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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.01/A1

Topic: A.02. Stem Cells and Reprogramming

Support: Academy of Finland grant
Business Finland grant
Instrumentarium Research Foundation grant
Aarne Koskelo Foundation grant
Paavo Nurmi Foundation grant

Title: Vascularized Immunocompetent Cortical Organoids for Disease Modeling and Drug Testing

Authors: *A. HALKOLUOTO¹, M. H. KEUTERS¹, T. ROLOVA¹, J. E. KOISTINAHO², H. DHUNGANA¹;

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Abstract: Neurodegenerative diseases, such as Alzheimer's disease, and cerebrovascular diseases, like an ischemic stroke, pose a substantial burden on societies. However, progress in translational research is limited by the lack of physiologically relevant human brain models. Three-dimensional human brain organoids mimic key aspects of brain physiology and laminar structures. However, even promising organoid models lack essential microenvironments, particularly vascular capillaries, and glial cells, which are crucial for mimicking in vivo conditions. This study addresses this gap by generating vascularized immunocompetent cortical organoids representing the human cortex with vascular and immune components. By co-culturing neural progenitor cells, vascular cells, and microglial progenitors we successfully created vascularized immunocompetent cortical organoids. Organoids were characterized using single-cell RNA sequencing and electron microscopy. Integration with host tissue was assessed following transplantation into mouse brain. At 35 days, organoids contained vascular cells, microglia, excitatory neurons, and glial progenitors, as confirmed by immunohistochemistry and single-cell RNA sequencing (Fig. 1A-B). Transmission electron microscopy revealed lumen in vascular-like capillaries. These organoids were implanted into the mouse brain, where they integrated with mouse tissue and created viable tissues capable of long-term survival. Organoid-derived vascular structures are in proximity to mouse vessels (Fig.1C). This platform can deepen our understanding of the pathophysiology of neurodegenerative diseases, potentially identifying novel therapeutic targets and treatment options for patients. Moreover, the model is equally suited for studying other neurological, neurodevelopmental, and psychiatric diseases.

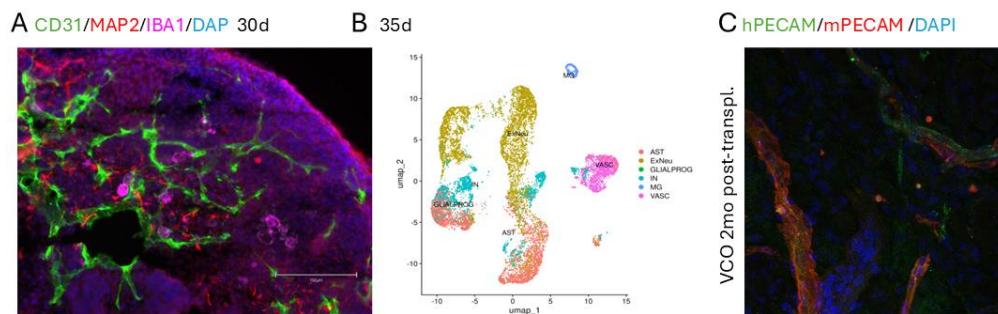


Fig .1 A. Human vascular immunocompetent cortical organoids (viCOs) stained with endothelial marker CD31 (green), microglial marker IBA1 (purple), neuronal marker MAP2 (red) and DAPI (blue). EVOSM500 20x. B. Single-cell RNA sequencing of viCOs annotated to Velmeshev fetal reference genome. C. Human vascular-like capillary (green, hPECAM) and mouse capillary (red, mPECAM) in close proximity. LSM90 40x.

Disclosures: **A. Halkoluoto:** A. Employment/Salary (full or part-time): Neuroscience Center, HiLIFE, University of Helsinki. **M.H. Keuters:** A. Employment/Salary (full or part-time): Neuroscience Center, HiLIFE, University of Helsinki. **T. Roloiva:** A. Employment/Salary (full or part-time): Neuroscience Center, HiLIFE, University of Helsinki. **J.E. Koistinaho:** A. Employment/Salary (full or part-time): HiLIFE, University of Helsinki, Division of Pharmacology and Pharmacotherapy, Univeristy of Helsinki. **H. Dhungana:** A. Employment/Salary (full or part-time): Neuroscience Center, HiLIFE, University of Helsinki.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.02/A2

Topic: A.02. Stem Cells and Reprogramming

Support: SynaptixBio

Title: Tubb4a suppression rescues h-abc cellular phenotype in human ipsc-derived oligodendrocytes and neurons

Authors: ***Z. HOU**^{1,2}, R. P. BATES², L. GARCIA², P. NAPIT², J. L. HACKER¹, A. D'ALESSANDRO², A. TAKANOHASHI², A. VANDERVER^{2,1}, S. SASE²;

¹Univ. of Pennsylvania, Philadelphia, PA; ²Children's Hosp. of Philadelphia, Philadelphia, PA

Abstract: Hypomyelination with Atrophy of Basal Ganglia and Cerebellum (H-ABC) is a rare leukodystrophy with heterozygous p.Asp249Asn (D249N) [P.745G>A] variant in the tubulin beta class IVA (*TUBB4A*). H-ABC presents in early childhood with developmental delay, speech

deficits, dystonia, dysphagia, progressive motor decline, and seizures. This leukodystrophy is defined by MRI with characteristic hypomyelination and atrophy in basal ganglia. The affected cell types include oligodendrocytes (OLs; myelin-producing cells), cerebellar granule neurons (CGNs), and medium spiny neurons (MSNs). However, the mechanisms through which *TUBB4A* mutations impact these cell types are unknown, and there is no cure for this debilitating disorder. Herein, we generated *TUBB4A*^{D249N} human induced pluripotent stem cells (hiPSCs) from H-ABC-affected individuals to model and understand the disease pathophysiology. We differentiated the *TUBB4A*^{D249N} hiPSCs into MSN- and OL-like cells. *TUBB4A*^{D249N} MSNs exhibited decreased maturation and increased apoptosis compared to wild-type control neurons. *TUBB4A*^{D249N} OLs fail to mature and show impaired complexity and myelination. Notably, CRISPR-mediated knockout (KO) of *TUBB4A* in *TUBB4A*^{D249N} (*TUBB4A*^{D249N}KO) hiPSC-derived neurons and OLs rescued cellular deficits without affecting wild-type *TUBB4A*^{KO} hiPSC neurons and OLs. This suggests that *TUBB4A* suppression rescues cellular phenotype and could be a viable treatment approach for H-ABC. To explore the therapeutic suppression of *TUBB4A*, we tested and screened antisense oligonucleotides (ASO) targeted against human *TUBB4A* in hiPSC-derived neurons and OLs. The selected ASO candidate showed no toxicity and significantly reduced the *TUBB4A* expression in wild-type and *TUBB4A*^{D249N} lines in both MSNs and OLs. ASO treatment in *TUBB4A*^{D249N} OLs partially improved OL maturation and complexity; however, it failed to improve the cellular phenotypes in MSNs. Ongoing studies are focused on treatment timing and developing a potent ASO candidate. Overall, this study provides proof-of-concept for ASO treatment and advances pre-clinical therapeutics for H-ABC disease.

Disclosures: **Z. Hou:** None. **R.P. Bates:** None. **L. Garcia:** None. **P. Napit:** None. **J.L. Hacker:** None. **A. D'Alessandro:** None. **A. Takanohashi:** None. **A. Vanderver:** 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, Passage BIO, NINDS, Biogen, NCATS, Children's Hospital of Philadelphia Center for Human Phenomic Science, NICHD, Ionis Pharmaceuticals., The children's hospital of Philadelphia cell & gene therapy acceleration grant, Sanofi Aventis U.S., Synaptix Biotherapeutics Ltd., Boehringer Ingelheim Pharmaceuticals, Eli Lilly and Co, Myrtelle, Sana Biotechnology, Takeda Pharmaceutical Company Limited, Shire Human Genetics Therapies. **S. Sase:** None.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.03/A3

Topic: A.02. Stem Cells and Reprogramming

Support: the National Natural Science Foundation of China Grants 82325015
the National Key Research and Development Program of China
2021YFA1101800

Title: Construction of human brain region specific organoids and assembloids reveal abnormal neurodevelopment in Down syndrome

Authors: *Y. LIU;
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Abstract: Down syndrome is a genetic disorder that results from an extra copy or part of chromosome 21 in the human genome. Individuals with Down syndrome have significant developmental delays, intellectual disabilities, and MRI results suggest that patients with Down syndrome have volumetric atrophy in multiple brain regions, including the cortex, cerebellum, and basal forebrain. The cognitive impairments in DS may result from early developmental defects in various brain regions, and the underlying molecular mechanism remains unknown. Here we differentiated human pluripotent stem cells into basal forebrain cholinergic organoids (hBFCOs) and cerebellar organoids by treating differentiation morphogens, respectively. Then we fused hBFCOs with cortical organoids (hCOs) to form assembloids and transplanted hBFCOs into immunodeficiency mice to construct chimeric brains. We validated the structural and functional connectivity of basal forebrain cholinergic neurons to the cortex in assembloids and chimeric brains. In addition, we identified neural morphological and projective defects in cholinergic neurons in Down syndrome patient iPSC-derived assembloids by histological assays. Cognitive impairment was further detected in chimeric mice that transplanted with Down syndrome patient iPSC-derived organoids. After performing single-cell/single-nucleus RNA-Seq analysis on organoids in the normal and Down syndrome groups, we found that axonogenesis was significantly affected in Down syndrome group, given the decreased projected fibers in the cortex of assembloids or chimeric brains; this may have been responsible for the cognitive impairment due to projection defects. Furthermore, suppression of the DSCAM pathway, which showed enhanced activity in DS, using CRISPR/Cas9 and RNA interference, reversed morphological defects in cholinergic neurons, thereby increasing the projection fiber in the cortex. Meanwhile, cerebellar organoids derived from individuals with Down syndrome recapitulated disease phenotypes, including decreased levels of cell proliferation and differentiation. More importantly, our data showed that Down syndrome cerebellar organoids have longer primary cilia, which may ultimately contribute to abnormal cerebellar development. Our work models the abnormality of basal forebrain-cortical cholinergic circuit and the developmental disorder of cerebellar cell proliferation in DS by using organoids and chimeric brains, which presents the direct influence of the DSCAM pathway and length of primary cilia on developmental brain defects in DS.

Disclosures: Y. Liu: None.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.04/A4

Topic: A.02. Stem Cells and Reprogramming

Support: NIH R01AG068992
NIH R41AG084421

Title: A scalable platform for consistent generation of functional microglia from induced pluripotent stem cells (iPSCs)

Authors: *A. DUONG¹, P. ZHOU¹, R. RAMIREZ², N. BUTELET¹, W. W. POON^{1,2};
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Abstract: Microglia are essential immune cells in the central nervous system (CNS), playing a critical role in brain health by surveying the environment, clearing debris, and regulating immune responses. Microglia dysfunction is implicated in numerous CNS disorders, driving research in disease modeling, therapeutic discovery, and cell therapy. While deriving microglia-like cells from induced pluripotent stem cells (iPSCs) holds great promise, scaling production for clinical and commercial use remains challenging due to inefficiencies in current methods. Existing approaches often rely on conventional 2D microplate systems, which are labor-intensive, inefficient, and unable to achieve high yields across multiple donor iPSC lines. To address these challenges, we developed a scalable platform for generating functional microglia from multiple iPSC donors and gene-edited lines. By optimizing differentiation conditions and implementing a rapid expansion system, we achieved high yield and purity, typical morphology, classical phagocytic function, and a transcriptomic profile that closely resembles both adult and fetal human microglia. These cells exhibited high post-thaw recovery and viability, secreted cytokines in response to inflammatory stimuli, and successfully could be co-cultured with other neural cells in both 2D and 3D systems. Our platform supports both scale-up and scale-out processes, enabling large-scale production of functional microglia for disease modeling, drug screening, and cell therapies. This approach has the potential to meet the high cell number demands required for CNS disease research and therapeutic applications.

Disclosures: A. Duong: A. Employment/Salary (full or part-time): NeuCyte, Inc. P. Zhou: A. Employment/Salary (full or part-time): NeuCyte, Inc.. R. Ramirez: None. N. Butelet: A. Employment/Salary (full or part-time): NeuCyte, Inc. W.W. Poon: A. Employment/Salary (full or part-time): NeuCyte, Inc..

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.05/A5

Topic: A.02. Stem Cells and Reprogramming

Support: NWO Gravitation Grant 024.004.012

Title: Ranbp1 haploinsufficiency contributes to impaired neuronal development in human 22q11.2 deletion syndrome

Authors: *S. ENUGALA^{1,2}, M. PEITZ³, O. BRUESTLE³, A. B. SMIT², R. E. VAN KESTEREN²;

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Abstract: 22q11.2 deletion syndrome is one of the most common micro-deletion syndromes in humans and is linked to disorders such as Di-George syndrome, attention deficit hyperactivity disorder (ADHD), schizophrenia (SCZ) and autism spectrum disorder (ASD). Neurons derived from human 22q11.2 deletion patient iPSCs could provide insights in neuron-specific cellular and molecular pathways underlying these disorders. Here, patient (n=3) and control (n=4) iPSC-derived NGN2-induced neurons were cultured on a rat astrocyte monolayer for 4, 8 or 11 weeks and subjected to molecular and cellular analyses. Proteomics analysis revealed a dysregulation of mRNA metabolism and transport with a key role for the 22q11.2 gene RANBP1. RANBP1 is known to control the Ran pathway which regulates nuclear protein and mRNA export and has been implicated in cell cycle progression, RNA processing and mitotic spindle assembly. Patient-derived neurons further showed premature network development as evidenced by increased dendrite length and synapse numbers at 4-8 weeks in culture. Next, we tested whether restoring RANBP1 expression in 22q11.2 patient-derived neurons is sufficient to rescue the molecular and cellular phenotypes of these neurons. Neurons were transduced with two different lentiviral SINEUP constructs to increase RANBP1 translation, cultured on a rat astrocyte monolayer for 4, 8 or 11 weeks and subjected again to proteomics and cellomics analyses. To observe neuronal morphology and synapse densities, neurons were stained with Hoechst, anti-microtubule-associated protein (MAP2), anti-RANBP1 and anti-synapsin1. One of the SINEUPs increased RANBP1 levels in transfected cells only. Importantly, physiological expression levels were reached that were only ~1.8 fold higher compared to non-transfected control cells, as opposed to the high expression levels that are often observed using conventional over-expression methods. Our findings identify RANBP1 as one of the potential genes in 22q11.2 locus that contributes to neuronal impairments underlying changes associated with 22q11.2 deletion syndrome and may help to further understand the cellular and molecular pathways involved in these neurological conditions.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.06/A6

Topic: A.02. Stem Cells and Reprogramming

Support: NIH R01 NS111986
Eagles Autism Foundation

Title: Elucidating the Role of Non-imprinted Genes in Dup15q Syndrome Neuronal Phenotypes

Authors: *D. ANJAN KUMAR, T. ROBINSON, E. S. LEVINE;
Univ. of Connecticut Sch. of Med., Farmington, CT

Abstract: Dup15q syndrome (Dup15q) is a rare neurodevelopmental disorder caused by duplication or triplication of maternal chromosome 15q11-q13 region. It is characterized by seizures, intellectual disability, and autism. Of the many duplicated genes in this region, *UBE3A*, a gene that encodes for a ubiquitin ligase, is solely expressed from the maternal allele and silenced by genomic imprinting on the paternal allele in neurons. While *UBE3A* has been hypothesized to be the main driver for Dup15q phenotypes, the role of other genes remains unknown. Because current mouse models only address overexpression of *UBE3A*, we have utilized patient specific neurons derived from induced pluripotent stem cell lines and isogenic CRISPR corrected control lines to formulate a better understanding of the role of non-imprinted genes in the pathology of Dup15q. Using the whole cell patch clamp technique to record electrophysiological parameters from 10-15 week old neurons, we have found that Dup15q neurons exhibit a hyperexcitability phenotype characterized by increased action potential firing frequency and decreased potassium current density compared to isogenic control neurons. Additionally, at these developmental timepoints, the percentage of cells receiving active GABAergic inputs is significantly higher in the Dup15q neurons compared to isogenic control neurons. Of the non-imprinted genes that are duplicated in this region, we focused on normalizing the expression of *GABRB3*, a gene that encodes for the β_3 subunit of the GABA_A receptor, because there is an increased percentage of Dup15q cells receiving GABAergic inputs, and gain of function mutations in this subunit have been implicated in epilepsy. To normalize the expression of this gene early in development, we utilized specific antisense oligonucleotides and assessed electrophysiological parameters in 10-15 week old neurons. We found that normalizing the expression of this gene prevented the development of increased GABAergic transmission phenotype but did not affect the increased action potential firing frequency and decreased potassium current density in Dup15q neurons. These data suggest that overexpression of *GABRB3* contributes to the synaptic phenotypes in Dup15q neurons. Understanding the role played by non-imprinted genes in addition to *UBE3A* will be crucial in not only identifying targets to develop more effective therapies, but to also develop improved mouse models for Dup15q.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.07/A7

Topic: A.02. Stem Cells and Reprogramming

Support: NIH R01 HD107489
CIRM Stem Cell and Gene Therapy program 2.0
The Egyptian Ministry of Higher Education

Title: Folic acid imbalance disrupts human cortical development and neural network function in cerebral organoids: implications for autism spectrum disorders

Authors: *S. M. ALI^{1,2,3,4}, V. HAGHANI⁵, R. KLEIN⁶, N. MCCULLOH², P. KNOEPFLER^{6,2}, J. LASALLE⁵, R. BEN-SHALOM⁷, R. GREEN^{6,2}, K. ZARBALIS^{6,2,7};

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Microbiology and Immunol., Univ. of California, Davis, CA; ⁶Pathology and Lab. Medicine, Sch. of Med., Univ. of California, Davis, Sacramento, CA; ⁷MIND Institute, Univ. of California, Davis, CA

Abstract: Background: Folate, essential for nucleotide synthesis and methylation, is critical for fetal brain development. Folic acid (FA), oxidized folate form, has been mandated in U.S. food fortification since 1998 besides the supplementation guidelines to prevent neural tube defects. Elevated maternal folate levels beyond WHO recommendations are linked to increased autism spectrum disorder (ASD) risk in children. Despite these concerns, the impact of excessive FA intake on human fetal brain development remains poorly understood. This knowledge gap is particularly pressing given the sharp rise in ASD prevalence in the U.S., along with evidence that disruptions in embryonic cortical development are common pathological features in ASD—underscoring the need to investigate how excess FA may alter early brain development.

Objectives: To examine the neurodevelopmental, neurophysiological, and transcriptional effects of folate imbalance during human cortical development using cerebral organoids, establishing a translational model to inform prenatal nutrition strategies. **Methods:** Human cerebral organoids were generated from two independent induced pluripotent stem cell lines and cultured for 40 days under three conditions: control, folic acid excess (FAE) and folinic acid excess (reduced folate). We employed immunohistochemistry, flow cytometry, WGBS and RNA sequencing to assess cellular and molecular changes. Neurophysiological activity was recorded in 100-day organoids using high-density microelectrode arrays (HD-MEAs). **Results:** FA excess disrupted cortical organization and neural progenitor dynamics via expanding the neural progenitor populations, reducing neurogenic output and altering the organoids' size. Folinic acid excess preserved normal progenitor dynamics and neurogenesis. HD-MEA recordings showed impaired network synchronization in the FAE conditions resembling ASD-related deficits, while excess folinic acid maintained normal function. RNA sequencing showed downregulation of genes linked to neuronal differentiation, axonogenesis, synaptic signaling, and ASD-associated genes in the FA excess conditions accompanied by altered DNA methylation profiles. **Conclusion and Significance:** Optimal folic acid levels are crucial for human cortical development and neural function. Folic acid excess disrupt neurogenesis and network synchronization, potentially contributing to ASD risk. These findings underscore the need for tailored prenatal nutrition strategies and highlight organoids as a robust platform for studying neurodevelopmental disorders

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

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Program #/Poster #: PSTR255.08/A8

Topic: A.02. Stem Cells and Reprogramming

Support: NIH Grant NS122902
RUCCTS pilot grant
Torsten N. Wiesel Fellowship

Title: Modeling Ataxia-Telangiectasia (A-T) in a 3D-Bioengineered Laminar Human Cerebellar Neural Circuit

Authors: *N. CHENG, J. ZHAO, E.-E. GOVEK, M. E. HATTEN;
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Abstract: The human cerebellum is most well-known for its role in motor coordination and increasingly appreciated for its non-motor roles in cognition, emotion, and language. The cerebellum has an archetypal cortical structure composed of two primary classes of cerebellar neurons, Purkinje cells (PCs) and granule cells (GCs), with other interneurons to modulate their activity. The current methodologies of modeling human cerebellum using self-organizing brain organoids suffer from a lack of repeatable, correctly formed neuronal layers. We have generated a new system for studying human cerebellar neuron development and physiology by modeling the human cerebellum in bio-engineered 3D laminar cerebellar neuronal circuits. It is achieved by seeding hPSC-derived cerebellar neurons into different “layers” in a specifically engineered polydimethylsiloxane (PDMS)-based microfluidic device. The PCs and GCs attach, survive, and extend processes through the special “microgrooves” structure to contact each other, thus forming basic cerebellar circuits in our new 3D laminar microfluidic system. Our system provides a unique platform for the study of human cerebellar developmental features and human cerebellum disease mechanism. This robust system is ideal for studying the CNS disease Ataxia-telangiectasia (A-T) caused by null mutations in the *ATM* gene. A-T results in cerebellar degeneration that prominently affects PCs and increase susceptibility to cancer and immune deficiency. *ATM* loss of function mouse models do not have an A-T cerebellar phenotype, which highlights the advantage of using our human cerebellum modeling system. Combining our human 3D laminar cerebellar neuronal system with iPSC differentiation technology, we have found a collection of developmental defects in *ATM* patient-derived cells. Our results to date show that fewer hPCs are generated from A-T iPSCs and that they make fewer synapses than control cells, suggesting that A-T might involve defects in developmental pathways, and that A-T iPSC-PCs have altered spiking patterning. Ongoing work includes using molecular biology

analysis to identify potential disease-causing pathways and using calcium imaging as well as electrophysiology to study changes in circuitry properties of A-T cerebellar neuronal circuits.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

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Topic: A.02. Stem Cells and Reprogramming

Support: NINDS F31 1F31NS141217
NIH T32 GM144233
Mayo Clinic Center for Individualized Medicine RFA
Pediatric DPD Deficiency Foundation

Title: Patient-derived cerebral organoids as a model for investigating the molecular mechanisms contributing to pediatric dihydropyrimidine dehydrogenase deficiency

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Abstract: Dihydropyrimidine dehydrogenase (DPD) deficiency is caused by inactivating genetic variations in the *DPYD* gene, resulting in a metabolic disorder that has been correlated with neurologic and developmental conditions that arise during childhood. Case studies have reported associations between DPD deficiency and neurological symptoms, including epileptic seizures, microcephaly, delayed motor skill development, and autism spectrum-associated behaviors. The goal of this study was to characterize the cellular and molecular changes associated with DPD deficiency using patient- and control-derived cerebral organoids differentiated from human induced pluripotent stem cells (iPSC). Patient iPSC lines were obtained from affected individuals with three different genotypes associated with full or partial DPD deficiency; control lines were from related, unaffected individuals and non-related individuals with no *DPYD* genetic variation. Differential gene expression analysis with DESeq2 was performed comparing embryoid bodies and mature organoids by patient phenotype. Overall, dysregulation of genes related to synaptic function, neurotransmitter signaling, cell-type markers, and neurological structural components was observed at all stages of development when comparing cases and controls. To further investigate dysregulation at a single cell level, single cell RNA sequencing was performed on mature organoids. A decrease in the proportion of neural stem cells and immature neurons was observed in cases when compared to DPD deficient controls. Three-dimensional immunofluorescence analyses were performed to mechanistically investigate observed cellular composition and gene expression changes. A decrease in GABAergic neurons and neuroepithelial cells was observed in cases when compared to controls. Based on our results, we

hypothesize that altered expression of genes involved in neurological development results from a reduction of β -alanine availability via DPD deficiency, leading to reduced modulation of glutamatergic signaling, which provides a potential explanation for the epileptic-type seizures that have been reported in patients. Further, delayed or abnormal GABAergic neuron development and the loss of GABAergic neurons within organoids derived from cases may suggest a mechanism contributing to an excitatory-inhibitory signaling imbalance within patients with pediatric DPD deficiency. Collectively, our data provide key insight into the etiology of this condition.

Disclosures: B. Bembenek: None. K. Bouchonville: None. R. Schmidt: None. S. Offer: None.

Poster

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Topic: A.02. Stem Cells and Reprogramming

Support: Unrestricted Grant from Research to Prevent Blindness (RPB)
Knights Templar Eye Foundation
California Institute for Regenerative Medicine (CIRM)

Title: Spatiotemporal Role of mGluR6 in Human Retinal Synapse Assembly Revealed by Inducible Knockout Organoids

Authors: *L. BAHMANI^{1,2,3}, P. GALVAN², J. BAI^{2,3}, S. PRAMEELA BHARATHAN^{2,3}, H. HOSSEINI⁴, A. NAGIEL^{2,3,5};

¹Children's Hosp. Los Angeles, Los Angeles, CA; ²The Vision Center, Dept. of Surgery, Children's Hosp. Los Angeles, Los Angeles, CA; ³The Saban Res. Institute, Children's Hosp. Los Angeles, Los Angeles, CA; ⁴Keck Sch. of Medicine, Univ. of Southern California, Los Angeles, CA; ⁵Roski Eye Institute, Keck Sch. of Medicine, Univ. of Southern California, Los Angeles, CA

Abstract: Background Metabotropic glutamate receptor 6 (mGluR6), encoded by the *GRM6* gene, is expressed in the retina where it plays a pivotal role in mediating synaptic transmission from photoreceptors to depolarizing bipolar cells (DBC). The precise mechanisms of synaptic assembly in human retina and the role of mGluR6 in this process remain poorly understood. This study will utilize *GRM6*-inducible knockout human retinal organoids (*GRM6* iKO HROs) to investigate the role of mGluR6 in different developmental stages of human retinal synaptogenesis. **Methods** To explore *GRM6* expression profile, we analyzed single-cell RNA sequencing datasets on mouse and human retina. Then, we performed RNA fluorescence *in situ* hybridization (RNA-FISH) and immunofluorescence staining (IF) to confirm different expression patterns of *GRM6* between mouse and human. Combining CRISPR/Cas9-mediated gene editing with the Cre/LoxP system, we developed tamoxifen-inducible *GRM6* knockout

(iKO) induced pluripotent stem cells (iPSCs). These iKO iPSCs were subsequently used to differentiate 3D HROs and treated with 2.5 μ M 4-hydroxytamoxifen (4-OHT) at specific ages. Localization and apposition of presynaptic and postsynaptic proteins were analyzed by confocal immunofluorescence microscopy in iKO HRO groups at day 220 (D220) and compared to the corresponding vehicle-treated control HROs. **Results** In contrast to mouse where mGluR6 is expressed only in DBCs, our RNA-FISH and IF experiments in human fetal retina and HROs revealed photoreceptor expression of mGluR6, in addition to its expression in DBCs. 4-OHT was applied to iKO HROs at D130 and D190 to model loss of mGluR6 during synaptic assembly and maintenance, respectively. At D220, the cellular organization of control and *GRM6* iKO HROs appeared similar. However, closer examination of HROs treated with 4-OHT at D130 revealed pre and postsynaptic abnormalities by D220, including a notable reduction in dystrophin and PSD-95 at photoreceptor terminals and TRPM1 and GPR179 at DBC dendritic tips. Mislocalization of presynaptic protein bassoon and retraction of cone photoreceptor pedicles were also observed in D130-Treated HROs resulting in the formation of a pseudo-outer plexiform layer (OPL). In contrast, synapse morphology was preserved in HROs treated with 4-OHT later at D190. **Conclusion** By utilizing iKO HROs with precise temporal control of mGluR6 knockout, our study may elucidate how mGluR6 interactions with other synaptic proteins orchestrate the synapse assembly process in the human retina. Our data in iKO HROs suggests that mGluR6 may be necessary for synaptic assembly but not maintenance.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.11/A11

Topic: A.02. Stem Cells and Reprogramming

Support: NIH-NIMH grant RM1MH133065

Title: Loss of function mutations in neurodevelopmental and psychiatric risk genes alter neuronal morphology and synaptic transmission in human neurons

Authors: *M. DÍAZ DE LEÓN GUERRERO^{1,2}, H. ZHANG⁵, P. GOWDA^{1,2}, D. J. SIRKIN⁵, M. ELACHWAH^{1,2}, G. TRACY⁵, A. MCCARROLL⁵, R. POLLAK³, R. P. HART⁴, A. SANDERS^{5,6}, A. KREIMER³, J. MULLE³, J. DUAN^{5,6}, Z. P. PANG^{1,2};

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Abstract: Neurodevelopmental and psychiatric disorders (NPDs), including autism, schizophrenia, bipolar disorder, and major depressive disorder, are complex conditions with high heritability and significant genetic contributions. Over the past decade, increases in genome-wide association studies (GWAS), whole-exome sequencing (WES), and study cohort sizes have enabled the detection of rare genetic variants associated with various NPDs. However, the effects of these mutations on neuronal function and synaptic transmission have not been assayed on a large scale. To address this gap, we developed a pipeline for clonal loss-of-function (LoF) mutagenesis in human induced pluripotent stem cells (hiPSCs) using cytosine base editing to introduce premature stop codons. Using this pipeline we have generated LoF mutants for >60 NPD risk genes across 4 different hiPSC lines, which were verified for pluripotency, genomic integrity, and gene LoF through transcript and protein-level analyses. These edited lines were differentiated into 2D cocultures of excitatory and inhibitory neurons to assess the impact of individual gene disruptions on neuronal morphology and function. We used high-content imaging to assay morphological differences in neuronal cultures and identified mutations that decrease neurite branching and synaptic puncta density. To evaluate synaptic transmission, we used the optical calcium sensor jRCaMP1f to measure spontaneous neuronal activity at single-cell and network levels. Our analysis revealed that specific LoF mutations result in altered calcium dynamics, suggesting changes in baseline synaptic activity and excitability. Additionally, using fluorescent sensors for neurotransmitter detection we've identified mutations that alter glutamate and GABA spontaneous release in our mutant lines. By combining these complementary assays, we aim to uncover shared functional signatures and identify common cellular pathways to deepen our understanding of the biology of NPD.

Disclosures: M. Díaz De León Guerrero: None. H. Zhang: None. P. Gowda: None. D.J. Sirkin: None. M. ElAchwah: None. G. Tracy: None. A. McCarroll: None. R. Pollak: None. R.P. Hart: None. A. Sanders: None. A. Kreimer: None. J. Mulle: None. J. Duan: None. Z.P. Pang: None.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.12/A12

Topic: A.02. Stem Cells and Reprogramming

Title: Improving Functional Activity of Human Pluripotent Stem Cell-derived Neural Organoids with BrainPhys™ Neuronal Medium

Authors: N. WILLIAMS¹, G. YOGARAJAH¹, J. BAGLEY², L. CHEW¹, A. MACCIONE³, S. A. LOUIS¹, A. EAVES^{1,4}, B. M. FONTINHA², M. GANDOLFO³, *C. K. MAK¹, E. KNOCK^{1,5};

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³Brain AG, Waedensville, Switzerland; ⁴Terry Fox Laboratory, BC Cancer, Vancouver, BC, Canada; ⁵Simon Fraser Univ., Burnaby, BC, Canada

Abstract: Organoids hold promise for replicating physiological neural network activity, including both spontaneous and coordinated firing patterns, due to their increased cellular diversity and structural complexity. However, currently available culture media may not adequately support functional neuronal activity, while media designed to promote activity (e.g., BrainPhys™ Neuronal Medium) may not support the full spectrum of neural and glial cell types. This trade-off limits the usability of organoids for functional studies and drug screening. To address this, we compared the effects of cell culture media on the long-term culture and electrophysiological activity of neural organoids generated using the STEMdiff™ Dorsal Forebrain Organoid Kit or STEMdiff™ Cerebral Organoid Kit. Fifty-day old organoids, cultured in Organoid Maturation Medium (OMM), were either switched to BrainPhys™ (supplemented with 10 mM glucose) or retained in OMM as a control. Cultures continued until day 150 - 200. To evaluate the effects of neuromodulatory drugs, we treated day-200 organoids with 100 µM 4-aminopyridine (4-AP) to induce an epileptiform phenotype, followed by treatment with 1 mM valproic acid. Functional activity was assayed using a high-density multielectrode array (HD-MEA) from 3Brain. These drug treatment recordings were performed in low-protein BrainPhys™ Medium. Day-150 dorsal forebrain organoids in BrainPhys™ showed a higher mean firing rate (MFR) and network burst frequency (NBF) compared to those in OMM (MFR: 1.3 ± 0.3 Hz vs. 0.3 ± 0.1 Hz; NBF: 14.6 ± 7.0 Hz vs. 0.6 ± 0.4 Hz, for BrainPhys™ and OMM, respectively [mean $\pm$ SEM; n = 3 - 9 organoids]). Day-150 cerebral organoids also displayed similar improvements in MFR and NBF when cultured in BrainPhys™ Medium compared to OMM (MFR; BrainPhys™: 0.2 ± 0.344 Hz vs. OMM: 0.03 ± 0.001 Hz and NBF; BrainPhys™: 4.5 ± 0.5 Hz vs. OMM: 0.5 ± 0.3 Hz; n = 3 organoids, mean $\pm$ SEM). Acute 4-AP treatment caused a 10- to 15-fold increase in the MFR of cerebral organoids, while valproic acid reduced the MFR by 3- to 5-fold relative to baseline (n = 3). These results demonstrate that neural organoids generated using STEMdiff™ kits exhibit robust neural activity when cultured in BrainPhys™. Introducing BrainPhys™ after terminal differentiation, when key neural and glial cell types were already established, highlights the importance of timing to preserve cell-type diversity. Additionally, glucose supplementation may be necessary to support metabolically demanding cell types and sustain high levels of functional activity.

Disclosures: **N. Williams:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc. **G. Yogarajah:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc. **J. Bagley:** A. Employment/Salary (full or part-time): a:head Bio AG. **L. Chew:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc., **A. Maccione:** A. Employment/Salary (full or part-time): 3Brain AG. **S.A. Louis:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc. **A. Eaves:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc., Terry Fox Laboratory, BC Cancer. **B.M. Fontinha:** A. Employment/Salary (full or part-time): a:head Bio AG. **M. gandolfo:** A. Employment/Salary (full or part-time): 3Brain AG. **C.K. Mak:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc. **E. Knock:** A. Employment/Salary (full or part-time): STEMCELL Technologies Inc., Simon Fraser University.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.13/A13

Topic: A.02. Stem Cells and Reprogramming

Title: In vitro neural and microglia co-culture systems to identify mechanisms underlying the therapeutic effects of an iPSC-based cell therapy for aging and Alzheimer's disease

Authors: ***L. DIMAS-HARMS**, E. VALENZUELA, M. ALCANTARA, S. BELL, V. MOSER, C. N. SVENDSEN;

Regenerative Med. Inst., Cedars-Sinai Med. Ctr., Los Angeles, CA

Abstract: With rates of Alzheimer's disease (AD) projected to double by 2060, understanding the underlying interactions to develop effective therapeutics is critical. Previous studies have shown cognitive improvements in aged and AD mouse models following treatment with young blood, plasma, or bone marrow; however, these approaches face challenges in scalability. Building on this work, we evaluated induced pluripotent stem cell (iPSC)-derived mononuclear phagocytes (iMPs) as a potential therapy for age- and disease-related cognitive decline. We find that bi-weekly, intravenous administration of iMPs over one month results in significant improvements in cognition in mouse models of both aging and AD. Immunohistochemical analyses demonstrate reductions in micro- and astro-gliosis and increased markers of neural health in treated animals. Single-nucleus RNA sequencing of hippocampus and proteomic profiling of plasma identifies several potential mechanisms through which iMPs may be beneficial. These include reductions in age-associated increases in plasma levels of serum amyloid proteins, and improved health of a key neural subtype. To further investigate potential mechanistic pathways, we employed in vitro approaches, using iPSC-derived cortical neuron and microglia co-cultures. Both, iMP-conditioned media and extracellular vesicles derived from iMPs, protected cultured cells against a variety of insults, including treatment with apoptotic agents and with the serum amyloid proteins we find are reduced in aging iMP-treated animals. Additionally, iMP-conditioned media was profiled in order to determine key proteins secreted by iMPs, which will now be evaluated for their therapeutic potential in vitro. By determining iMP protein secretion and the mechanistic interactions between iMPs and microglia/neurons in vitro, we aim to pinpoint the key cellular interactions and further our understanding of this promising cell-based therapeutic.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.14/A14

Topic: A.02. Stem Cells and Reprogramming

Support: California Institute for Regenerative Medicine (EDUC4-12751)
Winnick Family Foundation

Title: Generation of region-specific human iPSC-derived motor neurons (iMNs) to model regional vulnerability in ALS

Authors: *K. NING¹, I. TINDEL², B. SINGH², A. DAWSON², B. ZOGU², M. RAMOS², S. MUNAMARTY^{2,4}, S. ARFE², R. HO³;

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²Cedars-Sinai Med. Ctr., Los Angeles, CA; ⁴UCLA, Los Angeles, CA

Abstract: Amyotrophic Lateral Sclerosis (ALS) is a progressive neurodegenerative disease characterized by the degeneration of motor neurons (MNs), leading to paralysis and death. A defining feature of ALS is the selective vulnerability of MNs in different regions of the nervous system. Whether cervical, lumbar, or hindbrain MNs degenerate first determines the site of disease onset. Contemporary iPSC-derived models (iMNs) predominantly generate hindbrain and cervical MNs. Thus, they fail to capture the full spectrum of regional identities and potentially distinct pathogenic events, particularly those of lumbar MNs. Additionally, the heterogeneous nature of these models, which often produce mixed populations of MNs, can obscure critical region-specific characteristics and mechanisms of vulnerability, limiting their ability to fully model ALS pathology. To address these limitations, we optimized differentiation protocols by incorporating small molecules to direct the development of brachial, lumbar and hindbrain MN identities within 32 days. These protocols are reproducible across multiple healthy control and ALS cell lines, generating iMNs that closely resemble human spinal motor neurons with distinct segmental identities. Compared to traditional iMN induction methods, these optimized protocols produce iMNs with higher homogeneity and more well-defined regional identities. Notably, the protocols preserve key motor neuron and corresponding muscle innervation markers, which are critical for accurately modeling the vulnerabilities observed in ALS patients with different sites of onset. This enhanced consistency and regional specificity provide a robust platform for studying region-specific motor neuron properties and offer new opportunities to explore the diverse roles of spinal motor neurons in health and disease with greater precision.

Disclosures: **K. Ning:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **I. Tindel:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **B. Singh:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **A. Dawson:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **B. Zogu:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **M. Ramos:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **S. Munamarty:** None. **S. Arfe:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center. **R. Ho:** A. Employment/Salary (full or part-time): Cedars-Sinai Medical Center.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.15/A15

Topic: A.02. Stem Cells and Reprogramming

Support: BMBF Grant FKZ 01KI2311A/B

Title: Establishing a human stem cell-derived infection model to visualize the effects of *Listeria monocytogenes* on the developing brain

Authors: *A. FISCHER^{1,3,4}, C. LEMKE², L. HAIBER³, M. FULDE², B. SEEGER³, S. C. BRÖER^{1,4};

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Abstract: Prenatal zoonotic infections involving the central nervous system (CNS) are strongly associated with neurodevelopmental disorders in offspring. *Listeria monocytogenes* is a key example, with prenatal exposure linked to long-term neurological impairments. However, the mechanisms driving these outcomes remain poorly understood. To investigate underlying pathogenic pathways, we established a human induced pluripotent stem cell (iPSC)-derived neurosphere model that emulates critical aspects of early brain development, enabling systematic study of infection consequences. A standardized infection protocol was implemented: Bacteria were cultured to 10⁸ CFU/mL (OD 0.5) and applied at multiplicities of infection (MOI) of 50, 100, and 150 for 1-4 hours. Extracellular bacteria were removed via gentamicin, and neurospheres were fixed at 1-, 24-, and 48-hours post-infection (hpi). Infection was validated through immunofluorescent staining using *Listeria*-specific antibodies and Phalloidin to visualize the actin cytoskeleton. All experiments are performed in biological triplicate, each comprising a minimum of three technical replicates. Preliminary findings revealed heterogeneous invasion patterns across conditions. All tested MOIs successfully resulted in neurosphere infection; thus, MOI 50 was selected for subsequent experiments, as it more closely reflects physiologically relevant infection levels. Furthermore, infections demonstrated greater consistency after 2 hours of bacterial exposure compared to 1 hour, leading to the adoption of the 2-hour timepoint for future assays. Initial assessments of viability and cytotoxicity indicated that cell viability exceeded 80% relative to non-infected controls, while cytotoxicity remained below 5% relative to lysis controls, indicating that the model is appropriate for studying infection dynamics without significantly compromising cell survival. Ongoing studies will evaluate infected cell populations, assess proliferation and differentiation, and evaluate migration capacity and electrophysiological alterations post-infection. This model addresses a critical gap by enabling human-specific investigation of fetal CNS infections without the ethical barriers of *in vivo* studies. The observed invasion heterogeneity parallels clinical variability in congenital listeriosis outcomes, providing a platform for decoding the host-pathogen dynamics that drive neurodevelopmental sequelae.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.16/A16

Topic: A.02. Stem Cells and Reprogramming

Support: P30AG072976
U54AG090792

Title: Investigating polygenic contributions to Alzheimer's Disease pathology using a human NGN2-based iPSC-derived neuronal model

Authors: ***R. Afify**^{1,2}, **L. VARGHESE**^{1,2}, **M. HERNANDEZ**¹, **J. S. MEYER**^{1,2};
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Abstract: Alzheimer's disease (AD) is a polygenic neurodegenerative disorder in which the combined impact of genomic variants contributes to disease susceptibility and progression. Although genome-wide association studies (GWAS) have significantly advanced our understanding of the genetic architecture underlying AD, the functional impact of polygenic risk on human neurons remains largely unclear. This highlights the need for physiologically relevant human neuronal models that capture the genetic complexity of AD pathology. To more properly elucidate polygenic contributions to AD pathology, we generated multiple lines of human induced pluripotent stem cells (iPSCs) from patient samples from the ADNI study based upon varied polygenic risk scores (PRS), including patients from the uppermost and lowermost quartiles. With these iPSCs, we then aimed to optimize an NGN2-induced neuronal differentiation protocol to enable robust, efficient, and reproducible generation of excitatory cortical neurons from iPSCs derived from AD patients with variable PRS. iPSC lines were successfully differentiated into neurons across all groups, with neuronal identity and morphology validated using immunocytochemistry (ICC), confirming the robustness of the protocol across genetically diverse backgrounds. Ongoing experiments aim to functionally and molecularly characterize these neurons to uncover genotype-associated differences. Neuronal activity is being assessed using multi-electrode array (MEA) technology, while Western blotting, ELISA, and immunocytochemistry (ICC) are used to quantify AD-relevant protein burden, including amyloid-beta (A β) and phosphorylated tau. We anticipate that neurons, particularly those derived from individuals with high PRS, will exhibit altered electrophysiological properties, disrupted synaptic integrity, and dysregulated expression of AD-related proteins compared to neurons from low-PRS or healthy control individuals. Ultimately, this study utilizing NGN2-based iPSC-derived neuronal platform offers a promising tool to unravel how polygenic risk shapes AD pathophysiology. It holds potential to uncover polygenic contributions to disease pathology, support genotype-guided therapeutic development, and advance personalized risk prediction in AD.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.17/A17

Topic: A.02. Stem Cells and Reprogramming

Support: NIH grants NS127391
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the Parkinson's Cell Therapy Research Fund at McLean Hospital and
Massachusetts General Hospital
the Masson Family Endowed Scholar in Neurosurgery
the Korea-US Collaborative Research Project (RS-2024-00468036)

Title: Metabolic Acceleration of Dopaminergic Neuron Differentiation from Human Pluripotent Stem Cells for Cell Replacement Therapy in Parkinson's Disease

Authors: *H. JANG¹, Y. CHA¹, Y. HONG¹, Y. PARK¹, H. SEO², K. S. KIM¹;
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Abstract: Parkinson's disease (PD) is a neurodegenerative disorder characterized by the progressive loss of midbrain dopaminergic neurons (mDANs) in substantia nigra, making it a prime target for cell replacement therapy (CRT). Recent studies from our laboratory identified needle trauma as a major contributor to the early, selective death of grafted mDANs, primarily driven by injection-induced innate immune responses. We also demonstrated that intro-striatal co-transplantation of autologous regulatory T (T_{reg}) cells significantly enhances mDAN survival by suppressing these immune responses. These findings support the therapeutic use of highly enriched mDANs rather than mDA progenitors (mDAPs), which most current protocols prioritize. To address this challenge, we optimized in vitro differentiation by integrating metabolic and developmental signals. While human pluripotent stem cells (hPSCs) predominantly rely on glycolysis, their transition toward oxidative phosphorylation (OXPHOS) is critical to meet the bioenergetic and biosynthetic demands during neuronal differentiation. Leveraging this metabolic shift, we incorporated selected metabolic regulators into our established spotting-based differentiation protocol, significantly enhancing mDAN yield and maturation. We further found that small molecules targeting key transcription factors synergistically promoted dopaminergic differentiation and suppressed off-target cell fates. Importantly, combining this optimized protocol with T_{reg} cell co-transplantation led to improved functional recovery in rodent models of PD. This integrated strategy enhanced graft survival, minimized unwanted differentiation, and consistently yielded high proportions of mDANs. In summary, our approach improves both the safety and efficacy of mDAN-based CRT, offering a refined method with strong translational potential for clinical application in PD.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.18/A18

Topic: A.02. Stem Cells and Reprogramming

Support: Gilbert Family Foundation Grant #923016

Title: A Human iPSC-Derived Model to Assess Microglial Dysfunction in Neurofibromatosis Type 1.

Authors: ***M. GOMEZ**¹, **R. AFIFY**², **S. PATIL**³, **M. HERNANDEZ**², **J. S. MEYER**⁴;
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²Indiana Univ. Sch. of Med., Indianapolis, IN; ⁴Stark Neurosciences Res. Inst., Indiana Univ. Sch. of Med., INDIANAPOLIS, IN

Abstract: Neurofibromatosis type 1 (NF1) is a common neurogenetic disorder caused by mutations in the NF1 gene, leading to tumor formation and neurodevelopmental abnormalities. Microglia, the resident immune cells of the central nervous system, have emerged as key contributors to glioma development and disease progression in NF1. While animal studies have provided valuable insights into microglia-driven mechanisms, translating these findings into human models remains a major challenge. To overcome this, we used CRISPR/Cas9 gene editing in human induced pluripotent stem cells (iPSCs) to generate microglia-like cells (hMGLs). Using isogenic iPSC lines, we created control, heterozygous, and homozygous NF1 Arg816X mutant hMGLs to investigate the functional consequences of NF1 loss. iPSC-derived hMGLs expressed key microglial markers, including IBA-1 and TREM2, confirming their identity and successful differentiation. Further functional assays revealed that homozygous NF1 Arg816X hMGLs exhibited increased sensitivity to cytokine starvation, suggesting a heightened susceptibility to stress-induced cell death. Additionally, preliminary evidence of altered phagocytic activity was observed in NF1-mutant hMGLs compared to isogenic controls. These findings suggest that NF1 mutations may disrupt fundamental aspects of microglial biology, potentially contributing to disease pathology. This model offers a promising human-based platform for dissecting the cellular mechanisms underlying NF1 and highlights the critical need for further studies into microglial dysfunction in this disorder.

Disclosures: **M. Gomez:** None. **R. Afify:** None. **S. Patil:** None. **M. Hernandez:** None. **J.S. Meyer:** None.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.19/A19

Topic: A.02. Stem Cells and Reprogramming

Support: Research without animal tests (Forska Utan Djurförsök) Project-ID: N2023-0007

Title: Commercial Culture Media Trigger Acute Epileptiform Activity in Human iPSC-Derived Neurons: The role of calcium and magnesium.

Authors: *S. THEISS¹, E. ARTHURSSON², S. ILLES³;

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²Univ. of Gothenburg, Göteborg, Sweden; ³Inst. of Neurosci. and Physiol., Oscillation AB, Gothenburg, Sweden

Abstract: We demonstrate that commonly used cell culture media—including Neurobasal™ (+/- A), Neurobasal Plus™, and BrainPhys™—do not accurately replicate the ionic composition of human cerebrospinal fluid (CSF)¹. Using human induced pluripotent stem cell (PSC)-derived neuronal networks cultured on microelectrode arrays (MEAs), we show that the abnormally high extracellular potassium levels present in all tested media induce acute epileptiform activity, comparable to that triggered by the convulsant 4-aminopyridine¹. Building on these effects of elevated extracellular potassium, we also systematically examined how other ion concentrations, e.g. magnesium and calcium, affect human neuronal network activity. These ions are of particular interest, because all commercial media also fail to accurately reflect magnesium levels found in human CSF. To address this, we used a high-capacity assay platform using 3D PSC-derived neural cultures integrated with microelectrode arrays (MEAs) in a 96-well format. This system enables simultaneous recording of spiking activity and LFPs, and thereby combines the electrophysiological depth of brain slice models with the scalability of cell culture systems, providing a human-relevant alternative to animal-based assays. Using this model, we reveal that even subtle deviations in extracellular calcium and magnesium concentrations were sufficient to change human neuronal network activity. Our findings emphasize that precise control of extracellular ion composition is essential for maintaining normal human neural network function and avoiding artifactual hyperexcitability in vitro. Altogether, these results underscore that small changes in extracellular ion concentrations have large effects on human neuronal network activity. This work lays the foundation for enhancing the fidelity of human iPSC-based brain models in mimicking both normal and disease-relevant network functions. *References:* 1. Lyckenvik T, Izsak J, Arthursson E, Forsberg M, Johansson K, Zetterberg H, Axelsson M, Wasling P, Theiss S, Hanse E, et al. Non-Physiological Potassium Concentrations in Commercial Culture Media Trigger Acute Epileptiform Activity in Human iPSC-Derived Neurons. bioRxiv. 2025.

Disclosures: S. Theiss: None. E. Arthursson: None. S. Illes: A. Employment/Salary (full or part-time): Oscillation AB. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Oscillation AB.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.20/A20

Topic: A.02. Stem Cells and Reprogramming

Title: Dissecting in vitro mechanisms of GABAergic inhibition in a co-culture system using human pluripotent stem cell-derived pallial interneurons

Authors: ***J. SALVATIERRA**¹, **G. SUBRAMANYAM**¹, **Y. MAURY**³, **M. BERSHTEYN**², **S. HAVLICEK**¹, **C. NICHOLAS**¹;

¹Neurona Therapeut., South San Francisco, CA; ²Discovery Res., Neurona Therapeut., Redwood City, CA; ³Discovery research, Neurona therapeutics, South San Francisco, CA

Abstract: In approximately one third of patients with epilepsy, seizures do not respond adequately to anti-seizure medications. For these individuals, alternative treatments to surgically resect or ablate affected brain tissue are tissue-destructive and may cause irreversible neurocognitive impairment. GABAergic interneurons play a crucial role in regulating brain activity, preventing the excessive synchronization that can lead to seizure onset and propagation. We have developed a cell therapy product, NRTX-1001, composed of human pluripotent stem cell (hPSC)-derived post-mitotic MGE pallial-type GABAergic interneurons (hpINs), designed to restore balanced neural activity in drug-resistant focal epilepsy, which is being investigated in ongoing clinical trials (NCT05135091, NCT06422923). To elucidate mechanisms by which GABAergic interneurons suppress neuronal network hyperexcitability, we have developed an in vitro model using multi-electrode array (MEA) recordings of hpINs co-cultured with hPSC-derived glutamatergic neurons. This platform allows real-time assessment of network activity, dynamics, and kinetics using a variety of parameters, to isolate the role of GABAergic hpINs over time. We employed optogenetic tools and pharmacological manipulations to functionally dissect GABAergic inhibition in this system. We used excitatory opsins (e.g. ChR2) to selectively activate hpINs and to evaluate their potential to suppress synchronized network firing. Conversely, inhibitory opsins (e.g., ArchT) were used to transiently silence hpINs, and reveal their requirement for dampening intrinsic network hyperactivity. Additionally, perturbation of GABA signaling using toxins and receptor antagonists enabled specific disruption of GABA release and/or its binding to receptors, which further helped to delineate the role of synaptic inhibition in seizure-like dynamics. Our results provide mechanistic insight into how GABAergic interneurons regulate excitatory network activity in vitro and lay the foundation for evaluating therapeutic strategies that target inhibitory circuitry in epilepsy and related disorders.

Disclosures: **J. Salvatierra:** A. Employment/Salary (full or part-time): Neurona Therapeutics. **G. Subramanyam:** A. Employment/Salary (full or part-time): Neurona Therapeutics. **Y. Maury:** A. Employment/Salary (full or part-time): Neurona Therapeutics. **M. Bershteyn:** A. Employment/Salary (full or part-time): Neurona Therapeutics. **S. Havlicek:** A. Employment/Salary (full or part-time): Neurona Therapeutics. **C. Nicholas:** A. Employment/Salary (full or part-time): Neurona Therapeutics.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.21/A21

Topic: A.02. Stem Cells and Reprogramming

Support: Loulou Foundation

Title: Cortico-thalamic assembloids as a platform model CDKL5 Deficiency Disorder and to test Gene Therapy strategies

Authors: *R. BLANCH¹, M. TENREIRO¹, A. M. BOVE⁵, C. ESCALANTE VERA², T. ZHOU³, A. E. SAJCHE SAPON⁴, F. PUPPO¹, A. R. MUOTRI¹;

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Abstract: Mutations in the X-linked cyclin-dependent kinase-like 5 (CDKL5) gene cause CDKL5 Deficiency Disorder (CDD), a rare, severe neurodevelopmental condition marked by early-onset intractable epilepsy, developmental delay, motor dysfunction, and autistic features. Currently, there are no effective treatments for CDD, and existing animal models fail to fully capture critical disease features, particularly spontaneous seizures. Thus, advanced human-based models are urgently needed to uncover CDD's molecular basis and accelerate therapeutic discovery. We previously generated and characterized a panel of induced pluripotent stem cells (iPSCs) from CDD patients harboring distinct CDKL5 loss-of-function mutations, along with isogenic controls. Differentiation into 3D cortical organoids (COs) revealed molecular, cellular, and functional abnormalities, including early hyperexcitability and impaired network maturation, which align with clinical observations. Recognizing the role of the cortico-thalamic (CTh) circuit in epilepsy, we hypothesized that modeling reciprocal axonal connectivity between cortex and thalamus using patient-derived cells would better reproduce the spontaneous seizures observed in CDD. To this end, we generated 3D CTh assembloids by fusing COs and thalamic organoids (ThOs), and functionally characterized them in the CDD context. Multi-electrode array (MEA) recordings and pharmacological assays in control assembloids showed enhanced network maturation and a more balanced excitation/inhibition profile compared to individual organoids. To address the absence of disease-modifying therapies, we tested two AAV-based gene therapy strategies. First, COs and ThOs were transduced with Prime Editors to correct CDKL5 mutations, and editing efficiency was quantified. Second, we applied DNA methylation editors to reactivate the functional CDKL5 allele on the inactive X chromosome. Treated COs and ThOs were assembled into CTh assembloids and subjected to MEA to assess electrophysiological rescue. This human CTh assembloid model provides a powerful platform for modeling network-level dysfunction and offers a scalable, physiologically relevant system for testing therapies for CDD and related neurodevelopmental and epileptic disorders.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.22/A22

Topic: A.02. Stem Cells and Reprogramming

Support: Cellsight Development Fund
Reserch to Prevent Blindness
NIH (SPB)

Title: Human retinal organoid model of photoreceptor cell death identifies H105A peptide as a potential treatment for dry age-related macular degeneration

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Abstract: In dry age-related macular degeneration (AMD), photoreceptor degeneration leads to irreversible vision loss. Oxidative stress, mitochondrial dysfunction, and lipid peroxidation are major contributors to this process. Current therapies can slow the progression of retinal pigment epithelium (RPE) lesions, but they provide limited protection for photoreceptors. To address this therapeutic gap, there is an urgent need for human disease models that accurately replicate AMD pathology. Therefore, we developed a retinal organoid (RO) model using human induced pluripotent stem cells (hiPSCs) that replicates key aspects of photoreceptor degeneration associated with AMD, and used this platform to evaluate the neuroprotective potential of H105A, a therapeutic peptide derived from pigment epithelium-derived factor (PEDF). Specifically, two distinct hiPSC lines were utilized to generate ROs, which were cultured for 180 days before being exposed to either vehicle control or cigarette smoke extract (500 or 750 µg/ml) for 24 or 48 hours. Photoreceptor degeneration and cell death were assessed using immunofluorescence, TUNEL staining, and confocal imaging, and quantified using Fiji software. Additional readouts included live assessment of oxidative damage, lipid peroxidation, intracellular redox status, iron accumulation, and cell death pathways using 3D automated reporter quantification, and supported by proteomic analysis. Our results showed that CSE exposure led to elevated levels of reactive oxygen species, increased lipid peroxidation, disrupted redox homeostasis (reduced GSH/GSSG ratio), and excessive intracellular iron. These changes caused photoreceptor-specific cell death through both apoptotic and ferroptotic mechanisms in a dose- and time-dependent manner. Notably, co-treatment with the H105A peptide significantly reduced photoreceptor cell death under AMD-relevant stress conditions. Moreover, H105A-

treated organoids exhibited improved photoreceptor survival, as indicated by reductions in markers of apoptosis. These findings emphasize the significance of this novel human-derived model for therapeutic evaluation and spotlight H105A as a strong candidate for mitigating vision loss in patients with dry AMD.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

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Program #/Poster #: PSTR255.23/A23

Topic: A.02. Stem Cells and Reprogramming

Support: CMT4B3 Foundation

Title: Defining cell type-specific pathologies of CMT4B3 in a novel iPSC-based model system

Authors: *X. MAO¹, M. SHARMA², J. CHUA³;

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Abstract: Background: Charcot-Marie-Tooth Type 4B3 (CMT4B3) is a genetic neurological disorder caused by recessive mutations in the gene *Set-Binding Factor 1 (SBF1)*, which encodes the protein myotubularin-related phosphatase 5 (MTMR5), a negative regulator of autophagy. MTMR5 LoF in CMT4B3 patients results in peripheral axon loss, aberrant myelination, muscle atrophy, impaired mobility, and neuropathic pain. In severe cases, patients exhibit multiple cranial neuropathies, microcephaly, and intellectual disability. **Objectives:** This study aimed to generate patient-derived iPSC lineages of CMT4B3 capable of differentiating into principal cell types of the PNS: lower motor neurons, sensory neurons, and skeletal muscle cells. This new cell model enables (1) mechanistic investigations of autophagy dysfunction, MTMR5 biology, and cell degeneration on a cell type-specific basis and (2) testing of candidate molecular strategies of high therapeutic potential. **Methods:** Cassettes for doxycycline-inducible transcription factors (*NGN2/ISL1/LHX3* for iMotor Neurons (iMN), *NGN2/BRN3A* for iSensory Neurons (iSN), *MYOD/OCT4* shRNA for iMuscle (iM) cells) were stably integrated into the genome of CMT4B3 iPSCs with pathogenic compound heterozygous *SBF1* mutations via a transposon system. Cells were differentiated via doxycycline addition (7-14 days) and harvested for Western blot, immunocytochemistry, RT-PCR, or propidium iodide analyses. **Results:** WT and CMT4B3 iPSCs differentiated into iMNs, iSNs, and iMs with >99% efficiency based on immunocytochemistry for cell identity/maturity markers. Mutant iMNs showed extensive cell death within 2-6 days of differentiation; mutant iSNs and iMs showed minimal cell death. Mutant undifferentiated iPSCs demonstrated decreased autophagy substrate proteins LC3-II and p62 compared to WT, suggestive of increased autophagic degradation. Healthy iPSCs, iMN, and

iSN but not iM with *SBF1* KD showed decreased autophagy substrate levels. These substrates were decreased and not significantly changed in mutant iSN and iM cells. Addition of VPS34-IN1 successfully downregulated autophagy in *SBF1* KD CMT4B3 iMN and iSN. **Conclusions:** This study establishes the first human PNS cellular model of CMT4B3, facilitating mechanistic analyses of autophagy and MTMR5 dysfunctions in different cell types. *SBF1*/MTMR5 mutations lead to most autophagy disinhibition in neurons, hinting at a possible neuron-specific vulnerability in CMT4B3. This provides critical insights into CMT4B3 pathogenesis and lays a foundation for targeted therapeutic strategies.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.24/A24

Topic: A.02. Stem Cells and Reprogramming

Support: URF/1/5056

Title: Supernumerary X chromosomes shape brain organoid architecture and functions in a dose-dependent fashion

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Abstract: Klinefelter syndrome (KS, 47,XXY) is the most prevalent aneuploidy in males (1:400-1:600). High-grade sex chromosome aneuploidies (HGA-SCAs), such as 48,XXXY, and 49,XXXXY are rarer conditions occurring in 1:40.000-1:80.000 males. KS and HGA-SCA patients exhibit a broad spectrum of neuronal impairment, including cognitive deficits, seizures, autistic traits, and motor, speech, and language delays. While KS patients typically display a milder phenotype, HGA-SCAs are associated with profound cognitive defects. Despite the prevalence of X chromosome aneuploidies, there is a critical need for cellular models to define the transcriptional, epigenetic, and functional consequences of X chromosome overdosage during neurodevelopment. To this end, we derived cortical organoids from 47,XXY, 48,XXXY, and 49,XXXXY iPSCs. Allele-specific expression (ASE) analysis on X aneuploid organoids demonstrated a preserved epigenetic X inactivation status at different time points, from one to 12 months of differentiation in vitro. Through a multi-layered analysis integrating morphological, functional, bulk, and single-cell transcriptomics, we found that the additional X chromosomes lead to impaired neural patterning, disrupted cortical architecture, and altered electrophysiological properties of cortical organoids in a dose-dependent manner. While 47,XXY

organoids are phenotypically and functionally similar to 46,XY controls, HGA-SCAs display severe functional defects and aberrant transcriptomes. Through single-cell RNA analysis, we profiled the genes that escape X inactivation in neuronal and non-neuronal cell populations and revealed a dysregulated proliferation of neural progenitors in organoids carrying supernumerary X chromosomes. Additionally, severe astrocyte differentiation defects were observed in HGA-SCAs organoids, potentially contributing to synaptic dysfunction. Moreover, high-density microelectrode arrays (MEA) analysis revealed a higher mean spike firing rate and amplitude of HGA-SCAs compared to 46,XY organoids. Finally, patch-clamp studies demonstrated significant hyperexcitability of HGA-SCA organoids and X dosage-sensitive deficits in long-term potentiation (LTP). Our work leveraged the inaugural cohort of X aneuploid cortical organoids to unravel the functional consequences of X-linked gene overdosage during neurodevelopment

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Poster

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.25/A25

Topic: A.02. Stem Cells and Reprogramming

Title: Automated culture and development of iPSC derived 3D neural organoids followed by morphological characterization and functional analysis of calcium oscillation activity

Authors: *O. SIRENKO¹, P. MACHA², L. CHEW³;

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Abstract: Developing 3D brain organoids from human induced pluripotent stem cells (iPSC) is a rapidly developing technology with great potential for understanding brain development, neuronal diseases, and the impact of different genetic backgrounds. However, the formation of neural organoids is a complex and lengthy process, making it less suitable for compound screening. Here we demonstrate an automated protocol for the development of neural organoids using the CellXpress.ai Automated Cell Culture System. The system includes an automated liquid handler, imbedded imager, automated incubator, and process-controlling scheduling software. The system enables automation of most cell culture repetitive tasks, including 3D organoid culture with periodic media exchanges, agitation, and organoid development monitoring by imaging. The protocol includes stages of initial organoid formation using Accura plates or low attachment U-bottom plates, then continuous culture of organoids in 24-well or 6-well plates with periodic imaging and media exchange. Brain organoids were developed from iPSC cells using the dorsal forebrain development protocol and media from STEMCELL Technologies. Automation with the CellXpress.ai system was applied for most of the continuous

culture process, except the initial organoid formation step.

After development and maturation of organoids during 60+ days of culture, functional characterization of spontaneous neural activity of 3D organoids was measured by recording and analyzing calcium oscillations. Calcium oscillations were recorded on the FLIPR instrument that measured fast kinetic changes in calcium signal. Oscillation patterns were analyzed for multiple parameters including peak count, amplitude, and peak width. Organoids demonstrated spontaneous calcium oscillation activity. Morphological characterization of 3D organoids was done by imaging. Organoids diameters ranged from 1800-2000 μm , and the expression of neural markers including TUJ1 and GFP was detected with fluorescently labeled antibodies. For pharmacological characterization, several compounds were used to show appropriate functional responses. AMPA and 4 AP addition resulted in dose-dependent increase of frequency of calcium oscillations, while GABA and caused decrease in oscillation frequencies. Taken together, this biological system of iPSC derived 3D neural organoids paired with process automation and detailed analysis of calcium oscillations demonstrates a promising tool for compound testing.

Disclosures: **O. Sirenko:** A. Employment/Salary (full or part-time): Employment full time, Molecular Devices. **P. Macha:** A. Employment/Salary (full or part-time): Employment full time, Molecular Devices. **L. Chew:** A. Employment/Salary (full or part-time): Employment full time, STEMCELL Technologies.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.26/A26

Topic: A.02. Stem Cells and Reprogramming

Title: Mapping human brain development through combined tissue engineering and synthetic biology approaches

Authors: ***J.-P. URENDA**¹, G. QUADRATO²;
¹USC, Los Angeles, CA; ²USC, Los Angeles, CA.

Abstract: The human cerebral cortex has an unparalleled diversity of cell types, generated during development through a series of spatiotemporally orchestrated events critical for establishing cortical circuit functionality. Human induced pluripotent stem cell (hiPSC)-derived brain organoids provide the potential to address foundational questions in early human cortical development, such as which cell types arise from which progenitors at what times, and how those dynamics change in response to mutations or perturbations associated with neurodevelopmental disorders. However, brain organoids lack stereotypic macroscale anatomic structure, such as reproducible long-range connectivity, and have limited activity-dependent cellular maturation, making them ineffective for modeling later stages of human brain development and disease. To address these limitations, we have established a multi-organoid-on-

chip platform, combining tissue engineering technologies and new unbiased organoid analysis readouts, to recapitulate *in vivo* functional and dysfunctional connectivity of the human brain. Interestingly, organoids cultured under continual microfluidic perfusion for up to 3 months, maintain progenitor and neuronal cell viability at similar levels as bioreactor control organoids. Using scRNA-seq we show that progenitors are biased towards a neuronal differentiation fate and neurons increase expression of neuronal maturation and identity genes relative to bioreactor controls. Corroborating these findings, we see an increase in mature neuronal morphologies and an increase in both spontaneous network activity and synchronized activity in these microfluidic cultured organoids. We are further characterizing these neurons via whole-cell patch-clamping. Using our platform, we have induced long-range connectivity via hydrogel coated microchannels between cortical organoids and seen an improvement in spontaneous activity compared to static cultures. We are now developing functionalized microporous annealed particle gels to induce neurite extensions from specific subtypes of neurons to better control the connectivity between brain organoids. Our modular platform offers a novel framework that maintains organoid viability while simultaneously allowing the fine-tuning of neuronal connections made between brain-region specific patterned organoids to better model the co-development of distinct brain structures.

Disclosures: J. Urenda: None.

Poster

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Topic: A.02. Stem Cells and Reprogramming

Support: the Novo Nordisk Foundation (NNF18OC0030286, NNF17CC0027852 and NNF21CC0073729)
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Innovation Fund Denmark (BrainStem: 4108-00008A),
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Knut and Alice Wallenberg Foundation

Title: Mapping early patterning events in human neural development using an in-vitro microfluidic stem cell model

Authors: *G. RATHORE^{1,2}, A. KIRKEBY²;

¹Dept. of Neurol., Univ. of California San Francisco, San Francisco, CA; ²Novo Nordisk Fndn. Ctr. for Stem Cell Med. (reNEW), Univ. of Copenhagen, Copenhagen, Denmark

Abstract: *Stem cell models can provide insights into human brain development at embryonic stages which are normally inaccessible. We previously developed the Microfluidic Stem Cell Regionalisation (MiSTR) model, which recapitulates the rostro-caudal patterning of human*

neural tube through a WNT activation (WNTa) gradient. Through temporal single cell transcriptomics of rostro-caudal and dorso-ventral gradient-patterned MiSTR, we found that rostro-caudal subtypes were regionally specified and fate-determined already during the late epiblast stage, several days before onset of neuralisation at day 3-4. Rostral cells were characterised by expression of HESX1 and SHISA2 during pre-neuralisation and PAX6 during early neuralisation, whereas caudal cells expressed FST and HOXA1 during pre-neuralisation and SOX1 as the dominant neuralising factor. In contrast to the early rostro-caudal specification, response to ventralisation in telencephalic progenitors was developmentally delayed and occurred around day 9. We further uncovered temporal events in human midbrain-hindbrain boundary formation and ventral forebrain patterning, contributing new knowledge on early human neural region-specification.

Disclosures: **G. Rathore:** A. Employment/Salary (full or part-time): UCSF. **A. Kirkeby:** None.

Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

Location: SDCC Halls B-H

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Program #/Poster #: PSTR255.28/A28

Topic: A.02. Stem Cells and Reprogramming

Support: UCR startup fund
TRANSCEND fellowship from the California Institute of Regenerative Medicine(Award # EDUC4-12752).

Title: Bicontinuous interconnected porous scaffolds provide topographical cues that govern neuronal behavior and maturation

Authors: ***P. OKORO**, K. DALSANIA, S. IRAGAVARAPU, M. BASARANBILEK, B. ANVARI, I. NOSHADI;
Univ. of California, Riverside, Riverside, CA

Abstract: 3D tissue-engineered models hold great promise for recreating the intricate architecture and dynamic functions of neural tissues. However, replicating the nuanced structural cues of the brain *in vitro* remains challenging, as existing platforms often fail to capture the precise motifs that guide neural stem cell behavior and maturation. Here, we introduce a new architectural pathway toward physiologically relevant neural tissue models, integrating solvent transfer-induced phase separation (STriPS), microfluidics, and bioprinting to develop a bicontinuous interconnected porous emulsion system (BIPORES). This approach yields scaffolds featuring interconnected micropores and textured surfaces with hyperbolic curvature, seamlessly integrated within macroscale fibrous networks. Single-cell interactions assessed through optical trapping revealed rapid (within 30 seconds) ligand-free adhesion of human induced pluripotent stem cell-derived neural stem cells (i-HNSCs) to BIPORES, demonstrating for the first time geometry-driven neural attachment on PEGDA-based surfaces. Long-term cultures revealed

extensive migration toward the scaffolds with significantly higher cell densities on BIPORES compared to conventional polyornithine/laminin-coated 2D substrates. Immunofluorescence assays for β -III-tubulin and GFAP showed robust neuronal and astrocytic differentiation comparable to 2D controls. Calcium dynamics assessed via Fluo-8 AM revealed a twofold increase in spontaneous calcium transients, indicating pronounced functional neuronal maturation within BIPORES scaffolds. The interconnected microporous architecture sustained nutrient exchange and facilitated deep cell infiltration, supporting robust 3D neuronal network formation. These structural features persisted over two months with minimal degradation, highlighting the platform's suitability for protracted neuronal maturation. Collectively, our findings establish scaffold geometry as a powerful instructive cue, redefining bioactivity as a programmable feature of form rather than formula. This platform provides a versatile and scalable framework for advancing neural tissue modeling, regenerative therapies, and functional *in vitro* platforms.

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Poster

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Topic: A.02. Stem Cells and Reprogramming

Support: NIH Oxford Cambridge Graduate Partnership Program
Oxford Martin School Grant

Title: 3D constructs and differentiation timing shape maturation and connectivity of human NPCs allowing for generation of a bilaminar cortical implant

Authors: *G. GREENE¹, S. SAU², L. CAMPOS SOARES³, M. BARKAT¹, M. LI⁴, M. LAWSTON⁵, L. ZHOU⁴, H. BAYLEY⁴, A. KORETSKY⁶, F. G. SZELE¹, Z. MOLNÁR¹; ¹Physiology, Anat. and Genet., ²Univ. of Oxford, Oxford, United Kingdom; ³Physiology, Anat. and Genet., Univ. of Oxford, OXFORD, United Kingdom; ⁴Chem., Univ. of Oxford, Oxford, United Kingdom; ⁵NIMH, ⁶NIH, Bethesda, MD

Abstract: Neural progenitor cell (NPC) transplantation aims to replace lost tissue and stimulate regeneration following brain injury. Various studies have demonstrated the potential for implanted cells or organoids to survive and mature; however, optimizing the organization and neural connectivity of implant-derived cells remains a challenge. This study aims to examine whether the implantation of a pre-organized, microfluidic generated three-dimensional NPC construct can enhance integration and connectivity with the host brain circuitry. Additionally, we investigate whether the duration of *in vitro* differentiation of human iPSCs into cortical progenitor cells prior to implantation influences the implanted cells' tendency to maintain either

an upper or deep-layer cell phenotype post-implantation. Lastly, we evaluate the implantation of a fused bilaminar NPC construct with the goal of reconstructing a simplified cortical column. Human iPSCs were differentiated *in vitro* to generate early NPCs (ENPCs) resembling deep layer cortical cells or late NPCs (LNPCs) resembling upper layer cortical cells. The early or late NPCs were then either implanted as a cell suspension into the motor cortex of p8 mice or first incorporated into Matrigel constructs using microfluidic fabrication and subsequently implanted. Microfluidic generated constructs were implanted as either single ENPC or LNPC constructs or fused to create a bilaminar construct of ENPCs and LNPCs. Two months post-implantation (PI), NPCs in all groups formed mature neurons as seen by positive immunostaining for NeuN and extended axons to distant cortical targets. A higher percentage of NeuN positive cells was seen at 2 months PI in the 3D constructs in comparison to cells injected in suspension. In both the cell suspension and 3D construct conditions, LNPC implants projected primarily within the cortex, whereas early NPC implants extended axons cortically and to subcortical targets resembling the connectivity patterns of upper layer and deep layer neurons respectively. Our initial results provide a first step in controlling NPC connectivity following implantation which is crucial for potential clinical translation.

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Poster

PSTR255: Using Pluripotent Stem Cells to Model Human Diseases

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR255.30/A30

Topic: A.02. Stem Cells and Reprogramming

Title: Embodiment of the insoluble: Investigating nanoplastic neurotoxicity in models of the developing human brain

Authors: *A. HUDOCK, S. BENASSI, C. MARCHETTO;
Anthrop., Univ. of California San Diego, La Jolla, CA

Abstract: Recent reports suggest increased exposure of micro and nanoplastics (MNPs) in infants and young children compared to adults, underlining the need for experiments modeling chronic MNP during fetal development. Bioaccumulation of plastic occurs through ingestion, absorption, and inhalation of contaminated air, water, food, and soil sources. Across populations and tissues tested, plastic has been found in the placenta, lungs, blood, feces, urine, reproductive organs, gastrointestinal tract, and brain. There is an estimated total plastic weight of 4,917 µg/g in frontal cortex samples from people with a median age of 52, accounting for around 0.5% of the total brain weight, with these amounts considerably increasing from samples taken only 8 years apart. Exposure to and bioaccumulation of plastic occurs regardless of socioeconomic status and geography. Evidence suggests that plastic exposure and bioaccumulation are linked to

inflammation, toxicity, and cellular death in animal and cell models. These processes are at even more considerable risk during fetal brain development and could pose harmful consequences to human health on a global scale.

Cultivated and matured neural organoids and other cell culture techniques allow for an appropriate and direct model of human fetal brain development, and can provide information about the direct effects of exposure to different plastic types and concentrations. Using human cortical organoids, neural stem cells, and nanoplastics, we investigated the possible effects on neurogenesis, neural proliferation, cell death, cell viability, and neuronal activity for human cell cultures exposed to 3 different plastic concentrations versus controls, with the assistance of fluorescence microscopy, immunostaining, and microelectrode arrays. The goal of this project was to investigate processes associated with nanoplastic toxicity in a model of developing human brain tissue over 60 days, (representing a large portion of the first trimester of gestation in humans), to contribute to the growing body of international and interdisciplinary research outlining the possible genetic, behavioral, and physiological effects of environmental pollutants on human development. Overall, our study affirms neurotoxicity in cultures treated with nanoplastics versus controls. Additionally, we demonstrate using microelectrode arrays that nanoplastics, especially in more concentrated dosages, reduce the neuronal activity of human neurons compared to control groups, reiterating the possible consequences of chronic nanoplastic exposure experienced during fetal development.

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Poster

PSTR256: Autism: Physiology and Systems

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Topic: A.06. Developmental Disorders

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Academia Sinica Grant AS-IA-106-L04
NSTC Grant 112-2326-B-001-008
NSTC Grant 113-2326-B-001-008

Title: Mesoscopic mapping of circuit deficits in autism mouse models

Authors: *T.-T. HSU¹, C.-P. CHEN², M.-H. LIN¹, T. HUANG¹, C.-Y. WANG², Y.-P. HSUEH¹;

¹Inst. of Mol. Biology, Academia Sinica, Taipei, Taiwan; ²Inst. of Information Science, Academia Sinica, Taipei, Taiwan

Abstract: Autism spectrum disorder (ASD) is a disconnectivity syndrome, making it crucial to identify circuit deficits for understanding its etiology. We established a whole-brain mapping platform specific for histological fluorescent images from consecutive brain cryosections to analyze the circuit rewiring. Using this platform, we demonstrated structural and functional circuit rewiring in multiple ASD mouse models. First, we studied circuits from the basolateral amygdala (BLA) in a *Tbr1*^{+/-} autism mouse model by expressing oChIEF-Citrine in the unilateral BLA and analyzing Citrine⁺ axons and C-FOS expression. We found that *Tbr1* haploinsufficiency leads to inter- and intrahemispheric connectopathies, altered neuronal activities, and impaired synchronization among brain regions. Optogenetic theta-burst stimulation (TBS) of BLA partially restored the synchronization of the default mode network, which is linked to social behaviors. Consistently, TBS of BLA increases nose-to-nose social interactions in *Tbr1*^{+/-} mice, suggesting amygdalar stimulation may improve social function. Second, we analyzed the circuit rewiring with Thy1-YFP reporter across three ASD mouse models (*Tbr1*^{+/-}, *Nf1*^{+/-}, and *Vcp*^{+R95G}). While each model resulted in distinct connectivity alterations, visual, somatosensory, and piriform areas—were consistently affected across all models. The impact on visual and somatosensory areas varied among the models, but the piriform area consistently showed impairment, with reduced YFP signals and fewer YFP⁺ neurons in all three models. Moreover, all three mutants showed similar olfactory discrimination deficits, highlighting the vulnerability of sensory regions—particularly the piriform area—to ASD-related mutations. Overall, our whole-brain mapping platform effectively screens for circuit deficits in ASD mouse models, facilitating the identification of key brain regions for further investigation. More advanced reporter systems can be employed to analyze specific or multiple circuits, uncovering complex connectivity deficits associated with ASD.

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Poster

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Title: Sensory symptoms in autism spectrum disorder are associated with EEG complexity: multiscale entropy analysis

Authors: *T. NAOE¹, Y. SATO², Y. SHIKAUCHI¹, K. TANAKA², M. NAKAMURA¹;
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Abstract: The dynamic complexity of brain signals is linked to various psychiatric symptoms. The multiscale entropy (MSE) of electroencephalography (EEG) provides an index of the irregularity of temporal patterns of neural activity using sample entropy (SampEnt) over multiple time scales. Prior work has reported that individuals with autism spectrum disorder (ASD) exhibit reduced SampEnt at longer temporal scales—which correspond to lower complexity at the coarse scale—relative to typically developed (TD) controls. However, which ASD symptoms are the most closely associated with alterations in neural complexity remains unclear. Although the core features of ASD include impairments in reciprocal social communication and restricted repetitive behaviors (RRBs), several individuals with ASD also experience difficulties with sensory processing. This study investigated the relationship between resting-state EEG MSE and ASD-specific symptoms. Resting-state EEG data were acquired from 44 participants (ASD = 29; TD = 15). Statistical analysis confirmed that the results were consistent with those of previous studies which showed that SampEnt in the coarse-scale (i.e., time series scaled to 11.76-7.69 Hz) was lower in ASD than in TD. We also observed for the first time that SampEnt in the fine-scale (i.e., time series scaled to 25-50 Hz) was higher in ASD than in TD ($ts > 2.492$, $ps < .05$). Furthermore, we examined the Pearson correlations between EEG complexity and symptom severity using these scales, controlling for age and sex. In the ASD group, the fine-scale SampEnt was correlated with the severity of sensory symptoms (subscore of the adult/adolescent sensory profile). Specifically, the fine-scale SampEnt showed a significant negative correlation with sensory sensitivity (hypersensitivity), especially in the frontal regions ($r = .605$, $p < .01$), and with low registration (hyposensitivity), especially in the occipital regions ($r = .589$, $p < .01$). No significant correlations were observed between the severity of social communication or RRB symptoms and SampEnt on at these timescales in the ASD group. We demonstrated that EEG complexity reflects distinct ASD characteristics across different timescales.

Disclosures: T. Naoe: None. Y. Sato: None. Y. Shikauchi: None. K. Tanaka: None. M. Nakamura: None.

Poster

PSTR256: Autism: Physiology and Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR256.03/A33

Topic: A.06. Developmental Disorders

Support: DOD Grant AR240285

Title: Attention control contributions to sensory challenges in autism

Authors: *Y. TAKARAE¹, J. TOWNSEND², M. SOLOMON¹;

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Abstract: Sensory challenges are frequently experienced by autistic persons and could negatively impact attendance at social events, sleep quality, and other aspects of quality of life. Sensory challenges could occur because of alterations in fundamental sensory drives as suggested by studies indicating greater neural responses to sensory stimuli. Sensory challenges could, however, also reflect difficulty in regulating sensory input, including selecting and filtering sensory input, because it is well established that attention can enhance or dampen sensory-related neural activations. This perspective suggests that reduced attention control leads to inappropriate allocation of attention, for instance, resulting in hyperfocus on irrelevant and/or unpleasant stimuli (yielding sensory hypersensitivity or hyperreactivity) or difficulty filtering noise or distractors (sensory filtering problems). The current study investigated relationships between attention control and sensory challenges via an event-related potential (ERP) study. 9 autistic and 14 non-autistic adolescents/young adults participated in the study. They completed EEG recordings while they listened to repeating tones or watched a silent movie while the same tone played in the background. Participants also completed surveys of daily sensory challenges. Global Field Power based ERPs to the tone were compared between groups and conditions using running F tests. The group by condition interaction was significant in post-stimulus 204-266 ms and 330-398 ms, characterized by a greater reduction of auditory ERPs in the movie watching condition in the non-autistic group than in the autistic group. The finding could suggest that they were filtering the background tone while watching the movie, but to a lesser degree in the autistic group. Individual levels of this attention modulation of auditory ERPs were strongly related to sensory challenges in the autistic group, with greater attention modulation associated with less severe sensory challenges ($r=.59$). These findings suggest that attention control to regulate sensory input could be important in understanding the experience of daily sensory challenges in autism.

Disclosures: Y. Takarae: None. J. Townsend: None. M. Solomon: None.

Poster

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Program #/Poster #: PSTR256.04/A34

Topic: A.06. Developmental Disorders

Title: Whole-brain E/I imbalance linked to anxiety in autism via the Hurst exponent

Authors: *S. LAKAMSANI¹, S. BEELER-DUDEN², J. EILBOTT³, K. PELPHREY²;

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Abstract: Autism spectrum disorder (ASD) is characterized by social communication difficulties, restricted and repetitive behaviors, and a high prevalence of anxiety. A leading neurobiological hypothesis suggests that ASD involves an altered balance between neural excitation and inhibition (E/I), which may contribute to both core features and affective comorbidities. The Hurst exponent (H), a scale-free metric derived from resting-state fMRI (rs-fMRI), provides a noninvasive index of E/I dynamics, with lower H values reflecting E/I imbalance. While prior studies have identified reduced H in default mode and prefrontal regions in ASD, the relationship between whole-brain E/I dynamics and anxiety symptoms remains unclear. We analyzed rs-fMRI data from N= 148 (62 autistic individuals; 8-18 years) and computed Hurst exponent values across 268 brain regions defined by the Shen functional-connectivity atlas. Anxiety symptoms were assessed using the Child Behavior Checklist Anxious Depressed Subscale. We examined correlations between regional H values and anxiety scores across large-scale functional networks using multiple regression models controlling for age and motion. Autistic individuals showed widespread decreases in H values compared to control individuals, indicating increased E/I imbalance. Notably, decreased H in the right ventral temporal cortex, right caudate, left sensorimotor. There was also a significant interaction effect in the right anterior temporal gyrus between the diagnostic group and the Anxious/Depressed subscale of the CBCL. Control individuals exhibited a negative relationship between the subscale and H, while ASD individuals did not. This suggests that anxiety in Control individuals may reflect disruptions in intrinsic neural dynamics associated with heteromodal cortical integration. Autistic individuals showed altered neural dynamics indicative of increased excitatory/inhibitory imbalance, with key regional differences and distinct associations with anxiety-related symptoms compared to controls, suggesting divergent brain-behavior relationships in autism. Hurst mapping offers a promising neurophysiological marker for identifying individual differences in affective vulnerability and may inform targeted interventions to reduce anxiety in autistic individuals.

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Topic: A.06. Developmental Disorders

Support: NIH Grant R15NS141197
WWU Scientific and Technical Services

Title: Cannabidiol and terpenes modulate endocannabinoid expression in the BTBR mouse model of autism spectrum disorder

Authors: K. POTTER, J. CALDWELL, J. MUCKERHEIDE, Z. WILSON, H. R. JAMES, *J. KAPLAN;

Behavioral Neurosci. Program, WESTERN WASHINGTON UNIVERSITY, BELLINGHAM, WA

Abstract: Cannabidiol (CBD) is a non-intoxicating phytocannabinoid with emerging therapeutic benefits across numerous neurological conditions including autism spectrum disorder (ASD). We've previously demonstrated that CBD increases social interaction preference in the BTBR mouse model of ASD, and this enhancement is more reliable in the presence of a cannabis inspired terpene blend. Therefore, CBD-based treatment approaches may be improved through the systematic addition of cannabis terpenes. BTBR mice, as well as individuals with ASD, often have reduced expression of the endocannabinoid, anandamide, and boosting anandamide signaling in mouse models improves ASD-related symptoms. Therefore, anandamide may be a targetable substrate for screening CBD and terpene compositions that most effectively and reliably improve behavioral phenotypes. We hypothesized that combinations of CBD and terpenes would lead to more pronounced increases in anandamide with little impact on the expression levels of the other endocannabinoid, 2-AG. We tested this hypothesis in this exploratory study using two-dimensional MALDI-TOF mass spectrometry on sagittal brain slices of adult male and female BTBR mice following acute passive vapor inhalation of either CBD, terpenes, or CBD and terpenes in combination. Our preliminary data suggest that CBD increases anandamide, but not 2-AG. Further, although the prominent cannabis terpene, beta-caryophyllene, did not impact anandamide levels on its own, combining it with CBD led to a substantial increase in global anandamide expression. Thus, our preliminary results support our hypothesis that adding terpenes, such as beta-caryophyllene, to CBD increases its impact on anandamide levels, and thus, should increase its predictive validity as a reliable treatment. Sex differences in CBD sensitivity contribute an additional challenge to the predictive validity of CBD-based treatment approaches. We hypothesized that CBD's impact on anandamide expression would vary as a function of estrous period. We tested this hypothesis in a separate cohort of female BTBR and found that CBD's increase in global anandamide expression was most pronounced during estrus. In summary, our results suggest that terpenes can enhance CBD's effects on one potential therapeutic target in ASD while revealing an important impact of sex hormones on the magnitude of these effects. Our future studies seek to continue optimizing terpene compositions, applying those formulations in behavioral assessments, and confirming their therapeutic mechanism of action.

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Poster

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Program #/Poster #: PSTR256.06/A36

Topic: A.06. Developmental Disorders

Support: NIH Grant, PA-18-401, 5R01NS121120-05

Title: Atypical visually guided precision grip control and its association with perceived physical health quality in middle-aged and older autistic adults

Authors: *E. M. PARKS¹, H. QU², D. CHRISTENSEN², H. M. GEMMELL², K. E. WETHERINGTON¹, A.-M. ORLANDO³, R. A. ROMERO³, B. KARMAKAR³, D. E. VAILLANCOURT², Z. WANG²;

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Abstract: Sensorimotor impairments are common in autism spectrum disorder (ASD), yet little is known about how these difficulties manifest in middle-aged and older autistic adults and their relation to broader physical health outcomes. This study examined 52 autistic and 56 age- and sex-matched non-autistic adults (aged 30-73) using a visually guided precision grip task to assess temporal (reaction time and duration), spatial (force accuracy and variability), and dynamic (rate of force change) features of grip control under varying motor output demands (target force test) and visual gain feedback (visual gain test). Quality of life was examined using the World Health Organization Quality of Life-BREF (WHOQoL-BREF) instrument. Autistic adults exhibited prolonged duration, delayed reaction time, and greater target overshooting at lower force levels during the rise phase of precision gripping. During the sustained phase, autistic adults exhibited increased grip force variability compared to non-autistic adults across both tests. In contrast, autistic individuals exhibited shorter reaction times during the relaxation phase during these tests. We observed that quality of life was significantly lower in autistic adults across all WHOQOL-BREF domains, and poorer self-reported physical health ratings were significantly associated with several grip force measures. These findings demonstrate that manual motor impairments are persistent beyond childhood and across adulthood in ASD. This work also suggests a potential link between manual motor control deviations and perceived health outcomes, offering a promising avenue for future interventions targeting manual dexterity to promote physical health and autonomy in aging autistic individuals.

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Poster

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Program #/Poster #: PSTR256.07/A37

Topic: A.06. Developmental Disorders

Support: US Army

Title: Involvement of amygdala neurons in male predominance of autism spectrum disorder

Authors: *N. MONTEFIORE¹, Y. YAROM²;

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Abstract: Male predominance is one of the least understood characteristics of autism spectrum disorder (ASD). While more than a hundred genes are involved in ASD, only a few of them show sex differences in behavior. In previous work, we found that male mice harboring a mutation in the *Pogz* gene (*Pogz*^{+/-}) show sexually dimorphic overly friendly behavior in several sociability assays. Mapping the engaged neurons during social recognition revealed that in *Pogz*^{+/-} males, there are more activated neurons in the basolateral amygdala (BLA) than in control and *Pogz*^{+/-} female mice. To characterize the morphophysiological properties of these BLA neurons, we conducted in-vitro whole-cell recordings in acute brain slices from both *Pogz*^{+/-} and control mice. We employ targeted recombination in active populations (Trap2) to label the neurons in response to a social odor test. Our results from single-cell recording from the BLA present two types of principal neurons, characterized by spike burst with the initiation of current pulse, unique AHP and spike train accommodation in response to a long current injection. Our results also indicate that the neurons active during the social odor test exhibit characteristics similar to a specific subtype of principal neurons we identified in the BLA of mice harboring the *Pogz* mutation. Initial experiments are now underway to optogenetically label the olfactory inputs to the BLA in order to further examine the activity of these neurons in response to a novel social odor. In these experiments, we activate ChR2-labeled terminals in brain slices containing the BLA while recording from labeled and unlabeled neurons in mice that have undergone the social odor test. These experiments will further expose any correlation between male predominance and overly friendly behavior and the basis of the properties of the neuronal network. Our results will pave the way to understanding the role of amygdala neurons in ASD model mice and advance our knowledge about autism spectrum disorder.

Disclosures: N. Montefiore: None. Y. Yarom: None.

Poster

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Topic: A.06. Developmental Disorders

Support: NIH Grant R01MH096847
NIH Grant R01NS110307

Title: Neurobiology of NDD Elopement: Syngap1 expression within cortical-amygdala circuits regulates the intersection of sensory processing and negative valence memory retrieval

Authors: *C. Y. PAN^{1,2}, D. G. BALAZSFI^{3,4}, T. VAISSIÈRE⁴, C. ROJAS⁴, G. RUMBAUGH^{2,4};

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Abstract: Elopement, characterized as sensory-induced escape independent of context, frequently occurs in individuals with autism spectrum disorder (ASD) and intellectual disability (ID). These abrupt reactions, triggered by sensory discomfort or anxiety, significantly increase the risk of serious injury or fatal outcomes, underscoring the urgent need to understand the neural circuits underlying these behaviors. Elopement is an issue in *SYNGAP1-Developmental and Epileptic Encephalopathy*, a genetic ID disorder with co-occurring ASD caused by *SYNGAP1 de novo* loss-of-function variants. Prior work from our lab has shown that *Syngap1* regulates cortical sensory processing linked to cognitive impairment and behavioral dysregulation. We hypothesized that *Syngap1* deficiency within cortico-amygdala valence circuits disrupts the processing of auditory-based fear reminders, which in turn drives expression of improper defensive behaviors. *Syngap1* heterozygous mice, which models the human genetic disorder, dart rather than freeze specifically in response to the presentation of an auditory tone that elicits retrieval of a negative valence (e.g., fear) memory. Strikingly, expression of cue-induced darting coincided with a heart-rate spike rather than the expected bradycardia in this model, demonstrating that a fear reminder elicited a maladaptive brain and behavioral state. Restricting *Syngap1* haploinsufficiency to forebrain excitatory neurons (*EMXI*+ population) phenocopied the cue-induced brain and behavioral state as evidenced by darting accompanied by a heart-rate spike. Conversely, targeted *Syngap1* re-expression within the *EMXI*+ population normalized the fear-induced brain and behavioral response - freezing with bradycardia was now elicited by the fear reminder. Furthermore, *Syngap1* knockdown largely restricted to amygdala neurons recapitulated coordinated cue-induced darting with a heart rate spike. Finally, neuronal calcium imaging through a GRIN lens implanted above the basolateral amygdala (BLA) of *Syngap1* heterozygous mice revealed evidence of altered auditory cue processing at the time of fear memory retrieval. Together, our data demonstrate that *Syngap1* expression within valence processing circuitry is essential for coordinating proper adaptive behavioral and autonomic responses elicited by a neutral sensory stimulus associated with past negative experiences. Ongoing experiments are focused on understanding how *Syngap1* regulates neuronal representations of neutral, aversive, and appetitive stimuli within the BLA, and how these representations guide coordinated brain/body adaptive responses to the environment.

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Poster

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Topic: A.06. Developmental Disorders

Support: NIMH Grant R01MH103494

Title: Cerebellar lobular volumes in middle-age and older adults with autism spectrum disorder

Authors: ***B. BLOOMER**¹, S. PEDRAHITA², M. CORDOVA¹, J. S. KOHLI², S. C. TUY², A. C. LINKE², J. HAU², M. ELDEN², G. TORO², M. KINNEAR ELSON², R.-A. MUELLER², R. A. CARPER²;

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Abstract: Evidence from lesion and neuroimaging studies point to an important role of the cerebellum in higher order cognition and motor behavior. Decreasing cerebellar volume during typical aging has been linked to declining implicit and procedural learning, gait, balance, and working memory. The cerebellum is also impacted in autism spectrum disorder (ASD), with converging evidence across postmortem and neuroimaging studies finding reduced Purkinje cell counts in ASD cases as young as 4 years and cerebellar hypoplasia across multiple lobules. In children and adults with ASD, structural and functional differences are associated with social-communication impairments and increased restricted and repetitive behaviors, hallmark domains of ASD. Most research in autism has been conducted in child, adolescent, and young adult samples, with scant research on aging. Individuals with ASD experience atypical trajectories of early brain development, raising the possibility that brain aging may differ as well. Critically, altered early neurodevelopment may interact with the typical processes of aging, creating a risk of early-onset or accelerated brain aging in autism. We hypothesized that total cerebellar volume, Crus I, Crus II, lobules VI, VII, VIIIA, and VIIIB would be decreased in ASD compared to NT groups, and expected ASD adults to show steeper negative effects of age on subregion volumes compared to NT. 32 adults with ASD and 42 NT adults aged 40-70 years were included. T1-weighted MRI scans were collected on a 3T GE MR750 scanner and processed using CERES2 software to parcellate all cerebellar lobules (12 lobules per hemisphere). All group difference and interaction analyses controlled for intracranial volume (ICV) and image quality rating (QR). Right Lobule VI volumes were significantly decreased in ASD compared to NT ($\beta = 0.43$, $p = 0.045$). However, this finding did not survive FDR correction. Additionally, there was a significant Group x Age interaction for right cerebellar Lobule VI volume ($\beta = 0.07$, $p = .035$), but NT individuals showed a positive age-related age effect, while ASD individuals showed no significant age effect. Contrary to hypotheses, total cerebellar volume and volumes of other lobules were not significantly different between ASD and NT groups (*all* $p > 0.05$). While these cross-sectional data did not provide strong evidence of differential cerebellar aging in ASD, longitudinal follow-up (currently underway) may better disentangle developmental effects from aging-specific effects. Examination of associations between cerebellar structure and cognitive measures will help clarify the potential consequences of cerebellar change during aging in ASD.

Disclosures: **B. Bloomer:** None. **S. Pedrahita:** None. **M. Cordova:** None. **J.S. Kohli:** None. **S.C. Tuy:** None. **A.C. Linke:** None. **J. Hau:** None. **M. Elden:** None. **G. Toro:** None. **M. Kinnear Elson:** None. **R. Mueller:** None. **R.A. Carper:** None.

Poster

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Topic: A.06. Developmental Disorders

Support: NIH Grant R21AG 065621
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Title: Persistent deviations in non-constrained walking and grooved pegboard test performance in middle-aged and older autistic adults

Authors: *H. QU^{1,2,3}, M. TERZA³, H. GEMMELL^{2,3}, D. SHIRLEY^{2,4}, A.-M. ORLANDO^{5,6,7}, B. KARMAKAR⁸, Z. WANG^{2,3};

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Abstract: Autism Spectrum Disorder (ASD) is a lifelong neurodevelopmental condition that requires sustained support across the lifespan. Recent evidence suggests an elevated risk for early-onset neurodegenerative conditions, including Parkinsonism, among autistic individuals. However, empirical research on neuromotor function in middle-aged and older autistic adults remains scarce, and age-related changes in gross and fine motor skills are poorly understood in this unique clinical population. Motor atypicalities—such as increased gait variabilities and impaired finger coordination—are commonly observed in young autistic individuals and recognized as sensitive biobehavioral markers predicting the onset and progression of neurodegenerative conditions. Despite their clinical relevance, deviations in gait kinematics and manual dexterity have not been comprehensively examined in autistic adults beyond early adulthood. To fill this gap, we administered non-constrained walking and grooved pegboard tests to 46 autistic adults aged 30 to 73 years and 46 age-, sex-, and full-scale IQ matched non-autistic controls. We derived multiple spatiotemporal and joint angle measures from the non-constrained walking. For the pegboard test, we assessed the completion times and number of pegs dropped for both dominant and non-dominant hands. Compared to non-autistic individuals, autistic adults exhibited greater stride-to-stride variability in cadence, step time, step length, step velocity, hip angle at heel strike, and knee angle at toe-off. Autistic adults also had significantly longer completion times for both hands during the pegboard test, although the number of dropped pegs did not differ between groups. Across both groups, older age was associated with increased variability in knee angle at toe-off and longer completion times for the non-dominant hand; however, no significant group-by-age interactions were observed. These findings highlight the persistence of neuromotor control impairments in autistic adults into later life. Age-related changes were observed in some neuromotor measures, though they appeared to affect both groups similarly. Further research is warranted to determine whether these neuromotor tests may serve as early indicators of neurodegenerative risk and to explore their potential as intervention targets for promoting autonomy and functional independence in autistic adults.

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Poster

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Title: Does the personality of individuals on the autism spectrum influence their ability to decode mental states?

Authors: *E. PISANU¹, M. ZANON², C. STEA⁴, L. COLLARI⁴, A. RIOLO⁵, R. RUMIATI³; ¹Neurosci. Area, ³Area of Neurosci., ²International School for Advanced Studies (SISSA), Trieste, Italy; ⁴Ctr. for Autism Spectrum Disorders and Cognitive Disabilities, Univ. Hosp. ASUGI, Trieste, Italy; ⁵Ctr. for Autism Spectrum Disorders and Cognitive Disabilities, University Hosp. ASUGI, Trieste, Italy

Abstract: Personality modulates several socio-emotional processes related to Theory of Mind (ToM) in neurotypical individuals, however, little is known about how these dynamics work in neurodivergent populations particularly in individuals with Autism Spectrum Disorder (ASD). The role of personality and individual differences in ASD is an emerging research topic that has received limited attention. To date, research has primarily focused on behavioral outcomes, relying on self-reports or other-reports, with conflating results from samples differing in age and diagnosis, therefore making results difficult to interpret. Furthermore, behavioral studies alone are not sufficient to unveil potential compensatory strategies, as they only capture the final output of complex internal processes. This EEG examines whether young adults with ASD without language or intellectual impairments have a distinct personality profile and by exploring how agreeableness personality trait may modulate brain processing during different socio-emotional tasks related to ToM. Personality traits of 42 young adults with ASD, assessed using the Big Five Inventory, were compared to those of 462 typically developing controls. ToM performance and neural activity, measured via EEG, were further compared to 62 controls selected from the original pool based on agreeableness scores.

ASD participants scored lower on all Big Five traits except openness, with substantial within-group variability. They showed comparable mental state decoding accuracy to less agreeable controls but underperformed relative to highly agreeable ones. ERP data revealed a delayed and reduced N170 response in ASD participants during stimulus processing and a smaller late slow wave (LSW) during mental state decoding. The LSW differences were more pronounced when comparing ASD participants to highly agreeable controls than to less agreeable ones. This study

highlights that individuals with ASD generally score lower on most Big Five traits, although they do not consistently exhibit extreme trait values, underscoring the importance of tailored interventions that leverage individual strengths. Having the same diagnosis does not necessarily imply uniformity among individuals with that diagnosis. Notably, ASD individuals showcased a capacity for compensatory mechanisms, achieving behavioral performance in decoding mental states akin to less agreeable healthy controls despite neural differences. Overall, this study offers valuable insights into the multifaceted nature of personality within the ASD context.

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Poster

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Program #/Poster #: PSTR256.12/A42

Topic: A.06. Developmental Disorders

Title: Functional disruption of the LHb-VTA pathway reveals core ASD-like behavioral deficits

Authors: *M. SCORRANO^{1,2}, G. SAGONA^{1,3}, A. ALLEGRA MASCARO^{1,4}, G. TARGA⁵, M. CAMBIAGHI^{5,6}, L. BARONCELLI^{1,3};

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Abstract: The Lateral Habenula (LHb) integrates aversive and reward-related information by exerting top-down control over midbrain dopaminergic pathways, positioning it as a key regulator of motivational states. While its function has been thoroughly characterized in the context of affective disorders, its contribution to neurodevelopmental conditions such as Autism Spectrum Disorders (ASDs) remains insufficiently explored. ASDs are characterized by impairments in social behavior, communication, and cognitive flexibility, and disruptions in reward processing have been increasingly implicated in their etiology. Intriguingly, neuroimaging evidence of habenular enlargement in individuals with ASD raises the possibility of a functional role for LHb hyperactivity in the emergence of core symptoms. To investigate this hypothesis, we employed a chemogenetic approach to selectively activate LHb neurons in wild-type mice. Global stimulation of the LHb produced pronounced deficits in sociability and cognitive flexibility, while targeted activation of LHb projections to the Ventral Tegmental Area (VTA) selectively recapitulated the social impairments. These findings pinpoint the LHb-VTA circuit as a critical substrate modulating social behavior and reveal that its dysregulation is capable of inducing behavioral signatures that mirror key features of the autistic phenotype. This

work positions the LHB not merely as a relay for aversive signaling but as a modulatory hub whose activity shapes fundamental dimensions of social cognition.

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Poster

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Topic: A.06. Developmental Disorders

Support: ANR
IReSP
INSERM

Title: Co-emergence of hippocampal circuit properties and cognitive function during development

Authors: *X. LEINEKUGEL¹, A. FERREIRA GOMES DA SILVA², O. J. DUBANET³, H. ROUAULT⁴;

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Abstract: During development, the maturation of functional inhibition and of specific neuronal activity patterns are essential prerequisites for the emergence of cognitive functions, and this structure-function relationship may underlie the cognitive deficits observed in neurodevelopmental disorders. We used advanced in vivo electrophysiological recordings together with a novel behavioral phenotyping platform (Phenotypix) to investigate the co-development of hippocampal circuit properties and cognitive capacities.

A prominent pattern of activity is hippocampal Sharp Wave Ripples (SPW-Rs), considered essential for learning and memory. During SPW-Rs, sequences of awake coding activities are replayed at fast time scale, in a rhythmic timing that might promote synaptic plasticity and a high level of synchrony that may facilitate summation and transfer of information to downstream structures. Previous work reported the emergence of ripples at P10 in the CA1 region, together with the development of inhibition. On the other hand, neither the relationships between perisomatic inhibition and ripples or their developmental emergence have been investigated in CA3, in which ripples have a different frequency profile (90-110Hz instead of 140-200Hz in CA1), functional perisomatic inhibitory circuits have different properties, and some developmental aspects such as neurogenesis or interneuron maturation are early compared to CA1. We have here investigated the hypothesis of a conjoint and earlier appearance and maturation of ripples and perisomatic inhibition in the CA3 hippocampal region compared to

CA1.

We report that the emergence of perisomatic GABAergic synaptic activity correlates with that of ripple activity. Interestingly, perisomatic inhibition and ripple activity follow parallel maturation trajectories, beginning in CA3 at P7 with immature (unfunctional) inhibition and proto-ripples that exhibit labile frequency. A transition period occurs between P10 and P12, during which mature functional perisomatic inhibition and stable ripple activity progressively emerge, reaching adult-like properties by P13. This developmental trajectory also meets the emergence of key aspects of cognition, as evidenced by behavioural investigations.

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Poster

PSTR256: Autism: Physiology and Systems

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Topic: A.06. Developmental Disorders

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Title: Neural responses to infant-directed speech and song in autism spectrum disorder

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Abstract: Many treatment studies in autism spectrum disorder (ASD) have shown that toddlers with ASD often respond poorly to speech cues, particularly to high-affect, infant-directed speech, which elicits strong engagement in typically developing (TD) toddlers. In contrast, a growing body of research indicates that music-based interventions incorporating speech elements through singing can be highly effective for children with ASD. This raises a critical question: what specific features of enhanced musicality drive stronger responses in ASD, and are these responses purely behavioral, or do they reflect underlying neural mechanisms? We hypothesized that in toddlers with ASD, activation in typically speech-processing areas of the temporal cortex would correlate positively with the musicality of the stimuli rather than with the amount of speech. Specifically, we predicted that instrumental music would evoke the strongest neural response, while infant-directed speech would elicit the weakest. Conversely, we anticipated that TD toddlers would exhibit robust temporal cortex activation in response to social speech, but only nonspecific auditory processing in response to music, reflecting a lack of speech-related

engagement. To test these hypotheses, we conducted a multimodal study involving behavioral and cognitive assessments, structural and functional MRI, and parental surveys across a cohort of over 50 toddlers, including those with ASD, non-ASD developmental delays, and typical development. During fMRI scanning, toddlers were exposed to three auditory paradigms: infant-directed speech ("motherese"), vocal (a cappella) lullabies, and instrumental lullabies. We observed distinct patterns of neural activation, particularly in the temporal cortex, across diagnostic groups in response to these stimuli and identified meaningful links between neural responses and clinical phenotypes. Our findings contribute to a deeper understanding of how musical and speech stimuli are processed differently in ASD and may help guide the development of targeted interventions. Specifically, they highlight the potential of music-based approaches to enhance engagement and communication in young children with ASD, and offer insight into how speech-based methods might be adapted for greater efficacy.

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Poster

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Title: Preliminary evidence of atypical white matter microstructure along the 'third' visual pathway in children and adolescents with autism

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Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental condition associated with challenges in social communication and the presence of restricted and repetitive behaviors. Individuals with ASD often struggle to interpret social cues such as eye gaze, facial expressions, and gestures, which rely on the perception of biological motion. Atypical functioning of the posterior superior temporal sulcus (pSTS), known to selectively respond to biological motion, has been reported in people with ASD. However, the structural connectivity supporting this region remains understudied. Recent evidence suggests a distinct "third" visual pathway linking visual cortices to the pSTS, hypothesized to support visual input for social perceptual and multisensory processing. This study examined the anatomy and microstructure of this pathway in

ASD utilizing archival diffusion MRI data (61 directions at $b=1000\text{s/mm}^2$, voxel size = $1.88 \times 1.88 \times 2\text{ mm}^3$) acquired in a cohort of children and adolescents aged 7 to 19 years ($n=97$; ASD: 15 female, 47 male; typically developing [TD]: 5 female, 30 male). The diffusion images underwent standard preprocessing to correct for head motion and image distortions. Following whole-brain probabilistic tractography, left and right pSTS-occipital tracts were segmented by filtering streamlines terminating in the banks of the STS and all occipital regions from the Desikan-Killiany atlas. The tracts were visually inspected for quality control. Fractional anisotropy (FA) and mean diffusivity (MD) were sampled along all tract streamlines and averaged per tract. Analyses of covariance were performed to test for differences in tract FA and MD between diagnostic groups with age and in-scanner head motion as covariates. The left and right tracts showed similar distributions of occipital terminations with the majority in the lateral occipital cortex (75%), followed by the lingual gyrus (12%), pericalcarine cortex (7%), and cuneus (6%), consistent with known connections to early visual (V1, V2) and motion-sensitive (V5/MT) areas. Compared to TD peers, the ASD group showed significantly reduced FA (left: $p<.001$, $\eta^2=0.13$; right: $p<.001$, $\eta^2=0.16$) and increased MD (left: $p=0.03$, $\eta^2=0.05$; right: $p=0.03$, $\eta^2=0.09$). Significant age effects were found for FA (positive; left: $p<.001$, $\eta^2=0.11$; right: $p<.001$, $\eta^2=0.13$) and MD (negative; left: $p<.001$, $\eta^2=0.26$; right: $p<.001$, $\eta^2=0.37$), as expected for this age group. These results provide evidence of disrupted structural connectivity, and potentially atypical maturation, of the third visual pathway in individuals with ASD, suggesting a neural basis for difficulties in dynamic social perception.

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Poster

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Topic: A.06. Developmental Disorders

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Title: Altered microstructure of the cerebellar peduncles in young children with autism spectrum disorder

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Abstract: Gross and fine motor delay and atypical function are common in children with Autism Spectrum Disorder (ASD). Among non-heritable risk factors, cerebellar injury at birth carries the

greatest likelihood of developing autism, with the risk ratio being 5 times greater than that associated with premature birth, for instance (Wang et al., 2014). Further, the cerebellum has been increasingly recognized for its involvement in functions beyond motor coordination, including multisensory integration, executive function, and emotional regulation, all shown to be implicated in ASD. While evidence of atypical cerebellum morphology and cerebro-cerebellar functional connectivity in ASD is well-established, less is known about the cerebellar white matter connectivity, particularly in early childhood, when symptoms first manifest. Utilizing multi-shell diffusion magnetic resonance imaging (dMRI) data acquired during natural sleep (1.7mm³ voxels, 6 *b*0s and 47 or 46 directions at *b*=1500 and 3000s/mm², respectively) in a cohort of 1½-5 year old children enrolled in a study of early brain markers of autism (ASD: n=36, typically developing [TD]: n=26; *mean age* = 43±15 months), we aimed to examine microstructural indices of the inferior, middle, and superior cerebellar peduncles (i.e., the afferent and efferent fibers of the cerebellum). Following standard image preprocessing, the cerebellar peduncles were delineated via automated tract segmentation (performed by TractSeg), and the streamline-weighted averages of the outcome microstructural variables (fractional anisotropy [FA], mean diffusivity [MD], neurite density [ND], and orientation dispersion [OD]) were extracted for each tract. Linear regression models revealed significant effects of group on FA and ND of the bilateral superior and middle cerebellar peduncles and the left inferior cerebellar peduncles (ASD > TD, all *p* ≤ .02), while controlling for age, sex, and estimated in-scanner head motion (average root mean square displacement). Significant effects of age on all outcome variables (increased FA and ND, and decreased MD and OD) were also observed, as expected in this age group. Results point to an early maturation of cerebellar white matter microstructure - connecting the cerebellum indirectly to the cerebral cortex, via brainstem, and the spinal cord - in young children with autism. While these data do not allow inferences about when or how early in life the peduncles' overgrowth commences or its neurogenesis, they provide evidence that cerebellar white matter development is accelerated in early childhood in autism, potentially contributing to the diverse motor and non-motor manifestations of ASD.

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Poster

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Title: Machine learning assisted identification of frontotemporal pathway pathology in autism

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Abstract: Autism Spectrum Disorder (ASD) is a complex neurodevelopmental disorder characterized by atypical neural communication and connectivity. Prefrontal (PFC) and temporal cortices as well as the pathways connecting the two are affected in ASD but the mechanisms underlying pathology are not well understood. We used histopathology methods and high resolution bright-field (BF) and electron microscopy (EM) imaging to analyze the microstructure of frontal and temporal areas, as well as the deep white matter pathway that connects them, the uncinate fasciculus (UF). This analysis was conducted on post-mortem brain tissue from 13 control (CTR) and 12 age and sex-matched ASD adults. We focused on areas interconnected through the UF, including the frontal anterior and subgenual cingulate cortices, the orbitofrontal cortex, and the medial temporal areas 28, 35, and 36 (MTL). We collected hundreds of thousands of images at micrometer and nanometer scales and analyzed representative samples to quantitatively estimate neurotypical and atypical white matter features to compare across conditions. We identified novel pathological changes, especially among thin and thick axons, with decreased axon density and variable changes in myelin thickness in the UF and temporal regions and confirmed pathology in PFC. We then processed all captured photomicrographs to create a dataset with over one million images and, using machine learning approaches, we trained convolutional neural networks for histopathological image classification. To ensure generalizability of the models, we used different cases for training, validation, and testing, and achieved up to 92% accuracy in pathology classification in UF. We extended this to a multitask model using images from PFC, MTL, together with UF to classify pathology and region simultaneously. Moreover, in order to visualize and explain artificial intelligence network performance and classification processes, we used several approaches that can reveal traits: we used Grad-CAM heatmaps to highlight discriminative regions and DeepDream Images (DDI) to show the prototypical traits and patterns of the classified groups. High accuracy, up to 92%, in classification of pathology and discriminative Grad-CAM and DDI aligned with our image analyses and highlighted key differences across conditions and areas. We used Principal Component Analysis (PCA) to extract key features dimensions and the variance they explain. Together, these results confirmed our histological findings and demonstrated that explainable machine learning can reliably detect ASD-related microstructural pathology across frontotemporal regions.

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Poster

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Title: Association of glymphatic dysfunction, free water, white matter integrity and long-term memory performance in aging autistic brain

Authors: *Y. ZHANG¹, E. OFORI¹, S. A. HARKER¹, M. VELEZ GALINDO², S. GALLEGOS¹, D. HUGHES¹, L. BAXTER⁴, K. CHEN⁵, B. WOODRUFF⁴, H. VENKATESH¹, J. BLACK¹, S. STRASIA¹, B. BRADEN³;

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Abstract: Autistic adults are at elevated risk of accelerated cognitive aging and Alzheimer's disease and related dementias (ADRD), yet the underlying neurobiological mechanisms remain poorly understood. Dysfunction in the glymphatic system—a brain-wide network responsible for clearing waste via interstitial fluid flow—may contribute to this vulnerability by promoting extracellular free water accumulation and white matter (WM) degeneration. In this study, a total of 113 autistic and 90 age- and sex-matched neurotypical (NT) adults (aged 18-71 years) were included and underwent multimodal MRI scanning and episodic memory assessments. Diffusion tensor image analysis along the perivascular space (DTI-ALPS) index was computed, alongside free-water and fractional anisotropy (FA) maps, for each participant. Group comparisons, correlations, and mediation analyses were performed. Compared to NT adults, autistic adults showed significantly lower DTI-ALPS values, increased fornix free-water, reduced fornix FA, and poorer memory scores. For hippocampal free-water, a significant group by age interaction demonstrated a stronger positive correlation with age in the autistic group than the NT group. Mediation analyses revealed that fornix free-water mediated the relationship between DTI-ALPS and both fornix FA and hippocampal free-water. Long-term memory scores correlated with fornix FA and whole-brain gray matter free-water indices in autistic adults. Our results suggest that glymphatic dysfunction and free water accumulation may contribute to WM degeneration and episodic memory challenges in autistic adults across a broad age range. These findings point to potential biomarkers for identifying and intervening in the cognitive aging process in autism.

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Poster

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Title: Structural and functional graph measures: differences between aging autistic and neurotypical populations and correlations with longitudinal cognitive change scores

Authors: *D. HUGHES¹, L. RUHTER², Y. ZHANG³, B. BRADEN⁴, S. M. CROOK¹;
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Abstract: Recent studies suggest increased risk for dementia in aging autistic (ASD) adults. Our past work has shown accelerated memory and hippocampal volume decline in ASD aging adults compared to neurotypical (NT) aging adults. Here, we used graph theory to quantify structural and functional connectivity (SC, FC) to identify potential factors affecting cognitive aging outcomes in ASD adults. We identified baseline graph theory metrics that significantly differed between groups or significantly correlated with cognitive change over time. Participants include 33 ASD and 29 NT adults aged 39-71 at visit one. Subsequent visits occurred at two year intervals (2-5 total visits). Diffusion, T1, and functional MRI scans were acquired and the Auditory Verbal Learning Test (AVLT) was administered at each visit. We construct SC and FC matrices from MRI data and applied thresholds of 70% and 65% sparsity, respectively. We then computed global and nodal graph theory metrics. Nodal metrics were computed on the left and right hippocampus (L_Hippo, R_Hippo). Longitudinal AVLT long-term memory (A7) slopes were computed with linear mixed modeling. Baseline graph theory metrics were compared between ASD and NT groups and correlated with individual memory slopes. Significant differences were found between ASD and NT groups for the global measures structural radius and functional transitivity, and the nodal measure functional betweenness centrality for L_hippo. In the ASD group, SC characteristic path length ($r = -0.38$) and diameter ($r = -0.39$) were significantly correlated with memory slopes. Significant correlations were also observed in the ASD group between memory slopes and FC R_Hippo eccentricity ($r = -0.38$). In contrast, the NT group showed significant correlations between memory slopes and SC R_Hippo strength ($r = 0.47$), L_Hippo strength ($r = 0.37$), as well as L_Hippo clustering coefficient ($r = 0.38$). We identified structural and functional correlations between whole brain and hippocampal-specific baseline graph theory metrics and longitudinal memory change in ASD and NT groups. For ASD adults, measures where higher values indicate less tightly connected brain graphs are correlated to negative memory change scores (i.e. worsening AVLT A7 scores over time). Structural NT measures where higher values indicate the R_Hippo and L_Hippo have stronger connections or are more tightly locally connected are correlated with positive memory change scores. These preliminary findings suggest the potential for unique MRI-based biomarkers to identify ASD adults who are most vulnerable to accelerated memory decline, as well as potential brain mechanisms of risk.

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Poster

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Title: Age-related differences in behavioral and brain predictors of parkinsonism in autistic adults

Authors: *M. VELEZ GALINDO¹, G. L. WALLACE³, G. A. MCQUAID⁴, D. S. PETERSON², D. Q. BEVERSDORF⁵, S. A. HARKER¹, S. GALLEGOS¹, Y. ZHANG¹, A. RODI¹, C. BAYOUTH¹, J. SMITH-PLATA¹, K. GRABEEL¹, E. OFORI², B. BRADEN²; ¹Arizona State Univ., Tempe, AZ; ²Arizona State Univ., Phoenix, AZ; ³Speech and Hearing Sci., George Washington Univ., Washington, DC; ⁴George Mason Univ., Fairfax, VA; ⁵Dept Radiol, Neurol, Psychol Sci, DGS of INP, Univ. of Missouri Columbia, Columbia, MO

Abstract: Autistic adults may face elevated risk for parkinsonism, yet the behavioral and brain mechanisms underlying this vulnerability are poorly understood. Motor difficulties linked to restricted, repetitive behaviors and interests (RRBIs) are common in autism and may overlap with parkinsonism, but few studies have examined these associations across the adult lifespan. We examined self-reported parkinsonism features (Parkinsonism Screening Questionnaire; PSQ) and RRBIs (Adult Repetitive Behaviors Questionnaire-2; RBQ-2A) in two independent samples of autistic adults: an Arizona cohort (n=215; autistic=114, non-autistic=101; ages 18-71) and a national cohort from SPARK (Simons Foundation Powering Autism Research for Knowledge; autistic n=210; ages 42-80). In the Arizona sample, we also collected finger tapping and MRI-derived motor brain structure data. Pearson correlations and linear regression were used to examine group differences and associations with age, behavior, and brain measures. In the Arizona sample, autistic adults showed significantly higher PSQ (p=.001) and RBQ-2A scores (p=.001) than non-autistic adults. A group-by-age interaction was found for RBQ-2A Factor 1 (p=.047), with a negative correlation with age in autistic adults (p=.018) but not in non-autistic adults (p=.821). This age-related decline in Factor 1 was replicated in the SPARK sample (p=.041). PSQ scores were associated with RBQ-2A Factor 1 (AZ p=.034), Factor 2 (AZ p=.011), and Total Score (AZ p=.004), with replication in SPARK (all p<.001) and were linked to slower right-hand finger tapping in the Arizona cohort (p=.050). Autistic adults had significantly larger left (p=.013) and right (p=.020) caudate volumes and a steeper age-related

decline in left caudate volume ($p=.044$). RBQ-2A Factor 1 scores were positively associated with left ($p=.011$) and right ($p=.022$) caudate volumes. These findings suggest that aging autistic adults experience elevated parkinsonism symptoms, behaviorally linked to repetitive motor behaviors. Age-related changes in caudate volume were associated with repetitive motor behaviors, suggesting structural contributions to their persistence. RBQ-2A and PSQ associations replicated across cohorts, underscoring repetitive motor behaviors as potential early behavioral markers of parkinsonism risk. Findings highlight the need for longitudinal research on motor aging in autism to inform early detection and intervention strategies.

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Poster

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Topic: A.06. Developmental Disorders

Support: R01MH132746

Title: Age-related declines in verbal fluency and brain compensation in autistic adults

Authors: *S. GALLEGOS, Y. ZHANG, M. VELEZ GALINDO, S. A. HARKER, A. SABRI, S. A. CORTES CORIA, F. JOHNSON, K. FELIX, B. BRADEN; Arizona State Univ., Tempe, AZ

Abstract: Background: Older autistic adults report social communication challenges as a major concern, yet aging research rarely investigates verbal fluency, the crucial word-generation skill for everyday needs. Our lab's previous findings show potential age-related vulnerabilities for cortical thinning in Broca's area, reduced fluency, and compensatory anterior cingulate cortex recruitment. We evaluated how language/communication behaviors, in conjunction with brain metrics, contributes to quality of life (QoL) outcomes across the autistic lifespan. **Methods:** Cognitive and MRI data were drawn from a longitudinal study of autistic adults ($n=75$) and matched neurotypical adults (NT; $n=88$), aged 40-71 ($n=163$) tested biennial for up to 12 years. All had IQs in the non-intellectual disability range with autism diagnoses verified using ADOS-2. Verbal fluency was assessed with the Controlled Oral Word Association Test (COWAT) and the Animal Naming Test, social communication with the Social Responsiveness Scale-2 (SRS-2), and QoL with the 36-Item Short Form Health Survey (SF-36). FreeSurfer derived cortical thickness from T1 MRI scans, and functional brain activation associated with language was assessed via a phonemic fluency task modeled after COWAT where participants silently generated words while pressing a button for each letter-cued word. **Results:** Group differences

were observed in all language, communication, and QoL metrics in autistic adults compared to NT adults (all $p < .05$ for COWAT, Animal Naming, SRS-2, and SF-36). Cross-sectional linear regression revealed a steeper negative slope between age and pars opercularis (Broca's area) cortical thickness in the autism group (interaction $p = .004$), but no significant age by group interactions for behavioral variables. Higher SRS-2 totals were linked to lower SF-36 Social QoL scores in midlife and older autistic adults ($p = .009$). Group ICA task-fMRI and latent variable structural equation modeling are underway to test whether fluency neural metrics mediate the SRS-2 and SF-36 link. **Conclusions:** We confirmed previous findings suggesting that autistic adults demonstrate persistent challenges in phonemic and semantic fluency, aligned with potential age-related vulnerabilities for accelerated cortical thinning, and present new findings that social communication abilities are directly linked to QoL in midlife and older autistic adults. Ongoing multimodal analyses will evaluate whether fluency-related neural mechanisms explain this association. These findings suggest supporting autistic adults social communication abilities across the lifespan may alleviate well documented poor QoL outcomes.

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Poster

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Program #/Poster #: PSTR256.22/A52

Topic: A.06. Developmental Disorders

Title: Regulation of autism-related self-injurious behaviour by electrical stimulation of nucleus accumbens circuitry in mice and humans

Authors: *K. ZHANG^{1,5}, J. GERMANN⁶, R. MATIN^{5,1}, K. MITHANI^{5,2}, J. ELLEGOOD³, C. GORODETSKY⁴, J. P. LERCH³, F. VENETUCCI GOUVEIA⁷, G. M. IBRAHIM^{8,5};

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Abstract: Self-injurious behaviours (SIB) are prevalent among children with Autism Spectrum Disorder (ASD) and can lead to permanent disability or death. Deep brain stimulation (DBS) of the nucleus accumbens (NAc) has recently been developed in a world-first, phase I clinical trial at SickKids Hospital as a potential treatment for affected children (ClinicalTrials.gov: NCT03982888). However, the neural underpinnings of NAc-DBS for SIB are poorly understood, and multimodal translational studies using animal models and clinical data are necessary to explore the mechanisms of disease and treatment. Through a unique translational framework, we integrated data from the BTBR $T^+ Itpr3^{fl/J}$ (BTBR) mouse model of SIB with longitudinal

neuroimaging and clinical outcomes from the aforementioned clinical trial of NAc-DBS in children with severe SIB to provide mechanistic insight to the neural basis of SIB in ASD. BTBR mice received active or sham DBS to the bilateral NAc, followed by behavioural tests evaluating ASD-related phenotypes (self-injurious, repetitive, anxiety-like, and social behaviours). Neurocircuitry influenced by NAc-DBS was assessed using high-resolution magnetic resonance imaging and deformation-based morphometry. We found that chronic, high-frequency NAc-DBS reduced injurious self-grooming and repetitive behaviour in BTBR mice through morphological alterations in fronto-limbic-striatal brain regions. In parallel, children with severe SIB undergoing NAc-DBS exhibited longitudinal structural changes in homologous fronto-limbic-striatal regions, including reduced volumes in the somatomotor and somatosensory cortices, anterior cingulate cortex, caudate, putamen, thalamus, and periaqueductal grey. These findings highlight conserved circuit-level plasticity associated with NAc modulation and provide foundational evidence to support NAc-DBS as a mechanistic therapy for affected children.

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Poster

PSTR256: Autism: Physiology and Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR256.23/A53

Topic: A.06. Developmental Disorders

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Italian Ministry of University and Research through the PRIN, Grant 20207S3NB8 (ACT2)

Title: Mistimed Brain Maturation in Toddlers with Autism Spectrum Disorder Revealed by Voxel-Level BrainAGE Analysis

Authors: E. TRIMARCO^{1,2}, L. ASTOLFI¹, S. LUBIAN LOPEZ^{2,3}, I. BENAVENTE FERNANDEZ^{2,3}, A. C. LINKE⁴, *I. FISHMAN⁴;

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Abstract: Early brain overgrowth, characterized by increased brain volume, cortical surface area, and structural connectivity, is one of the most consistent neuroimaging findings in autism spectrum disorder (ASD) during the first years of life. Additionally, atypical cortical thickness and white matter development trajectories suggest accelerated yet dysregulated or mistimed brain maturation, underscoring the need for advanced neuroimaging tools to better quantify these

early neurodevelopmental alterations. One promising approach is Brain Age Gap Estimation (BrainAGE), which uses machine learning models trained on typically developing (TD) populations to predict "brain age" from MRI data. The difference between predicted and chronological age serves as a proxy for atypical brain maturation. While BrainAGE has shown utility in studying neurodegenerative and psychiatric conditions in adults, and more recently in adolescents with neurodevelopmental disorders, its application to toddlers with ASD remains unexplored. In this study, we applied a voxel-level BrainAGE framework to assess deviations in brain maturation in a cohort of 1½-5 year old children enrolled in a study of early brain markers of autism (ASD: n=45, F/M: 11/34; mean age: 46 ± 13 months; TD: n=29; F/M: 13/16; mean age 46 ± 12 months), utilizing high-resolution T1-weighted MRI data acquired during natural nocturnal sleep (voxel size=0.8mm³, TE/TI=min full/1060ms). Using the VolBrain platform, T1-weighted images were preprocessed and segmented into 133 regions of interest (ROI), with BrainAGE calculated for each ROI and compared between ASD and TD groups using t-tests. After FDR correction, significantly greater BrainAGE (ASD >> TD) was found for the right temporal pole ($p = 0.027$), the left lateral orbital gyrus ($p = 0.028$), and the left temporal pole ($p = 0.035$). This work represents the first application of voxel-level BrainAGE analysis in toddlers with ASD and may offer a novel neuroimaging biomarker for early identification and tracking of atypical neurodevelopment in autism.

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Poster

PSTR256: Autism: Physiology and Systems

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Topic: A.06. Developmental Disorders

Support: Office of the Assistant Secretary of Defense for Health Affairs through the Autism Research Program under Award No. W81XWH-16-1-0321

Title: Functional Connectivity and Volume of the Amygdala as Predictors of the Response to Propranolol for Anxiety in Autism: A Pilot Study

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Abstract: Many autistic individuals experience higher rates of anxiety than both the general population and those with other neurodevelopmental disorders. Altered amygdala connectivity and atypical volume development in autism are linked to these anxiety symptoms which may be

important biomarkers for precision medicine efforts. Propranolol, a non-selective beta-adrenergic antagonist primarily used to treat hypertension, has shown promise in alleviating anxiety symptoms in individuals with autism. However, the response to propranolol is variable across autistic individuals. Thus, objective biomarkers capable of predicting treatment response are critically needed to identify which autistic individuals are most likely to benefit from propranolol. In this pilot study, we examined whether amygdala functional connectivity and volume are linked to anxiety improvement following propranolol treatment. Functional connectivity refers to the co-variation of brain signals between regions, measured via resting-state fMRI. Seventeen individuals with ASD (ages 15-21) were randomized to a 12-week double-blind trial of propranolol or placebo. Afterward, all participants received 12 weeks of open-label propranolol. Anxiety was assessed using the Clinical Global Impressions - Improvement (CGI-I) scale at baseline and after treatment. Resting-state fMRI scans were conducted at baseline. Participants who took propranolol for the blinded portion of the trial as well as those who took propranolol for the open-label portion were examined in this pilot study. Responders were defined as those minimally improved or better with propranolol, and non-responders were defined as those showing no change or worsening, based on CGI-I scores. Responders showed reduced left amygdala volume compared to non-responders, while no significant difference was observed in the right amygdala volume between the two groups. Furthermore, responders exhibited greater functional connectivity between the left amygdala and hippocampus compared to non-responders. In contrast, responders demonstrated reduced functional connectivity between the left amygdala and regions such as the cerebellum crus, frontal operculum, and left temporal pole, relative to non-responders. Although preliminary, the results of this pilot study suggest that neuroimaging biomarkers, including functional connectivity and amygdalar volume, may provide valuable predictive information for which autistic people may receive the most benefit for anxiety from propranolol. However, the results should be interpreted with caution until further randomized clinical trials with larger sample sizes are conducted.

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Poster

PSTR257: Animal Models of Developmental Disorders Other Than Autism II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR257.02/A55

Topic: A.06. Developmental Disorders

Title: Effects of mid-gestational prenatal cannabidiol exposure on hippocampal structure and behavioral outcomes in wistar rat offsprings.

Authors: *D. O. TAIWO-OLA¹, S. A. ISA³, R. O. IFARAJIMI⁴, J. A. ARIYO², O. A. BAMGBSE⁵, F. P. BAMIDELE², P. D. SHALLIE⁶;

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Abstract: Background: The most frequently used addictive substance during pregnancy is marijuana. Remarkably, 34% to 60% of individuals who use marijuana keep using it during pregnancy. Many women cite the belief that marijuana use is relatively safe during pregnancy, among other reasons for continuing use. Given the increasing prevalence of cannabis use among pregnant individuals, it is critical to examine how exposure at different stages of pregnancy impacts fetal development. This study evaluates the relationship between gestational cannabis intake and hippocampal development by systematically investigating the effects of cannabis exposure in mid-gestation. Methodology: This experimental study used 12 healthy pregnant Wistar rats divided into two (control and experimental) groups. Cannabidiol was administered orally at 150 mg/kg/day from gestational days 12-21 to the experimental group. Due to consistent cannibalism observed in the experimental group, foster parenting was adopted after birth. Key parameters assessed include neurobehavioral indices using the elevated plus maze; hippocampal tissues underwent histological and immunohistochemical analyses for GFAP, GLUT1, and gene expression of CNR1 and CAMK2G evaluated using qPCR. Result Cannibalistic behavior, involving maternal consumption of offspring, was observed in the Mid-CBD group but not in controls, suggesting CBD-induced maternal stress. In neurobehavioral tests, Mid-CBD rats spent significantly more time in closed arms (119.9s) and less in open arms (19.6s), with increased closed arm entries (3.9 vs. 1.6; $p < 0.01$). Histological analysis showed vacuolation in CA1, mild degeneration in CA3, and granule cell disorganization in the dentate gyrus. Significant increase in GFAP and GLUT1 expression were observed, indicating astrocytic activation and metabolic changes. CNR1 expression was significantly reduced, while CAMK2G levels remained unchanged. Discussion The mid-CBD group exhibited anxiety-like behavior and morphological changes, including vacuolation and uneven pyramidal cell distribution. While GFAP and GLUT1 levels remained stable, indicating minimal astrocytic activation, the significant decrease in CNR1 expression raises concerns about disrupted endocannabinoid signaling and potential long-term neurodevelopmental effects. Conclusion This study demonstrates that mid-CBD exposure results in increased anxiety-like behavior and significant morphological changes in the hippocampus, suggesting potential disruptions in neurodevelopment.

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Poster

PSTR257: Animal Models of Developmental Disorders Other Than Autism II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: A.06. Developmental Disorders

Support: Lo Kwee-Seong Biomedical Research Fund (J.I.)
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Title: A novel CDKL5 deficiency disorder mouse model: CDKL5 R550X Knock-in mouse model

Authors: *X. YANG^{1,2}, J. IP²;

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Abstract: Objective: CDKL5 deficiency disorder (CDD) is a neurodevelopmental disorder caused by mutations in the *CDKL5* gene, leading to early-onset epilepsy, motor deficits, cognitive impairments, and visual cortical damage. Over 250 mutations in *CDKL5* gene have been identified, including the nonsense point mutation CDKL5 R550X, which introduces a premature stop codon and truncates the protein. This study aimed to characterize the CDKL5 R550X mutation and its effects using a knock-in mouse model. **Methods:** We generated a CDKL5 R550X mutant knock-in mouse model and assessed its behavioral and neurobiological phenotypes. Behavioral tests were conducted to evaluate motor function, cognition, and autistic-like features. Additionally, *in vivo* two-photon imaging was employed to examine visual cortical defects. Further investigation focused on dendritic spine morphology in cortical visual neurons. **Results:** The CDKL5 R550X mouse model exhibited significant motor deficits, cognitive impairments, and autistic-like behaviors, recapitulating key features of CDD. Visual cortical defects were observed, consistent with clinical findings in patients. At the cellular level, cortical visual neurons showed reduced dendritic spine density and a lower proportion of mature spines, suggesting impaired synaptic connectivity. **Conclusions:** Our findings demonstrate that the CDKL5 R550X mutation leads to CDD-like phenotypes in mice, including neurodevelopmental and visual cortical impairments. The CDKL5 R550X knock-in mouse serves as a novel and relevant model for investigating the mechanisms underlying CDKL5 deficiency disorder.

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PSTR257: Animal Models of Developmental Disorders Other Than Autism II

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Topic: A.06. Developmental Disorders

Support: DHPS Foundation Grant
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Title: Lost in translation: how DHPS regulates neurodevelopment

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Abstract: Deoxyhypusine synthase (DHPS) deficiency is a rare monogenic disease characterized by biallelic variants in the *DHPS* gene. DHPS is an enzyme that functions in mRNA translation by catalyzing the post-translational modification, and therefore activation, of eukaryotic initiation factor 5A (eIF5A). The observed clinical outcomes associated with human mutations in *DHPS* include seizures, intellectual disability, and developmental delay. A previous mouse model of embryonic *Dhps* genetic deletion using the *Emx1-cre* resulted in agenesis of the cerebral cortex and hippocampus, leading to death by 4 weeks-of-age. Additional studies have shown that brain-specific deletion of *Dhps* caused a translation defect that impaired the synthesis of proteins involved in neurite outgrowth and synaptic signaling. Taken together, these data indicate that DHPS functions in the maintenance of eIF5A-mediated translation, which we hypothesize is needed to facilitate the elevated rate of protein synthesis during times of high translational burden such as neurodevelopment. Whereas these animal models clarified the outcome of total *Dhps* loss, the phenotypes are more severe than those observed in human DHPS deficiency—a disease not of total loss of DHPS abundance, but rather reduced enzyme activity. Thus, we have developed a mouse model that more accurately reflects the genetics of this disease. All individuals diagnosed with DHPS deficiency harbor a missense variant (*DHPS*^{MIS}) in *trans* with a functional null variant. Thus, we generated a *Dhps*^{MIS} knock-in allele and mated this animal with mice carrying the conditional *Dhps*^{LoxP/+} allele, *Emx1-cre* driver, and *R26R*^{TOMATO} reporter. We resultantly produced a mouse with deletion of one *Dhps* allele in the cortex and hippocampus beginning at embryonic day (E)9.5, and mutation in the other *Dhps* allele, which is the missense mutation common to all DHPS deficiency patients (*Dhps*^{MIS/LoxP}; *Emx1-cre*; *R26R*^{TOMATO}, denoted DHPS deficient mice). Preliminary data indicates that DHPS deficient mice do not have cortical agenesis as observed with complete genetic deletion of *Dhps*. Rather, we observe impaired cortical layer lamination, indicating that DHPS may be required to support proper migration and organization of cerebral cortex neurons. Ongoing experiments will determine how reduced DHPS function affects neuronal migration, differentiation, and arborization. These studies will decipher the regulatory role of eIF5A-mediated translation during neurodevelopment.

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Poster

PSTR257: Animal Models of Developmental Disorders Other Than Autism II

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Topic: A.06. Developmental Disorders

Title: Exploring the neurophysiologic and neuropathologic basis of visual deficits in a cystinosis mouse model

Authors: *H. CHANG¹, V. PRIFTI², A. SOLORZANO⁵, Y. DING⁵, J. FENG², K. PADMANABHAN², E. G. FREEDMAN³, K. H. WANG⁴, J. J. FOXE²;
²Neurosci., ³Del Monte Inst. for Neurosci., ⁴Dept. of Neurosci., ¹Univ. of Rochester, Rochester, NY; ⁵Neurosci., Univ. of Rochester Med. Ctr., Rochester, NY

Abstract: Cystinosis is a rare lysosomal storage disorder caused by mutations to the *Ctns* gene that result in a defective lysosomal transport protein called cystinosin. Cystinosin is responsible for the export of cystine - the oxidized form of cysteine - from the lysosome to the cytosol. Without a functioning transporter, cystine crystals accumulate in lysosomes and cause cell death. Typically, cystinosis presents early in childhood with kidney dysfunction that progresses to end-stage renal disease and early death. Certain medications can extend patient life span, but the disease will continue to progress over time, incorporating other body systems. Photophobia, or a painful sensation in response to light, is one of the most common and debilitating symptoms that patients experience. Historically, photophobia has been attributed to corneal cystine crystal accumulation in patients, as crystals are easily found within the eyes by just 16 months of age. Consistent usage of topical eye drops to remove these crystals, though, does not alleviate symptoms, suggesting a more complex etiology of patient photophobia. Interestingly, visually evoked potential (VEP) data collected in patients demonstrates an early cortical hyperexcitability, hinting that there may be a role of the central nervous system (CNS) in patient photophobia.

To understand whether the CNS plays a role in the etiology of photophobia in cystinosis, we have developed a *Ctns* knockout (*Ctns* ^{-/-}) mouse model using CRISPR/Cas9. This model introduces a frameshift mutation and premature stop codon early within the locus, producing a truncated 23 amino acid (AA) protein that is 344 AA shorter than the full-length CTNS. Early phenotyping with optical coherence tomography imaging, has confirmed that our *Ctns* ^{-/-} mice accumulate corneal crystals like that of patients. To further examine if our mouse model experiences the same hyperexcitability as patients with cystinosis, we have designed a VEP paradigm that presents a flash, grating stimulus to mice at varying intensities and contrast levels. In conjunction, the molecular basis of this pathology will be studied using sectioning and staining of areas in the visual cortex and visual thalamus for evidence of lysosomal dysfunction and loss of inhibitory interneuron signaling. Finally, using a light-dark box behavioral paradigm we will assess for whether our mice exhibit photophobic responses in response to varying intensities of light. In-depth neurophysiologic and neuropathologic characterization of this mouse model will facilitate future applications to examine the efficacy of potential gene therapies and medications for cystinosis.

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Research grant from the University of Pennsylvania Orphan Disease Center on behalf of the Loulou Foundation (J.I.)

Title: Altered granule dynamics in CDKL5 deficiency during development

Authors: *Z. ZHENG^{1,2}, Y. ZHU², S. YUAN², Y. CHAI², J. IP³;

¹The Chinese Univ. of Hong Kong, tai po, nt, Hong Kong; ³Sch. of Biomed. Sci., ²The Chinese Univ. of Hong Kong, Hong Kong, China

Abstract: CDKL5 deficiency disorder (CDD) is an X-linked neurodevelopmental disorder causing by loss of function of a serine/threonine kinase, Cyclin-Dependent-Kinase-Like 5 (CDKL5). CDD patients suffer from severe neurodevelopment defects, and the underlying pathological mechanism is not clear. In previous studies we identified nELAVL as novel substrates of CDKL5. Lack of phosphorylation by CDKL5 leads to altered nELAVL phase separation in rodent models. Induced pluripotent stem cells (iPSCs) derived neuron is a promising human-based model to study neurodevelopment disorders. We used CDD patient-derived neuron model and established an isogenic control by Adenine based editing (ABE). We found increased nELAVL granule formation and increased colocalization between nELAVL granules and P-bodies in CDD neurons. Our findings suggested possible altered granule interaction in CDD neurons, which might lead to pathological outcomes. Investigating the mechanism underlying the interaction between nELAVL and P-bodies would be an intriguing focus for future studies.

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Support: DISC-2 13417
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Title: Cortical Hyperexcitability, Sleep Deficits, and Abnormal Respiratory Physiology as Translational Biomarkers in a Female Mouse Model of CDKL5 Deficiency Disorder

Authors: *O. HAOUCHINE¹, J. L. SILVERMAN²;

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Abstract: CDKL5 Deficiency Disorder (CDD) is a rare X-linked neurodevelopmental disorder (NDD) primarily affecting females at a ratio of 4:1. Notably, female preclinical mouse models offer a novel and translationally relevant approach, given that pathogenic variants in CDKL5 are often lethal in males during infancy. Due to X-linked mosaicism, symptom severity of female patients is highly variable and uniquely captured in female mouse models, enhancing their translational value. Loss of function mutations in CDKL5 cause the disruption of neurodevelopmental processes like neurogenesis and synapse formation, informing the clinical presentation of CDD of severe epileptic encephalopathy accompanied by developmental delay, treatment-resistant epilepsy, sleep disturbances, and respiratory dysfunction including obstructive sleep apnea (OSA), as well as features common to most NDDs such as intellectual disability, motor impairment, delayed or absent speech and/or autism spectrum disorders. There is no effective standard of care, inadequate symptom management, and a lack targeted treatments being, presenting a critically unmet medical need. Our lab, and others, are utilizing clinically relevant rodent models to comprehensively characterize and identify phenotypes and/or biomarkers necessary for therapeutics to be assessed in clinical trials. Using multiple measures and intersecting analysis of sleep architecture, respiratory function, and electrophysiological signatures, we have identified and detailed robust and rigorous phenotypes in this exon 6 deletion model of CDD. The current study expands upon our previously published characterization work using a cohort of female mice implanted with wireless telemeters measuring both EMG and surface EEG activity. Respiratory physiology in these implanted females was acquired over the course of 4 hours in whole body plethysmography chambers and analysis was integrated with simultaneously collected EEG/EMG activity. Mutant females exhibited elevated baseline spike rates, prolonged spike trains, increased spectral power, reduced sleep spindles, and frequent apneas coinciding with EMG bursts. The linking of WBP and simultaneously collected EEG is important because it shows this model can recapitulate OSA phenotypes. This approach enables us to parse primary effects of CDD from secondary consequences of sleep disruption, strengthening the model's utility for therapeutic screening. The novel utilization of multiple technical approaches with intersecting analysis introduces a unique assay that has enabled us to report data that is the first of its kind amongst research using rodent models of NDDs.

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Poster

PSTR257: Animal Models of Developmental Disorders Other Than Autism II

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Topic: A.06. Developmental Disorders

Support: Hartwell fellowship, UC Davis MIND Institute and Department of Psychiatry and Behavioral Sciences.
The Orphan Disease Center, Million Dollar Bike Ride, University of Pennsylvania School of Medicine.

Title: Lifespan impact of early-life seizures in a novel mouse model of KCNT1-related epilepsy

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Abstract: Excitatory/inhibitory balance is critical for proper neuronal circuit formation and function wherein disruptions can lead to epilepsy. KCNT1 is a sodium-gated potassium channel essential for neuroexcitability with at least 64 human variants; many of which are gain-of-function (GOF). KCNT1-related epilepsy is a rare genetic neurodevelopmental disorder and severe developmental epileptic encephalopathy often diagnosed within the first 6 months of life. Employing a multi-disciplinary collaboration, we generated a novel mouse model C57BL/6J - *Kcnt1*^{em1Bryd}, with a knock-in mutation (G269S) corresponding to the human G288S variant. Our previous data demonstrates heightened network bursting and hypersynchronous activity in primary cortical neurons from *Kcnt1*^{+/G269S}, and *Kcnt1*^{G269S/G269S} *in vitro*. Additionally, *Kcnt1*^{+/G269S} mice were susceptible to early life (PND 21-23) hyperthermia-induced (fever/febrile) seizures compared to littermate controls. We hypothesize that *Kcnt1*^{+/G269S} mice with early-life seizure experience will better replicate the clinical presentation including changes in motor ability, learning and memory impairment, apneas, sleep disturbance, and increased spiking in EEG. We found that *Kcnt1*^{+/G269S} mice with prior seizure experience were more hyperactive in the open field and had impaired learning and memory. Additionally, early life seizures lead to increased erratic breathing, sleep disturbances, and increased spiking in EEG. Taken together, *Kcnt1*^{+/G269S} has construct and face validity as a model of KCNT1-related epilepsy and future studies are required to develop and evaluate novel therapeutics.
Funding: Hartwell fellowship, UC Davis MIND Institute and Department of Psychiatry and Behavioral Sciences. The Orphan Disease Center, Million Dollar Bike Ride, University of Pennsylvania School of Medicine.

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PSTR257: Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: PSTR257.09/A62

Topic: A.06. Developmental Disorders

Support: Hartwell fellowship, UC Davis MIND Institute and Department of Psychiatry and Behavioral Sciences.
The Orphan Disease Center, Million Dollar Bike Ride, University of Pennsylvania School of Medicine.

Title: Investigating Neuroanatomical, neurophysiological and epileptogenesis in a novel mouse model of KCNT1-Related Epilepsy

Authors: *S. BERG¹, S. L. OLGUIN¹, R. BEN-SHALOM², J. L. SILVERMAN³;
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Abstract: KCNT1-related epilepsy results from pathogenic variants in KCNT1 (potassium (K⁺) channel subfamily T-member-1), a sodium (Na⁺)-activated K⁺ channel, with at least 64 *de novo* pathogenic variants, causing a loss or gain of function. Depending on the site of the abnormal variant, there are heterogenous presentations that range in symptom severity, however all have severe treatment resistant epilepsy and cause a rare genetic neurodevelopmental disorder, with comorbidities in wide variety of clinical modalities. One of the most common clinical variants is *KCNT1* G288S (G269S in mice) and it is located in at the channel pore. Here, we investigated the C57BL/6J-*Kcnt1*^{em1Bryd} G269S knock-in mouse model using a multi-disciplinary approach to assess *in vitro* neuronal electrophysiology and local network activity concomitant with the *in vivo* formation of epileptogenesis and neuroanatomy by magnetic resonance imaging (MRI) using *Kcnt1*^{+/+} (WT) and *Kcnt1*^{+/G269S} mice. In our initial *in vitro* assessment, we examined WT, *Kcnt1*^{+/G269S}, *Kcnt1*^{G269S/G269S} and confirmed hyperexcitability and increased neuronal bursting, with excitotoxic death in homozygous neurons. These data were collected from HD-MEA chips recorded twice weekly from DIV 7 to DIV 28, to track the emergence and progression of network-level dysfunction. In our initial *in vivo* assessment, we observed no differences in developmental milestones in pups. Though adult *Kcnt1*^{+/G269S} illustrated poor motor coordination and exploratory anomalies. This motor phenotype is potentially explained by the volumetric differences obtained via *ex vivo* MRI analysis where the cerebellar nuclei were significantly smaller in HET compared to WT mice. Additionally, *Kcnt1*^{+/G269S} mice (PND 21-23) exhibited more intense fever-induced seizures at lower temperature thresholds, which aligns well with the clinical presentation of KCNT1-related epilepsy. Upon subsequent re-exposure to artificially induced seizures via the chemo-convulsant pentylenetetrazol and/or electroconvulsive-induced seizures via ocular electrode stimulation, *Kcnt1*^{+/G269S} mice were more profoundly affected than their age/sex matched WT littermates. Taken together, *Kcnt1*^{+/G269S} have elevated bursting and susceptibility to seizures increasing the likelihood of epileptogenic processes, compared to age/sex matched WT littermates. Future work will involve time synchronous collection of *in vivo* neurophysiology required to further understand the relationship between altered network activity and the behavioral presentation and to identify outcomes measures for therapeutic assessments.

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Poster

PSTR257: Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: PSTR257.10/B1

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Support: flash talk award

Title: Spatial distribution of sphingomyelin species dictates severe neurological pathologies in CerTra syndrome

Authors: *I. KAYSUDU;
EPFL, Lausanne, Switzerland

Abstract: Sphingolipids are essential in the central nervous system. Sphingolipids constitute 10-20% of the brain lipid composition. They sustain membrane stability, regulate neurogenesis and myelin formation. Any disturbance in sphingolipid metabolism has tremendous effects on the brain structure and function, thus the sphingolipid pathway needs to be strictly regulated. Ceramide transporter protein (CERT), which carries ceramides from the ER to the trans-golgi for further sphingomyelin synthesis, has a key role to maintain the sphingolipid homeostasis. It was reported that *de novo* heterozygous mutations in CERT1 gene, which lead its excessive activity, cause a severe sphingolipid-related neurodevelopmental disorder in newborns, called “CerTra Syndrome”. Among other symptoms, these patients primarily have intellectual disability, global developmental delay, sleep disturbances, ventriculomegaly and delayed or hypomyelination. However, it remains elusive how hyperactive CERT causes distinct neurological abnormalities in patients. Additionally, the neurotoxic sphingolipid species associated with the disease and their spatial distribution in the brain need to be elucidated. In this study, we demonstrated how constitutively active CERT affects the brain lipid composition and distribution by utilizing the MALDI-MSI, thereby, how it causes neurological pathologies. We generated a heterozygous CERT-S132L knock-in mouse model to characterize the disease in terms of physiology, locomotion, cognition and biochemistry. We showed that the CERT-S132L variant led to sleep disturbance, increased neuronal and physical activity. We also showed that these phenotypes in mice are in parallel with the symptoms in the patients. Biochemical findings obtained by integrative omics studies suggest that these traits are related to the spatial discrepancy of the sphingomyelin species in the ventricles and the white matter of the mouse brain. We hypothesize that increased and exclusively localized sphingomyelin species in the diseased brain should be the neurotoxic lipids to lead to ventriculomegaly and hypomyelination in CerTra patients. Overall, the discovery of neurotoxic sphingomyelin species in the CerTra disease and how they deregulate the brain function could promote therapeutic opportunities to the patients and possibly other neurodevelopmental or neurodegenerative diseases which might be affected by sphingolipid disturbances.

Disclosures: I. Kaysudu: None.

Poster

PSTR257: Animal Models of Developmental Disorders Other Than Autism II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR257.11/B2

Topic: A.07. Development of Neural Systems

Support: NIH Grant R01HD094809
NIH Grant R21HD106043
NHLBI T32 Hematology (PI: Greg Vercelotti)
University of Minnesota Masonic Institute for the Developing Brain Seed Grant
University of Minnesota Masonic Institute for the Developing Brain
Predoctoral Fellowship in Developmental Science

Title: Sex-dependent responses of hippocampal mitochondrial respiration to early-life iron deficiency

Authors: *J. BAILARD, T. R. MONKO, T. W. BASTIAN;
Dept. of Pediatrics, Neonatology Div., Univ. of Minnesota, Twin Cities, Minneapolis, MN

Abstract: Iron deficiency (ID) is the most common nutrient deficiency worldwide, affecting an estimated 1.9 billion people and 40-50% of pregnant women, fetuses, newborns, and toddlers. Early-life ID impairs brain development and cognitive function in children, reducing learning rates and processing speeds, while adolescents and adults who were iron-deficient in fetal-neonatal development have higher incidence of several cognitive and psychiatric conditions (anxiety, depression, ADHD, schizophrenia). Persistent neurobehavioral changes after early-life ID may arise from a metabolic reprogramming of the developing iron-deficient brain. We hypothesize that early-life ID induces acute and permanent sex-specific disruptions to hippocampal energy metabolism by persistently altering mitochondrial function. To test this, pregnant rats were given an iron-deficient diet beginning at gestational day 2 and switched to an iron-sufficient diet at postnatal day 7 (P7). Hippocampal mitochondria collected at P14 from both synaptic and non-synaptic compartments show trends of reduced respiration and ATP production rates in ID offspring, with greater decreases among females seeming to drive this difference. At P21, once the ID offspring's hematology measures have recovered from the anemia, the mitochondria's ongoing functional responses to early-life ID appear to depend on their subcellular location. Synapse-localized mitochondria from ID male hippocampus show trends of higher oxidative phosphorylation rates and more efficient ATP production, while non-synaptic mitochondria from glial and neuronal somas of ID female hippocampus have trending decreases in these measures. These data suggest that sex-dependent alterations to hippocampal mitochondrial respiration from early-life ID may arise during early postnatal brain development and persist even after iron treatment resolves the anemia. Furthermore, sex-dependent patterns of altered mitochondrial regulation in the hippocampus may differentially affect respiratory function among cell compartments that have unique energetic demands. Our preliminary findings are consistent with fetal-neonatal iron deficiency inducing a sex-dependent reprogramming of hippocampal mitochondrial function to alter the hippocampus's metabolic homeostasis. Ongoing

metabolic differences may contribute to the sex-dependent presentations of cognitive and neurobehavioral changes observed in adolescents and adults after early-life iron deficiency.

Disclosures: J. Bailard: None. T.R. Monko: None. T.W. Bastian: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.01/Web Only

Topic: A.09. Development and Evolution

Support: Mercy University, Faculty Development Grant

Title: Evolutionary constraints acting on classical cadherins in primates are associated with their expression in the central nervous system (CNS)

Authors: F. REYES VIGIL, M. CAMPO, *J. L. BRUSES;
Mercy Univ., Dobbs Ferry, NY

Abstract: Classical cadherins (CDHs) comprise a family of 18 single-pass transmembrane cell adhesion molecules composed of an ectodomain (ED) with five homologous cadherin domains (EC1 to EC5) and a highly conserved cytoplasmic domain (CD) with catenin binding sites. CDHs are divided into type I and type II based on the folding of EC1, which determines their homo and heterophilic binding specificity. CDHs are highly expressed in neurons, and the expression of distinct repertoires at precise stages of CNS development regulates cell migration, sorting, and the formation of synaptic contacts between specific partners. CDHs are exclusively found in metazoans, suggesting that their emergence contributed to the formation of architecturally complex tissues. To evaluate the extent of selective evolutionary pressure exerted on CDHs, this study examined the rate of amino acid and nucleotide substitution between *H. sapiens* CDHs and their orthologs in fifteen extant species of primates, including strepsirrhines, tarsiers, New and Old World monkeys, and apes (covering ~57 million years of evolution), to investigate whether differences in evolutionary constraints are associated with gene locus, essentiality, and expression. Pairwise comparisons of both amino acid substitutions and the ratio of non-synonymous substitutions per non-synonymous site (dN) to synonymous substitutions per synonymous site (dS) (dN/dS), show that CDH2, CDH4, and most type II CDHs have been under significantly higher negative selection compared to CDH1, CDH3, CDH5, and CDH19, which are primarily expressed outside the CNS. Analysis of amino acid substitutions within each EC reveals that CDH1 accumulated a higher number of substitutions including in EC1. The EC5 shows the greatest number of substitutions in most CDHs except CDH2, in which strong negative selective pressure is observed, suggesting an important role in neuronal function. Spearman's correlation analysis shows a positive correlation between CDHs expression levels (E) in mouse and human tissues and their rate of evolution (R), as is observed in most cell

surface proteins. However, CDHs expressed in the CNS show a negative E-R correlation, indicating that the negative selection exerted on CDH2, CDH4, and most type II CDHs is associated with their expression in the CNS. Our findings suggest that the strong negative selective pressure exerted on CDHs expressed in the CNS is due to their role in CNS formation and function and has contributed to the development of complex neuronal networks that shaped the evolution of the CNS in primates by regulating the specificity and properties of cell-cell interactions and synaptic transmission.

Disclosures: F. Reyes Vigil: None. M. Campo: None. J.L. Bruses: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.02/B3

Topic: A.09. Development and Evolution

Support: NSF IOS 2227963
NIH R01NS133654

Title: Transcriptomic analysis of molluscan chemosensory transduction genes suggests use of novel G proteins and ionotropic receptors

Authors: *J. L. PÉREZ-MORENO, C. LAWLESS, T. THATTACHERRY, E. JOHNSON, P. S. KATZ;

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Abstract: Vertebrate and insect chemoreception rely on distinct types of chemoreceptive molecules; vertebrates use G protein-coupled receptors (GPCRs), whereas insects use ionotropic receptors. The molecular transduction mechanisms for molluscs are not well-established and could shed light on the evolution of chemosensory systems in Bilateria. Our investigation centered on the nudibranch mollusc, *Berghia stephanieae*. We performed a transcriptomic analysis of genes involved in chemoreception focusing on 1) G proteins and 2) a recently described chemotactile receptor (CR). Our transcriptomic analysis of *Berghia*'s nervous system (brain, rhinophore ganglia) and sensory organs (rhinophores, oral tentacles) identified multiple $G\alpha$ subunit paralogs, including candidates orthologous to $G\alpha_s$, $G\alpha_{olf}$, and $G\alpha_q$. Large-scale phylogenetic analysis, using a custom molluscan transcriptome database, revealed one *Berghia* $G\alpha$ gene clustering with *Drosophila* $G\alpha_s$. This gene exhibited significantly higher expression in transcriptomes obtained from the rhinophores, which are the main olfactory organs. This suggests that $G\alpha_s$ may play a primary role in chemosensory transduction, representing a potential divergence from the canonical $G\alpha_{olf}$ -dependent signaling in vertebrate olfaction. Secondly, we explored CR expression. These ligand-gated ion channels, which mediate "taste by touch", were thought to be unique to cephalopods. Our transcriptomic analyses revealed 16 candidate CR-like

genes in *Berghia*. Phylogenetic analysis positioned these as a gastropod-specific sister group to known cephalopod CRs, indicating that CR-like receptors likely diversified before the evolutionary split between gastropods and cephalopods. This discovery suggests these receptors may have potential mechanosensory, chemosensory, or multimodal roles in non-cephalopod molluscs, broadening the functional and evolutionary landscape of CRs within Mollusca. Ongoing qPCR validation and HCR-FISH spatial mapping will further explore the expression specificity and distribution in sensory neurons of G α subunits and CR-like genes. Collectively, these findings underscore *Berghia*'s utility for identifying and characterizing novel chemosensory mechanisms, revealing greater evolutionary diversity in these pathways than previously recognized.

Disclosures: J.L. Pérez-Moreno: None. C. Lawless: None. T. Thattacherry: None. E. Johnson: None. P.S. Katz: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.03/B4

Topic: A.09. Development and Evolution

Support: NIMH ZIA MH002951-09

Title: A comparative transcriptomic analysis of the ventral midline thalamus between the Rhesus Macaque, Common Marmoset, and Laboratory Rat

Authors: *C. HUME^{1,2}, D. MATROV², Y. CHUDASAMA²;

¹Natl. Inst. of Mental Hlth., Rockville, MD; ²Section on Behavioral Neurosci., NIH, Bethesda, MD

Abstract: Previous efforts to define the molecular composition of thalamic nuclei describe a relatively conserved cellular heterogeneity between rodent and non-human primate models (Roy et al., 2022, *Nat Neurosci.* 25, 140-153). However, the specific comparison between non-human primates and rodent models in the ventral midline thalamus has not been reported. The ventral midline thalamus includes the reuniens and rhomboid nuclei and serves as important relay structures connecting the prefrontal cortex and the hippocampus. As such, they form a major part of the limbic thalamus and are involved in cognitive functions such as working memory, attention, and decision making that are often altered in psychiatric conditions. A focused investigation into the cellular composition of this area can enable a better understanding of the structure and function of this region for identification of potential therapeutic targets. Moreover, comparing the genetic conservation in brain structures between common mammalian models provides information about the evolution of this brain area. We collected brain tissue for a comparative single-nuclei transcriptomic analysis across three commonly used animal models:

the rhesus macaque (*Macaca mulatta*, n = 3), the common marmoset (*Callithrix jacchus*, n=3), and the Long-Evans rat (*Rattus norvegicus*, n=9). Brains were perfused, flash frozen, and the ventral midline thalamus was extracted under histological guidance. A minimum of 30 mg of tissue was collected per sample and the nuclei isolated and fixed. Split-pool combinatorial barcoding, library preparation, and sequencing were conducted according to the *Parse Biosciences Evercode*® v3 protocol using the *Evercode*® WT v3 kit. Gene expression analysis by cell count matrices will include cell-type identification and annotation, gene-ontology conversion, cross-species cell-type integration, and comparative differential gene expression. The knowledge from this study will help determine appropriate models for translational studies and contribute to the understanding of the organization and involvement of this important region in cognitive functioning.

Disclosures: C. Hume: None. D. Matrov: None. Y. Chudasama: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.04/B5

Topic: A.09. Development and Evolution

Support: JSPS KAKENHI JP17K07083

Title: Changes in ubiquitination of microtubule-cleaving enzyme katanin during vertebrate brain evolution

Authors: T. SHIGEMURA, S. SUZUKI, T. OYAMA, K. KOIKE, *K. HAYASHI;
Sophia Univ., Tokyo, Japan

Abstract: The reorganization of microtubules is required for axon branching and dendrite growth. Microtubule-severing enzymes rearrange the microtubule array or generate short microtubules which serve as new microtubule seeds for elongation. Katanin, is one of the microtubule-severing enzymes, and its catalytic subunit Katanin A has two isoforms - KATNA1 and KATNAL1. We have previously shown that KATNAL1 is strongly expressed in neurons and testis, has higher activity than KATNA1, and that KATNA1 undergoes intracellular degradation, while KATNAL1 is less prone to degradation (Hatakeyama and Hayashi 2018). In this study, we investigated the expression of KATNAL1 in mice in detail and examined the ubiquitination of katanin isoforms. RT-qPCR results showed that the ratio of KATNAL1 to KATNA1 expression in adult mice was predominantly higher in the brain than in other tissues. KATNAL1 expression in developing cerebral cortex is highest at around the time of birth when dendrites are actively growing. Immunostaining revealed that KATNAL1 is localized in the cell bodies of cerebral neurons. These suggest that KATNAL1 plays a significant role in brain development. We then tested if the difference in intracellular degradation between katanin

isoforms is due to differences in ubiquitination. FLAG-tagged mouse katanin cDNA and HA-tagged ubiquitin expression vector were co-transfected into HEK293T cells, and the proteins were immunoprecipitated with FLAG antibody, then followed by Western blotting with HA antibody. The results indicated that mouse KATNA1 was strongly ubiquitinated as has been previously reported, whereas mouse KATNAL1 was hardly ubiquitinated. Because KATNA1 gene and KATNAL1 gene were generated by chromosomal duplication at the early stage of vertebrate evolution, we investigated the ubiquitination of fish katanin isoforms. We cloned medaka katanins' cDNA and performed the same experiment and found that medaka KATNA1 and medaka KATNAL1 had almost the same levels of ubiquitination. This finding suggests that KATNAL1 underwent molecular changes during the evolutionary path from fish to mammals to ensure that it is not ubiquitinated, and to have stronger microtubule-severing activity. Thus, the molecular evolution of KATNAL1 may have contributed to the higher complexity of neuronal processes in mammalian brain.

Disclosures: T. Shigemura: None. S. Suzuki: None. T. Oyama: None. K. Koike: None. K. Hayashi: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.05/B6

Topic: A.09. Development and Evolution

Support: NIH Grant NS127051

Title: Spatiotemporal Profiling Neurodevelopment in the Fetal Göttingen Minipig

Authors: *J. D. PRASAD^{1,2}, Z. LI¹, A. FURUTA^{1,2}, S. HENMI², I. ANTONIOS², T. F. HAYDAR¹, N. ISHIBASHI^{2,1};

¹Anat. and Neurobio., Boston Univ., Boston, MA; ²Ctr. for Neurosci. Res., Children's Natl. Med. Ctr., Washington, DC

Abstract: The fetal period marks a critical window for establishing the structural and cellular foundation of the human brain, during which a series of intricately orchestrated events drive neocortical expansion and the formation of a highly complex, gyrencephalic structure. This cytoarchitecture arises from neural precursor cells (NPCs) in the ventricular-subventricular zone (VZ/SVZ), which generate all neurons and glia required for the precise formation of brain networks and the successful transition to life outside the womb. While this general plan for neurogenesis is conserved across mammalian species, recent studies suggest that specific NPC subtypes may be unique to gyrencephalic brain development (i.e. not present in lissecephalic brains). However, the limited number of prenatal animal models is the biggest rate-limiting step in advancing our understanding of these processes. Therefore, establishing a clinically relevant

gyrencephalic animal model that can be studied using modern genetic and cellular imaging techniques would improve our understanding of the cellular/molecular events underlying the mechanisms of human fetal brain development.

We propose the Göttingen Minipig as a preclinical model for studying fetal brain development due to the highly evolved gyrencephalic neocortex, comparative physiology, and cytoarchitecture. Fetal Göttingen minipig brains (5.5-13.5 gestation weeks [GW]; both sexes) were used to establish a time-course of neurogenesis and gyrencephaly. In utero electroporation (IUE) was performed in a subset of experiments to label cells in the VZ.

The minipig brain undergoes profound structural changes from 5.5-13.5GW, transitioning from a lissencephalic structure to a uniquely complex gyrencephalic structure. This period is marked by increased cortical thickness, reflecting neocortical expansion. PH3 analysis showed developmental shifts in proliferative activity from the VZ to SVZ, and pVIM analysis showed further displacement into the intermediate zone, subplate and cortical plate. This was accompanied by a shift from symmetric to asymmetric cell division. The emergence of cellular markers defining key stages of human brain development (i.e. microglia, radial glia and oligodendrocyte lineage cells) was also characterized. Finally, while intraventricular injections were technically challenging between 5.5-6.5GW, we successfully established and validated fetal IUEs between 7-8.5GW. Overall, our study indicates that the fetal Göttingen minipig brain shares many key features with human fetal brain development, making it a valuable model for studying the complexities of gyrencephalic brain development.

Disclosures: J.D. Prasad: None. Z. Li: None. A. Furuta: None. S. Henmi: None. I. Antonios: None. T.F. Haydar: None. N. Ishibashi: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.06/B7

Topic: A.09. Development and Evolution

Title: Comparative study of the roles of key autophagy genes in the developing brain

Authors: *G. ANGST¹, E. WALTER², G. HODSON¹, C. WANG¹;

¹OSUCCC-James Cancer Hosp. and Solove Res. Institute, Dept. of Radiation Oncology, The Ohio State Univ. Wexner Med. Ctr., Columbus, OH; ²Cancer Biol., Univ. of Cincinnati Col. of Med., Cincinnati, OH

Abstract: Autophagy is a highly conserved cellular process involving the packaging of cytoplasmic contents and organelles into double membrane structures and the subsequent delivery to lysosomes for degradation. Autophagy is broken down into different steps, including autophagy induction, phagophore nucleation, autophagosome elongation and closure, autophagosome-lysosome fusion, cargo degradation, and materials recycling. Autophagy

dysfunction is connected to infection, cancer, and neurodegenerative diseases. In this study, we examined the functions of autophagy genes of Fip200, Atg14, and Atg16l1 in postnatal brain development using a brain specific conditional knock out (cKO) system. As expected, we observed the accumulation of p62 puncta in the cortex and the granule layer of the hippocampus of the cKO mice but not in control mice. We noticed spongiform degeneration in several brain regions of Fip200 cKO mice and Atg14 cKO mice, but not in Atg16l1 cKO mice at eight weeks old. Similarly, our data revealed a significantly decreased number of NeuN+ neurons and Bcl11b+ neurons in Fip200 and Atg14 cKO mice. When the cortices of neonatal Fip200 cKO mice were observed, the number of Bcl11b+ neurons was similar to that in control mice, suggesting postnatal neuronal death in the mutant mice. In conclusion, we observed neurodegeneration in the young adult Fip200 and Atg14 cKO mice but not in Atg16l1 cKO mice, indicating distinct roles of autophagy genes involved in different steps in brain development.

Disclosures: G. Angst: None. E. Walter: None. G. Hodson: None. C. Wang: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.07/B8

Topic: A.09. Development and Evolution

Support: NSF 21-608

Title: Uncovering human-specific developmental signaling pathway responses in great ape forebrain neural progenitors

Authors: *R. C. MCMULLEN;
Neurol., Univ. of California San Francisco, San Francisco, CA

Abstract: Humans have a threefold enlargement of the brain compared to other great apes. Brain expansion (especially of the cortex) is thought to underly human-specific cognitive traits. One theory of cortical expansion implicates genetic changes affecting the proliferation or differentiation of the neuroepithelial progenitor (NEP) cells present in embryonic development. Genetic differences between humans and chimpanzees are mostly found in non-coding regions, and while several studies have described gene expression divergence between primates during development, isolating functional differences remains challenging. As potential regulatory sequences, these genetic changes may affect gene regulatory networks only in specific contexts, such as developmental stage, cell type, and stimulus. In some recent work, functional gene expression changes associated with genetic variants were unmasked only in response to physiological stimuli, and further, response genes had more phenotypic relevance than genes varying at baseline. We reasoned that by stimulating NEPs with physiologically relevant developmental signals, we would uncover species differences in stimulus response. We

generated putative cortical NEPs from pluripotent stem cells from dozens of individuals across human, chimpanzee, and orangutan species and treated them with 21 activators or inhibitors of 9 developmental signaling pathways, collecting single cell RNA-sequencing data at 3 time points. We found that molecular and cellular responses to developmental signals were highly conserved across species. Conserved response genes (rGenes) but not other rGenes or non-rGenes were enriched for loss of function (LOF)-intolerant genes and for diseases affecting proliferation. We identified approximately 2,400 genes with species divergence in expression, 79% of which were observed only in a stimulus response context. In response to FGF ligands, Human NEPs were more reactive to maintenance of progenitor fate, whereas in response to a NOTCH pathway inhibitor, Human NEPs were more refractory to neurogenesis. Species divergent rGenes were also enriched for LOF-intolerant genes, and in response to BMP7 ligand, were enriched for genes associated with Schizophrenia and Bipolar Disorder. Given the overlap of affected traits and associated genes, evolutionary changes in human brain development are hypothesized to underly human susceptibility to Schizophrenia. Intriguingly, we found that several divergent BMP7 rGenes overlap with loci associated with both human evolution and Schizophrenia, suggesting that BMP7 stimulus response in NEPs may be involved in the disorder's etiology.

Disclosures: R.C. McMullen: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.08/B9

Topic: A.09. Development and Evolution

Support: NSF CRCNS #2112862

Title: Serotonergic axons in the Pacific angelshark brain: From neuroanatomy to predictive stochastic modeling with supercomputing

Authors: *S. JANUSONIS¹, R. METZLER², T. VOJTA³;

¹Psychological and Brain Sci., Univ. of California Santa Barbara, Santa Barbara, CA; ²Inst. of Physics and Astronomy, Univ. of Potsdam, Potsdam-Golm, Germany; ³Dept. of Physics, Missouri Univ. of Sci. and Technol., Rolla, MO

Abstract: Serotonergic axons (fibers) are present in all studied vertebrate brains, where they form dense meshworks in many neuroanatomical regions. These meshworks are typically quantified with density measurements in descriptive studies. Serotonergic axons appear to fundamentally support neuroplasticity, but an in-depth understanding of their function is lacking (they also have no analogs in artificial neural networks). Their structural self-organization is also poorly understood, partly because of the strong stochasticity of individual fiber trajectories. In order to gain insights into the fundamental features of this system, it is instructive to examine

vertebrate clades that are separated by large phylogenetic distances and differ strongly in their physical geometry and differentiation of brain tissue. In this study, serotonergic fibers were investigated in the brain of the Pacific angelshark (*Squatina californica*), a representative of a unique (ray-like) lineage of the squalomorph sharks that dates back to the Late Jurassic. First, the fundamental cytoarchitecture of the angelshark brain was examined for the first time, including the expression of ionized calcium binding adaptor molecule 1 (Iba1, AIF-1), a microglia-specific protein in mammalian brains. Second, serotonergic fibers were visualized with immunohistochemistry, which revealed their major pathways and demonstrated their tendency to accumulate at pial borders in the forebrain. Third, a population of serotonergic fibers was simulated in a three-dimensional model of the angelshark brain by using supercomputing. The fibers were defined as paths of reflected fractional Brownian motion (FBM), a continuous-time stochastic process. The results reproduced key features of serotonergic fiber densities in the telencephalon, a brain division with a considerable physical uniformity and no dense axon tracts. This study contributes to comparative and computational neurosciences and provides evidence that serotonergic fibers can be modeled as paths of a rigorously-defined stochastic process. It also supports the hypothesis that the structural self-organization of the serotonergic system depends on axons that have strongly stochastic trajectories but also respond to external forces.

Disclosures: **S. Janusonis:** None. **R. Metzler:** None. **T. Vojta:** None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.09/B10

Topic: A.09. Development and Evolution

Support: NIH Grant R01 - MH132089

Title: Human accelerated region modulates chromosomal interactions to shape neurogenesis

Authors: ***F. MOSTI**¹, J. LIU², K. LAM³, S. SKAVICUS³, V. A. KAPPS³, K. POLLARD⁴, D. L. SILVER⁵;

¹Neurobio., ²MGM, ³Duke Univ., Durham, NC; ⁴Gladstone Inst., San Francisco, CA; ⁵Mol. Genet. and Microbiology, Duke Univ. Med. Ctr., Durham, NC

Abstract: Changes to the human genome have driven the evolution of human features such as a larger neocortex and enhanced cognitive functions. Human Accelerated Regions (HARs) are highly-conserved loci that exhibit unexpected high rates of mutations in our genome, with ~half are located near neurodevelopmental genes and acting as transcriptional enhancers in neural cells. However, functions of HARs remain largely elusive, and their mechanism of gene regulation is still widely unknown. Here, we show that the enhancer, *HAR1984*, determines neural progenitor cell identity and neurogenesis by influencing species-specific chromatin

interactions. Ancestralization of *HAR1984* in human cortical organoids led to reduction of intermediate progenitors and neurons, whereas humanized *HAR1984* knock-in of chimpanzee cortical organoids produced the opposite phenotype. Moreover, humanized *HAR1984* mouse model showed increased progenitor proliferation and neuron number resulting in a thicker cortex with cortical folding. Hi-C data revealed a chromatin loop of *HAR1984* with its target genes, *ETV5* and *TRA2B*, present only in human fetal brains, but absent in neural cells from chimpanzee, macaque or mouse. Furthermore, acquisition of a human-specific ETV5 binding site in a human-specific element of *Hs-HAR1984* lead to a positive feedback regulatory loop, driving robust transcriptional activity. Our work demonstrates a new molecular mechanism adopted by an enhancer HAR to acquire human specific features during neurodevelopment, establishing a direct link between chromatin architecture, cortical cell identity and evolutionary expansion of human brain.

Disclosures: F. Mosti: None. J. Liu: None. K. Lam: None. S. Skavicus: None. V.A. Kapps: None. K. Pollard: None. D.L. Silver: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.10/B11

Topic: A.09. Development and Evolution

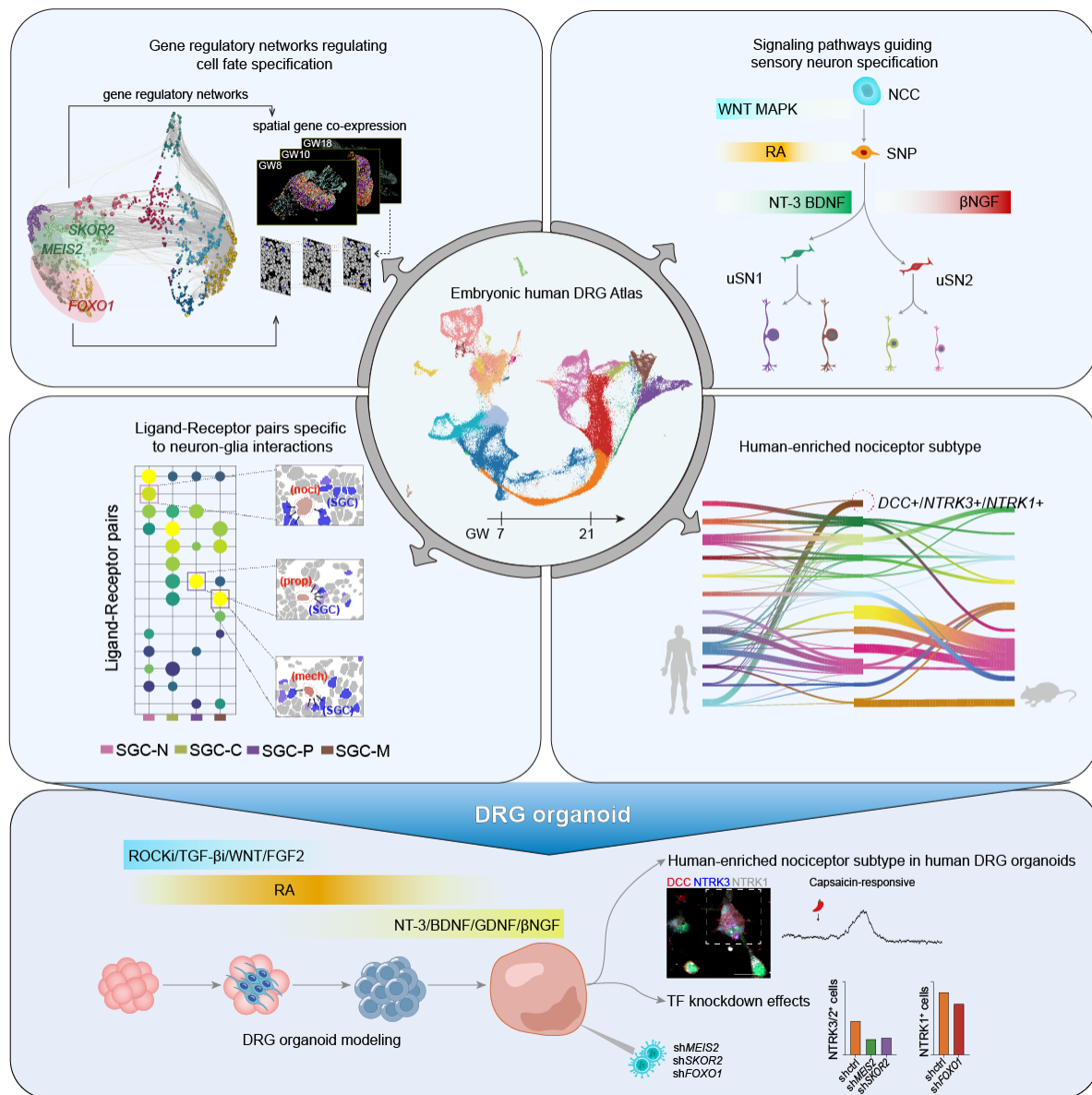
Support: Brain Science and Brain-inspired Intelligence Project of China
2021ZD0200102
Brain Science and Brain-inspired Intelligence Project of China
2021ZD0202403
CAS Project for Young Scientists in Basic Research YSBR-013

Title: Decoding Transcriptional Identity in Developing Human Sensory Neurons

Authors: *M. WANG;
CAS, Beijing, China

Abstract: Dorsal root ganglia (DRG) are pivotal in the sensory nervous system, harboring the cell bodies of sensory neurons responsible for detecting and transmitting sensory signals. Despite their importance, the cell heterogeneity and cellular mechanisms regulating the development of human DRG remain poorly understood. To address this gap, we constructed a single-cell spatiotemporal transcriptomic atlas of human embryonic DRG spanning from the first to second trimesters. This atlas profiled the cell type diversity and highlighted the extrinsic signaling cascades and intrinsic regulatory hierarchies guiding cell fate decisions, including neuronal/glia lineage restriction, sensory neuron specification and differentiation, and neuron-satellite glial cell (SGC) unit formation. We identified two unspecialized sensory neuron

populations (uSN1 and uSN2) emerging from two neurogenesis waves and revealed a human-enriched NTRK3+/DCC+ nociceptor subtype involved in multimodal nociceptive processing. Additionally, we explored the spatial interactions between neuronal subtypes and glial cells and found specific cell interactions that mediate the functional maturation of sensory neuron subtypes. In summary, our research elucidated the multilevel signaling pathways and TF regulatory hierarchies that underpin the diversity of human sensory neurons. **Keywords:** human dorsal root ganglia, sensory neuron, neurogenesis wave, transcription factors, single-cell spatial transcriptome, nociceptor, TF-seqFISH



Disclosures: M. Wang: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.11/B12

Topic: A.09. Development and Evolution

Support: NS095654
MH106934
MH116488
MH110926
MH129981

Title: Anti-correlated gene expression patterns along the developing sensorimotor-association axis

Authors: *I. PAVLOVIC, J. A. TSYPORIN, M. ZHANG, C. QI, S. BANDIERA, A. SEGAL, N. SESTAN;
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Abstract: The cerebral neocortex is organized along a dominant sensorimotor-to-association (S-A) axis, anchored at one end by modality-specific primary sensorimotor areas and at the other by transmodal association areas that form distributed axonal networks supporting higher-order cognition. Based on converging experimental evidence in several species, we recently proposed the Multinodal Induction-Exclusion in Network Development (MIND) model which describes developmental mechanisms underpinning S-A axis formation. We identified SEMA7A and PLXNC1, a mutually repulsive axon guidance pair characterized by anti-correlated expression, with SEMA7A being expressed in primary sensorimotor areas (S) and PLXNC1 being expressed in association areas (A). Utilizing immunohistochemistry (IHC) on developing wildtype murine brain tissue, we quantified the expression patterns of these proteins and observed an enrichment of SEMA7A in cortical sensorimotor areas and their complementary first-order thalamic nuclei while cortical association areas and high-order thalamic nuclei were enriched for PLXNC1. To investigate the evolutionary conservation of this patterning, we performed RNAscope on developing mouse, short-tailed opossum and chicken forebrain which showed a conserved pattern of Sema7a expression nested within a distributed Plxnc1 expression. These findings suggest that SEMA7A and PLXNC1 are enriched along the S-A axis with SEMA7A in S and PLXNC1 in A, and may regulate the connectivity of these networks. Next, we examined how the expression patterns of SEMA7A and PLXNC1 are affected by perturbations in neocortical development. To do so, we generated Zbtb18flox/flox; Neurod6-Cre (Zbtb18 cKO) mice characterized by defects in the primary area development and analyzed the expression of SEMA7A and PLXNC1 using IHC. We found defects in primary area development in Zbtb18 cKO mice along with dramatically decreased SEMA7A expression accompanied by a complementary expansion of the PLXNC1 domain. Our results underscore SEMA7A and PLXNC1 as being exemplary markers of S-A axis patterning and regulators of S-A network connectivity.

Disclosures: I. Pavlovic: None. J.A. Tsyporin: None. M. Zhang: None. C. Qi: None. S. Bandiera: None. A. Segal: None. N. Sestan: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.12/B13

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Title: Patterns of cortical connectivity across primates

Authors: *A. MURPHY¹, S. BARTON¹, M. MOHEBBI¹, J. PARGETER², M. SINHA¹, S. BROSINAN³, A. V. KUKEKOVA⁴, R. B. MARS⁵, L. A. PARR⁶, K. A. PHILLIPS⁸, C. C. SHERWOOD⁹, D. STOUT⁷, E. E. HECHT¹;

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Abstract: Understanding how corticocortical connectivity varies across primates offers critical insights into the neural architecture underlying cognitive and sensory integration. We tested two developmentally grounded hypotheses about brain connectivity: the rostrocaudal gradient hypothesis and the association network hypothesis. The rostrocaudal gradient hypothesis predicts that long distance connections increase progressively towards the rostral end of the cerebral cortex, and that this connectivity pattern scales with brain size. The association network hypothesis predicts that long distance connections increase with greater distance from primary sensory regions of the cerebral cortex. Our dataset includes over a dozen different primate species, including platyrrhine monkeys, cercopithecoid monkeys, and great apes, providing wide coverage of the phylogenetic breadth and brain size variation in the clade. Diffusion-weighted MRI scans were analyzed using probabilistic tractography. Voxelwise analysis was performed using white matter as a seed mask and cortical grey matter as a target mask. The resulting

connectivity matrix was used to produce measures of streamline count and average path length from each voxel in cortex to the rest of cortex. These variables were measured along the rostrocaudal axis of the brain as well as with increasing distance from primary sensory regions (V1, S1, A1). We found that as brain size increases, brains were increasingly characterized by association network patterns of connectivity. Comparisons with out-group carnivore brains, including silver foxes and domesticated dogs, suggest that there may be aspects of this connectivity pattern that are clade-specific, including potentially mosaic patterns that may have emerged in early primates to support life in the fine-branch niche.

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Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.13/B14

Topic: A.09. Development and Evolution

Title: Human brain size in adults: a reference database with 6,321 carefully screened brains as a tool for interdisciplinary research

Authors: *M. CHAMBERS¹, H. SANCHEZ²;

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Abstract: Motivation/problem statement: Brain weight is a foundational metric in neuroscience research. It serves as a reference point in studies of neurodevelopment, neurodegeneration, aging, brain injury, and comparative neuroanatomy. Deviations from normative brain weights can signal abnormal growth, atrophy, or pathology. Despite the centrality of brain weight in such studies, many reference tables in use today suffer from critical limitations: outdated datasets (some over a century old), small sample sizes, and inadequate screening for disease or postmortem artifact. These issues limit the accuracy and applicability of reference data, especially when used to draw inferences about normal versus pathological brain states. In response, we present updated, high-quality brain weight reference data designed to support modern neuroscience research and clinical interpretation. **Methods/approach:** Brain weights, along with demographic and body metrics, were collected from 8,280 carefully screened adult autopsies performed between 2009 and 2023 across five U.S. medical centers. Strict inclusion criteria were used to exclude cases with prolonged postmortem intervals, significant decomposition, or known neuropathologic processes affecting brain weight. Data were stratified by sex and evaluated using forward and stepwise regression analyses to assess associations between brain weight and indexing variables such as age, height, weight, and BMI.

These results were organized into updated reference tables and interactive graphs. **Results:** 8,280 decedents were included in the adult study, of which 6,321 met the inclusion/exclusion criteria for the brain. That women's brains were, on average, smaller than men's was confirmed and all subsequent analyses were conducted on men and women separately. Height was the single best fit metric in both women and men, followed by age. There was significant correlation between the different index metrics. Obesity was not significantly associated with brain weight.

Conclusion/implications: This updated brain weight dataset offers the neuroscience community a modern, rigorously curated reference resource. It addresses long-standing gaps in the literature and provides improved tools for evaluating brain size across the adult lifespan. The aggregate data is accessible as an interactive digital tool at organweights.theautopsybook.com, promoting transparency and collaboration. Line-item data, and opportunities for broader research collaboration, can be accessed through the digital tool and the authors herein.

Disclosures: M. Chambers: None. H. Sanchez: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR258.14/B15

Topic: A.09. Development and Evolution

Title: Neocortical and Corpus Callosum Morphology Across Mammalian Orders

Authors: A. GUZMAN¹, J. TODOROV¹, *W. E. GRISHAM²;

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Abstract: Various claims have been made about the relationship of the neocortex to white matter across mammalian orders, but there has not been an examination of the neocortex and a specific fiber tract. Here we examine measures of thickness of the neocortex and corpus callosum across different orders of mammals. Thirty-nine brains from 4 orders of mammals were obtained from the Comparative Mammalian Brain Collections (<https://brainmuseum.org/>) (16 carnivores, 8 primates (excluding human), 8 ungulates, & 7 rodents). Bilateral neocortex and corpus callosum thicknesses were measured using ImageJ. The corpus callosum and the thickest and thinnest neocortex were measured at the rostral-most point of the corpus callosum spanning the hemispheres, the rostral-most point of the hippocampus, and caudal-most extent of the corpus callosum. Corpus callosum measures were taken off the midline to avoid including the hippocampal commissure. Measures were independently averaged for each animal across rostral-caudal levels. Adjustments for phylogenetically relatedness were made. Data analyzed were the resultant residuals as well as raw data. Although we expected differences across orders in the ratio of CC/neocortex measures, none was found. Rather, the ratio components did show differences. Rodents had the thickest marginal mean raw and residual neocortex and differed from carnivores and ungulates. Ungulates had the thinnest marginal mean neocortex and differed

also from primates. The residual data showed the same pattern as the raw data. Further, regression lines of each order relating raw or residual neocortex thickness to log10 brain size differed between rodents and primates as well as between rodents and ungulates. Analyses of the raw CC data showed similarities to the raw cortex data. Ungulates had the smallest marginal mean and a significantly thinner CC than did any other order. The residual CC data showed the exact same pattern. Among the regression slope differences for individual orders, ungulates differed from carnivores in both the raw and residual data. Although we did not have the ability to determine densities of either neurons or axons, our findings suggest that ungulates may have fewer neurons/cortical sheet than the other orders measured and their connectivity across hemispheres may be poorer.

Disclosures: A. Guzman: None. J. Todorov: None. W.E. Grisham: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

Location: SDCC Halls B-H

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Topic: A.09. Development and Evolution

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Title: Cortex scaling differences exist in age-equivalent preterm and term neonatal brain

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Abstract: A universal scaling rule, $T^{0.5}A_t = kA_e^{1.25}$, for the human cortex has been previously proposed, where T is the average cortical thickness, A_t is true cortex surface area, A_e is the exposed cortex surface area (with the folded cortex area been ignored), and k is a dataset independent slope reflecting the linear relationship. This rule originates from the minimization of the free energy associated with the cortical shape. This rule has been widely validated using a) normal adult brain MRIs, including the HCP, OASIS, and NKI datasets; and b) diseased brain MRIs from ADNI dataset. However, the validity of this universal scaling rule in the infant brain remains largely unknown, most likely due to the limited infant image data and the need of specialized image processing approaches to mitigate the difficulty of accurately assessing cortical thickness and surface area.

Levering the neonatal brain MRI dataset from the developing human connectome (dHCP) project

and the iBEAT V2.0 infant brain imaging processing pipeline, we aimed to determine whether this scaling rule applies for the neonatal brain cortex, including the preterm and term infants. In addition, we statistically tested whether there are scaling differences in the slope (k) between preterm and term infants.

The dHCP brain dataset includes 885 scans. Of which, 308 scans were preterm (birth age <37 week) babies and 146 subjects were scanned at term equivalent age (scan age ≥ 37). Images were preprocessed using the iBEAT V2.0 pipeline to compute the hemispherical average cortical thickness (T), true cortex area (A_t) and the exposed cortex area (A_e). We then fitted these parameters using a linear regression to test the scaling rule's applicability on neonatal cortex. ANCOVA was employed for statistical analyses.

We found that the scaling rule perfectly applies to the neonatal brains for both term and preterm infant brains. A strong linear relationship between $T^{0.5}A_t$ and $A_e^{1.25}$ for both the left (with $p < 1e-16$, $R^2 = 0.965$) and right hemisphere (with $p < 1e-16$, $R^2 = 0.965$) was found. Moreover, there is no significant slope differences between the left and right hemisphere (with $p = 0.259$, $CI = [-0.002, 0.008]$). Importantly, however, the age-equivalent preterm and term born baby brains exhibited statistically significant differences in the cortex scaling for both the left (with $p = 0.001$) and the right (with $p = 0.006$) hemispheres. One of the most plausible factors leading to the observed differences is the potential injury of white matter, commonly observed in pre-term infants, which in turn may change the folding energy during the cortex folding processes.

Disclosures: Z. Wu: None. Y. Sun: None. L. Wang: None. W. Lin: None. G. Li: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

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Program #/Poster #: PSTR258.16/B17

Topic: A.09. Development and Evolution

Support: NIH Grant P20GM113109

Title: Analysis of Dopaminergic and Otp-Expressing Neurons in Zebrafish Reveals Evolutionary Constraints of the Extended Olfactory Amygdala

Authors: D. WEBSTER¹, V. WILLIAMS¹, A. SHEA¹, *T. MUELLER^{2,1};

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Abstract: Zebrafish has become an important model system for neural mechanisms of olfaction and neurodevelopmental disorders like autism that affect the sense of smell and taste. However, the zebrafish's extended amygdala (EA) and olfactory pallium have remained ill-understood. To define the zebrafish EA, we analyzed the distribution of the catecholamine-indicating protein tyrosine hydroxylase (TH), versus those of otpa-protein, calretinin, and GABA in zebrafish transgenic lines *Tg(lhx5:GFP)*, *Tg(isl1:GFP)*, *Tg(vGlut2a:GFP)*, and *Tg(lhx2a:GAP-YFP)*.

Comparing the distribution of TH and otpa with those of GFP in *Tg(lhx5:GFP)*, *Tg(isll:GFP)* and *Tg(vglut2a:GFP)*, we redefine the extended medial amygdala and portions of the olfactory pallium previously mislabeled as “Dp” in zebrafish. In sum, our results define the extended olfactory amygdala in zebrafish, thereby strengthening its use for spectrum autism disorders, which affect the sense of smell and social behavior.

Disclosures: D. Webster: None. V. Williams: None. A. Shea: None. T. Mueller: None.

Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

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Topic: A.09. Development and Evolution

Support: ASAP-025193

Title: Deep Cerebellar Nuclei Scaling in New World Monkeys: An Atlas-Based Volumetric Comparison of Marmoset and Cebus.

Authors: *M. LIN^{1,2}, C. M. CERKEVICH², A. C. BOSTAN², M. HANADA², P. L. STRICK²;
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Abstract: We introduce the first high-resolution, MRI-aligned histological atlas of the cerebellum of the capuchin monkey (*Cebus apella*). The atlas delineates each lobule of the cerebellar cortex as well as each of the deep cerebellar nuclei. The atlas fills a critical gap by providing an interactive, open-source platform that allows researchers to visualize labelled subdivisions, explore metadata, and overlay user-defined datasets. This resource includes a 2-D flatmap of the cerebellar cortex, which enables intuitive visualization of the entire cerebellar cortical sheet and facilitates direct comparison with the cerebellar cortex of marmosets, macaques and humans.

We leverage this resource, together with our recent Marmoset (*Callithrix jacchus*) Cerebellar Atlas (marmosetbrainconnectome.org/cerebellum), to gain insights into specific comparative alterations of the cerebellum in New World monkeys. In one initial analysis, we normalized the volumes of the intermediate (interpositus) and lateral (dentate) deep nuclei in the marmoset and cebus to the volume of the medial (fastigial) deep nuclei in these monkeys. We found that the relative volume of the cebus dentate is nearly 6 times larger than the marmoset dentate. In contrast, the relative volume of the cebus interpositus is only ~2 times larger than the marmoset interpositus. The striking relative expansion of the dentate in the cebus monkey may correlate with this nonhuman primate's remarkable ability to manufacture and use tools.

Overall, our atlas of the cebus cerebellum lays foundation for mapping experimental data (e.g., viral tracers, gene expression, cellular distributions) into a unified anatomical space and will

support a wide range of neurobiological applications from developmental and behavioral studies to comparative connectomics.

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Poster

PSTR258: Comparative and Evolutionary Mechanisms of Brain Development and Molecular Specialization

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Title: Comparative analysis of primary cilia in cortical regions across vertebrate species to interpret cortical evolution

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Abstract: Primary cilia are hair-like structures that are expressed in most mammalian cells, including neurons. We have shown that the primary cilia of principal neurons in loosely layered, or inside-out laminated, brain regions predominantly orient toward the pial surface. However, the primary cilia of principal neurons in compacted layered, or outside-in laminated, brain regions often manifest opposite directionality. In contrast, astrocytes and interneurons throughout the cerebral cortex, and neurons in nucleated brain regions, do not display directional cilia orientation. The directional cilia orientation present in pyramidal neurons in inside-out laminated regions reflects a reverse movement for positioning. The brain structure of newts (amphibians) shares basic pallial subdivisions - dorsal, lateral, and medial regions -with that of reptiles such as turtles. Unlike the mammalian or reptile brain, the chicken brain lacks a laminated cortical organization and follows a distinct structural arrangement. The primate brain has an abundance of loosely laminated regions except for the CA3 region and dentate gyrus. We examine cilia orientation in various cortical regions across multiple species - newts, turtles, chickens, mice, and macaques - to interpret the process of principal neuron positioning and cortical evolution.

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Topic: A.09. Development and Evolution

Support: 1U01DA055344
U01MH110274

Title: Emergence of anatomically symmetric and consonant redundant functional communities during early infancy

Authors: *X. LI¹, W. JIANG^{2,3}, W. YIN², T. LI², W. LIN²;

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Abstract: RsfMRI studies largely rely on simple pair-wise correlations, which cannot reveal what information is communicated across brain regions. Recent advances utilize information theory to decompose BOLD signals into synergetic and redundant communications among brain regions, reflecting functional integration and robustness, respectively. High redundancy marks *robustness*: if one node is compromised, the shared information—and its supported function—remains accessible through its partners. In this study, we hypothesized that (1) redundant information communication in the brain forms different functional communities from transitional functional networks and (2) charting developmental features of redundancy networks provide additional insights into brain development. We analyzed 525 resting-state fMRI scans from 240 typically developing infants who were stratified into four age groups: 0-6 mo, 6-12 mo, 12-18 mo, and 18-28 mo (n = 104, 153, 143 & 125). With preprocessed BOLD signals extracted from 232 regions, pairwise regional synergetic and redundant interactions were obtained via integrated information decomposition. We constructed group-level mean redundancy matrices quantifying probability that any two regions co-occupy a community across runs and generated 100 generalized Louvain partitions for each age group. Age-bin consensus partitions were identified by iterative reclustering; reliability was assessed with adjusted Rand index (ARI) across partitions withheld during jack-knife resampling. Network Correspondence Toolbox was used to compare redundancy defined communities to canonic functional networks. 5, 6, 7, and 6 highly reproducible communities were obtained for the four age groups (mean ARI = 0.824, 0.869, 0.845, 0.824), respectively, demonstrating age-dependent increases of numbers of communities. Topologies of these communities revealed left-right symmetric and anatomically consonant modules for all age groups. Dice correspondence with adult Yeo-7 atlas showed one redundancy

defined community sharing high similarity with the visual network independent of age ($p < 0.05$, spin test), while the remaining communities either blended multiple networks or showed no significant match, indicating distinct redundant communities from canonic networks. Our results demonstrate that redundancy-aware Louvain clustering reveals a robust and anatomically coherent functional architecture in the infant brain. These redundancy communities offer a novel framework for investigating atypical neurodevelopment, shifting focus from temporal synchrony to the nature of information exchanged between brain regions.

Disclosures: X. Li: None. W. Jiang: None. W. Yin: None. T. Li: None. W. Lin: None.

Poster

PSTR259: All About GPCRs

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.01/B21

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Dopamine D3 Receptor Mediated Action in Hippocampus CA1

Authors: *K. M. KEARY, III, E. HAMADA, V. A. SHVAREVA, K. J. BENDER;
Neurol., UCSF, San Francisco, CA

Abstract: Schizophrenia (SCZ) is a psychiatric disorder affecting over 1% of the global population. SCZ is characterized by positive, negative, and cognitive symptoms. Second generation antipsychotic (SGA) medications have high affinity for the D2 family of dopamine receptors, with highest affinity for D2 and D3 receptors (D2R, D3R). While positive symptoms can be treated with some success via antagonism of D2Rs, currently available SGAs provide limited relief for cognitive symptoms, which may originate from aberrant hippocampal signaling. Cognitive symptoms are often the first to present, compounding the severity of inadequate cognitive symptom treatment and establishing a clear and pressing need for investigations centered around SCZ cognitive symptom etiology and treatment. While D2R antagonism has not been successful in treating cognitive symptoms, the new antipsychotic cariprazine functioning as a D3R partial agonist has shown some signs of cognitive symptom relief. This introduces a new possibility that, within the hippocampus, D3Rs may be a more appropriate target for treating SCZ cognitive symptoms. Here, we report in-situ hybridization and electrophysiology studies examining the localization and function of hippocampal dopamine D3Rs. Investigation of D2-GFP and D3-Ai14 reporter lines shows much stronger D3 signal throughout the hippocampus as compared to D2. To further assess the relative concentrations of the D2-family dopamine receptor expressing cells, we utilized hybridization chain reaction (HCR)-FISH to quantify *Drd2*, *Drd3*, and *Drd4* transcript positive cells. Preliminary HCR-FISH experiments indicate over 70% of cells within the CA1 pyramidal layer are *Drd3* positive as compared to 7% *Drd2* positive. *Drd4* was not detected. Additionally, *Drd3* transcripts were detected in parvalbumin-positive, but not somatostatin-positive interneurons. To assess D3R-mediated cellular mechanisms of action, we conducted electrophysiological recordings from tdTomato positive pyramidal cells in D3-

Ai14 slices. The D3-Ai14 line is a strong reporter of active Drd3 expression as 89% of tdTomato positive cells are also Drd3 positive. When assessing miniature excitatory post-synaptic currents (mEPSCs), we found that D3R agonism via PD128907 application reduced mEPSC amplitude. Miniature inhibitory post-synaptic currents (mIPSCs) were also affected by D3R agonism as mIPSC amplitudes decreased and mIPSC frequency increased in the presence of PD. This demonstrates diverse D3R-mediated modulation of hippocampus CA1 circuitry. Future experiments will focus on examining the D3R-mediated effects of different antipsychotics on CA1 function.

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Poster

PSTR259: All About GPCRs

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Program #/Poster #: PSTR259.02/B22

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH Grant R01DA047130

Title: Drug development targeting the serotonin 6 G protein-coupled receptor (5-HT₆R) for cognitive disorders

Authors: *E. E. SULLIVAN^{1,2}, O. T. HOLBROOK³, C. F. FERRIS⁴, M. CUI⁵, R. BOOTH³; ¹Pharmaceut. Sci., ²Chem. and Chem. Biology, Ctr. for Drug Discovery, ³Pharmaceut. Sciences, Chem. and Chem. Biology, Ctr. for Drug Discovery, ⁴Pharmaceut. Sciences, Psychology, Ctr. for Translational Neuroimaging, ⁵Pharmaceut. Sciences, Ctr. for Drug Discovery, Northeastern Univ., Boston, MA

Abstract: The serotonin (5-hydroxytryptamine) type 6 G protein-coupled receptor (5-HT₆R) is expressed exclusively in the central nervous system, particularly in striatal, limbic, and cortical areas. Accordingly, drugs targeting the 5-HT₆R are proposed to treat disorders involving cognitive impairment, including Alzheimer's disease, dementia with Lewy bodies, and schizophrenia. Currently, however, no drugs are approved that specifically target the 5-HT₆R. To address this pharmaco-therapeutic gap, we used a 5-HT₆R structure-based drug design paradigm using a refined version of the reported 5-HT₆R cryo-EM structure (PDB: 7XTB) and 1 μ s molecular dynamics (MD) simulations, together with molecular pharmacological studies, to develop a series of 7-substituted-2-aminotetralin (7-SAT) type 5-HT₆R agonists and inverse agonists. The MD results suggested different 7-SATs differentially interact with a key tryptophan residue in transmembrane domain 6 (W6.48) that impacts stabilization of different 5-HT₆R conformations and activation vs. inactivation of the receptor. Experimentally, 5-HT₆R affinity of 7-SATs was determined by competitive displacement of [³H]-LSD and signaling activity was measured as cAMP accumulation in HEKt cells transiently expressing 5-HT₆Rs. A

new 7-SAT type 5-HT₆R agonist and inverse agonist ligand were identified; both, with potent functional activities (preliminary EC₅₀/IC₅₀ values ≤20.0nM). In our preliminary preclinical translational studies, we used only male Sprague Dawley rats. First, we conducted a pharmacological MRI (phMRI) study using awake rats approximately 7 weeks old. The phMRI study included 3 groups; vehicle (n=7), 7-SAT 5-HT₆R agonist (n=6, 5mg/kg), and 7-SAT 5-HT₆R inverse agonist (n=8, 5mg/kg), which provided comparative information regarding 7-SAT modulation of specific neuronal networks in the brain as measured by positive and negative blood oxygen level dependent (BOLD) signaling. Results were that the 7-SAT 5-HT₆R inverse agonist showed a more significant effect on brain activity than the agonist, impacting brain areas that included the hippocampus, cerebellum, hypothalamus, amygdala, midbrain, and cortex. In preliminary open field tests, we found that the lead 7-SAT inverse agonist did not impact general locomotor activity, and rats tended to spend increased time in the center zone. In pilot novel object recognition (NOR) assays, results with the lead 7-SAT inverse agonist indicated a trend to reduce cognitive impairment induced by scopolamine (2mg/kg), as evidenced by a higher NOR index total time exploring compared to the scopolamine-only and scopolamine plus inverse agonist conditions.

Disclosures: **E.E. Sullivan:** None. **O.T. Holbrook:** None. **C.F. Ferris:** A. Employment/Salary (full or part-time): Ekam Imaging/Solutions. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Ekam Imaging/Solutions. **M. Cui:** None. **R. Booth:** None.

Poster

PSTR259: All About GPCRs

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Program #/Poster #: PSTR259.03/B23

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: R01DK114008
F31DK142351
Indiana University Indianapolis Graduate Fellowship
Undergraduate Research Opportunity Program

Title: Are Tanycyte Cilia Interactions Influenced by Physiological Conditions

Authors: ***K. R. SCHWANTZ**¹, K. BREWER³, N. F. BERBARI⁴, J. BOONE²;
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Abstract: Tanycytes are glial cells which line the third ventricle (3V) and are important in sensing peripheral cues to regulate hypothalamic input and output. These cells appear to be unique in that they can alter their morphology in response to external stimuli and physiological changes, such as metabolic and seasonal fluctuations. Thus, their processes that extend into the

hypothalamic parenchyma appear to be dynamic and communicate with neurons, glia, and capillaries to regulate signaling via physical contact. Interestingly, primary cilia on neurons in the hypothalamus are also thought to contact neuronal synapses and CNS cell types to mediate signaling. Hypothalamic cilia are small, microtubule based structures known to mediate neuronal signaling and physiological processes, like feeding and circadian rhythm by localizing a variety of different signaling proteins, such as GPCRs. Here, we hypothesize that tanycytes and cilia within the hypothalamic nuclei contact one another to regulate signaling within the brain. These contacts are most likely dynamic in nature, changing based on different physiological states. We assessed tanycyte-cilia contacts in the hypothalamus of male and female mice as well as mice under different metabolic (*ad libitum*, calorie restriction, high fat diet) and seasonal (long and short photoperiods) conditions. This provides evidence for how tanycytes communicate peripheral cues (serum and CSF) to the hypothalamus under different physiological stress. Together, these data provide insight into different mechanisms cilia may deploy to mediate neuronal signaling and behaviors.

Disclosures: K.R. Schwantz: None. K. Brewer: None. N.F. Berbari: None. J. Boone: None.

Poster

PSTR259: All About GPCRs

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.04/B24

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIDA INVEST Fellowship
NIH grant # R01DA060923

Title: Mu-opioid receptor expression and function in heterogeneous neurons of the interpeduncular nucleus

Authors: *A. SZLAGA, A. J. FLORES, W. S. CONRAD, A. MAJUMDAR, T. S. HNASKO; Univ. of California San Diego, Dept. of Neurosciences, La Jolla, CA

Abstract: Interpeduncular nucleus (IPN) is a midbrain structure involved in avoidance, anxiety-related behaviors, and nicotine dependence. IPN is the main output of the medial habenula (MHb) and the habenulo-peduncular axis has also emerged as a relevant target in opioid dependence. While MHb was identified as an early hotspot of MOR expression and binding, a significant subset of IPN neurons also express MOR, but the neurochemical content of these cells and their projection targets has not been established. Here, we aim to characterize IPN cell heterogeneity and the influence of MOR activation in mice IPN. The IPN is considered a primarily GABAergic structure, but also contains populations of neurons that express vesicular glutamate transporters (VGLUT2 or VGLUT3). Vesicular monoamine transporter (VMAT2)-expressing serotonin neurons are also present and these cells may represent a rostral extension of raphe nuclei. We used fluorescent *in situ* hybridization to assess the relative fractions of these

neurons and determine which express MOR. Within IPN we found that approaching 20% of neurons express VGLUT2, these cells are most abundant in rostral IPN sub-regions, and are largely devoid of MOR expression. VGAT-expressing neurons make up the bulk of the remaining cells and nearly all MOR-expressing neurons express VGAT. VGLUT2 and VGAT thus represent largely non-overlapping populations of IPN neurons, and MOR+ neurons represent a subset of VGAT+ neurons. Moreover, a significant subset of VGLUT3+ cells co-express VGAT, but not VGLUT2, and many of them co-express MOR and/or VMAT2 and are more concentrated at the caudal portion of IPN. *Ex vivo* patch-clamp recordings showed that MOR activation directly inhibits MOR+ IPN neurons, simultaneously decreasing the frequency of spontaneous excitatory postsynaptic currents. Finally, projection-mapping experiments revealed non-overlapping innervation of brainstem structures made by MOR+ and VGLUT2+ IPN neurons. Altogether, our findings suggest that MOR is expressed in distinct populations of IPN neurons supporting distinct functional roles for heterogeneous IPN cell types.

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Poster

PSTR259: All About GPCRs

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.05/B25

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Interaction of 2,4,5-phenylethylamine psychedelics with the 5-HT_{2A} receptor: effects of transposing the position of the methoxy groups.

Authors: *B. CUCCURAZZU¹, N. SHACHAM², T. FANNANA³, P. WHITE³, S. CAMERON³, J. WALLACH³, A. L. HALBERSTADT⁴;

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Abstract: There is considerable interest in the therapeutic potential of classical psychedelic drugs (5-HT_{2A} receptor agonists). In previous investigations, the activity of phenylethylamine (PEA) psychedelics at 5-HT_{2A} was enhanced when there are methoxy groups at the 2- and 5-positions and a bulky hydrophobic 4-substituent. However, other ring-substitution patterns have received less attention. To fill that gap, we explored the effect of transposing one of the methoxy groups in the 2,4,5-phenylethylamines 2C-B (4-bromo-2,5-dimethoxy-PEA) and 2C-D (2,5-dimethoxy-4-methyl-PEA) to a different position (moving the 2-methoxy to the 3-position, or the 5-methoxy to the 6-position). Because α -methyl and *N*-(2-methoxy)benzyl (NBOMe) groups enhance the activity of some PEAs, the effect of adding those groups was evaluated across the three series. Activity at 5-HT_{2A} was assessed using multiple assays, including binding affinity (displacement of the radioligand [³H]ketanserin), potency (EC₅₀) for inducing Gq dissociation (measured using Bioluminescence Resonance Energy Transfer [BRET]), and potency (ED₅₀) in

the head-twitch response (HTR) assay in male C57BL/6J mice (a behavioral proxy for human psychedelic effects). Across the three series, the 2,4,5-trisubstitution pattern consistently had the highest activity at 5-HT_{2A}. Transposing the 5-methoxy to the 6-position reduced 5-HT_{2A} activity by one-half to one-fifth. In comparison, the effect of transposing the 2-methoxy to the 3-position was less consistent: there were typically modest reductions in potency in the HTR assay, whereas 5-HT_{2A} affinities and agonist potencies were reduced 5-20-fold. Adding an α -methyl group increased activity in the 2,4,5- and 3,4,5-series but reduced activity in the 2,4,6-series. Adding an NBOMe group produced a robust increase in the activity of the 2,4,5- and 2,4,6-series, but had much less of an effect on the 3,4,5-series. The effect of adding an NBOMe substituent was markedly attenuated when it was combined with an α -methyl group. PEAs containing both substituents induced seizures in mice, suggesting potential off-target activity. There were significant correlations between in vivo and in vitro measures of 5-HT_{2A} activity, confirming that the HTR assay serves as a readout of 5-HT_{2A} activation. However, based on a subset analysis, the strength of the relationship is much greater when the 3,4,5-series is excluded. Overall, these results provide important insight into the structure-activity relationships of PEA psychedelics. Future investigations will attempt to understand the molecular ligand-receptor interactions responsible for the observed substituent effects.

Disclosures: **B. Cuccurazzu:** None. **N. Shacham:** None. **T. Fannana:** None. **P. White:** None. **S. Cameron:** None. **J. Wallach:** None. **A.L. Halberstadt:** None.

Poster

PSTR259: All About GPCRs

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.06/B26

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Characterizing Gi GPCRs using high throughput fluorescence and electrophysiology based assays for drug discovery

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Abstract: G protein-gated inwardly rectifying potassium (GIRK) channels are critical regulators of neuronal and cardiac excitability. In the brain, GIRK primarily assembles as a Kir3.1/Kir3.2 heterotetramer, activated by G $\beta\gamma$ subunits released upon stimulation of Gi/o-coupled GPCRs. GIRK activity serves as a direct and physiologically relevant readout of Gi/o signaling; however, conventional approaches—including FRET, BRET, and cAMP assays—are constrained by biological complexity or require artificial adenylate cyclase stimulation. We describe a Gi/o GPCR assay that enables high-throughput, real-time quantification of GIRK activation using both fluorescence-based and automated electrophysiology approaches. This

assay utilizes a HEK293 cell line stably expressing Kir3.1 and Kir3.2, with the flexibility to transiently express a range of Gi/o-coupled GPCRs using just-in-time expression constructs. Using this platform, we measured receptor-driven GIRK activation for the M2 muscarinic receptor, yielding EC₅₀ values that align with published data. The EC₅₀ for ML297, a GIRK activator, was found to be 100nM and 72 nM using fluorescence and automated patch clamp assays, respectively. What is more, GIRK channels could be activated by acetylcholine/carbachol in a concentration-dependent manner with an EC₅₀ of 15 and 20 nM, using fluorescence and automated patch clamp assays, respectively. This effect was not observed in HEK cells without M2 or GIRK channel expression.

This approach provides a physiologically relevant, scalable, and high-throughput method for identifying modulators of Gi/o-coupled GPCRs. Its compatibility with a variety of neurotransmitter receptors, and ability to generate reproducible pharmacological data using fluorescence and electrophysiological measurements, make it a valuable tool for both research and drug discovery efforts targeting Gi/o GPCRs.

Disclosures: **T. Strassmaier:** A. Employment/Salary (full or part-time): Nanion Technologies, Nanion Technologies. **A. Obergrussberger:** A. Employment/Salary (full or part-time): Nanion Technologies GmbH. **D. Hernandez:** A. Employment/Salary (full or part-time): Ion Biosciences. **Y. Lu:** A. Employment/Salary (full or part-time): Nanion Technologies. **N. Fertig:** A. Employment/Salary (full or part-time): Nanion Technologies GmbH. **S. Wiley:** A. Employment/Salary (full or part-time): Ion Biosciences.

Poster

PSTR259: All About GPCRs

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.07/B27

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: SERB, India (Grant no.- CRG/2022/001291)

Title: Internalization of group I metabotropic glutamate receptors and synaptic AMPA receptor endocytosis by dopamine

Authors: ***K. ARUNA**¹, **M. KULKARNI**², **S. BHATTACHARYYA**³;

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Abstract: Metabotropic glutamate receptors (mGluRs) are members of the G-protein coupled receptor (GPCR) family. Group I mGluRs consist of mGluR1 and mGluR5. These receptors play crucial roles in neuronal circuit formation and various forms of synaptic plasticity, including learning and memory. Dysregulation of these receptors may result in a variety of

neuropsychiatric disorders like fragile X syndrome, autism etc. Appropriate delivery of these receptors to the cell surface is a crucial step to place these receptors at the specific region of the neuron for proper signaling. The process of receptor trafficking plays a critical role in controlling the precise spatio-temporal localization as well as regulation of the activity of these receptors. Dopamine plays a vital role in the brain in reward and movement regulation. Hippocampus receives dopamine projections from the ventral tegmental area (VTA) and the substantia nigra pars compacta (SNc). Multiple evidences suggest a potential crosstalk between the dopaminergic system and the glutamatergic system in the hippocampus. These evidences tempted us to investigate whether dopamine, although not a cognate ligand for glutamate receptors, could modulate the group I mGluR trafficking and in turn, could also regulate mGluR-mediated AMPA receptor endocytosis which is the cellular correlate for the mGluR-mediated synaptic plasticity in the hippocampus. We found that dopamine induced internalization of both members of the group I mGluR family; mGluR1 and mGluR5. Dopamine-mediated internalization of mGluR1 was observed to be clathrin-dependent, but did not depend on dynamin. Subsequent to the dopamine-mediated endocytosis, majority of the receptors recycled to the cell membrane, much faster than what was observed when the receptors internalized through the activation by the agonist. Unlike the agonist-induced internalized receptors, recycling of the dopamine-mediated internalized mGluR1 was protein phosphatase 2B (PP2B)-dependent and did not depend on protein phosphatase 2A (PP2A). Finally, we demonstrate that dopamine plays an important role in the mGluR-mediated internalization of synaptic AMPA receptors. Taken together, these findings reveal a novel mechanistic role of dopamine in the trafficking of group I mGluRs and suggest its physiological implications in the brain.

Disclosures: K. Aruna: None. S. Bhattacharyya: None.

Poster

PSTR259: All About GPCRs

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Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: 1RF1NS132920-01A1 HEAL Initiative
1UF-1NS133763-01 NIH BRAIN Initiative
ZIA MH002970-06
NIH CCB Graduate Student Fellowship

Title: Kappa- and mu-opioid receptors are differentially compartmentalized and activated by endogenous peptides across paraventricular thalamic circuits

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Abstract: The paraventricular nucleus of the thalamus (PVT) is a stress-sensitive region of the brain that regulates emotional and motivational processes. The PVT controls behavior largely through projections to limbic regions including the nucleus accumbens (NAc), central amygdala (CeA), medial prefrontal cortex (mPFC). Neuromodulatory systems implicated in emotion and motivation, like endogenous opioids, are highly expressed in PVT neurons, and understanding how endogenous opioids are configured to modulate PVT neurons is crucial for elucidating how this system drives behavior. The PVT expresses both kappa- (KOR) and mu-opioid receptors (MOR) which are activated by the endogenous peptides dynorphin (Dyn) and enkephalin (Enk), respectively. Interestingly, KOR and MOR are both inhibitory Gi-coupled GPCRs, but Dyn/KOR signaling promotes aversive states while Enk/MOR signaling promotes euphoric states, suggesting an antagonistic relationship between KOR and MOR. Yet, the differences in KOR and MOR expression patterns and inhibition of PVT subcompartments is unknown. Using patch-clamp electrophysiology, we show PVT cells generate inhibitory currents in response to Dyn and Enk stimulation of somatodendritic opioid receptors, suggesting the presence of MOR and KOR. A larger proportion of opioid peptide responsive cells are found within PVT neurons that project to the NAc and CeA than mPFC projectors. Interestingly, pharmacological and genetic manipulations have uncovered that both Dyn and Enk currents are MOR-mediated, and KOR is not present at PVT dendrites/soma. In contrast, in PVT axon terminals in the NAc, pharmacological and genetic manipulations demonstrate that both KOR and MOR are present on PVT terminals, and KOR is required for Dyn signaling. Additionally, MOR-mediated inhibition is larger than and occludes inhibition by KOR, while KOR inhibition only partially occludes MOR inhibition. This suggests that either MOR is expressed on more terminals than KOR or has stronger inhibition than KOR on terminals. In contrast, investigation of PVT terminals in the mPFC and CeA shows strong MOR inhibition and weak KOR inhibition. This highlights the widespread pattern of MOR localization to all PVT neural compartments and circuits assessed and the specificity of KOR localization to primarily axon terminals in the NAc. Ongoing work is aimed at identifying the role PVT KOR and MOR play in modulating reward-seeking and aversive behaviors. Together, this work uncovers a potential mechanism for how KOR and MOR may have different effects on behavior by driving differing degrees of neural inhibition from separate PVT subcompartments and circuits.

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Poster

PSTR259: All About GPCRs

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.09/B29

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Characterization of NTX94827 a Novel mGlu7 Negative Allosteric Modulator in Preclinical Pharmacological Models

Authors: C. MOMBEREAU¹, J.-L. PAPARIN¹, C. FERRY¹, A. JAVAULT¹, D. S. DUPUIS¹, L. PARK², J.-P. ROCHER¹, M. KALINICHEV¹, ***R. LUTJENS**¹;

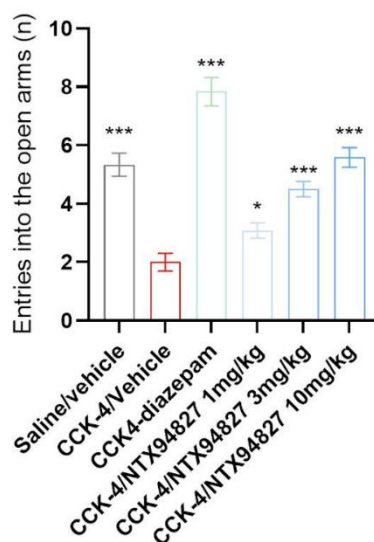
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Abstract: The role of mGlu7 receptors remains partly elusive - while it is the most widely expressed from all eight subtypes of metabotropic glutamate receptors, it is also the one with the lowest affinity for glutamate. Its highly specialised expression in active zones of synapses of glutamatergic and gabaergic neurons, its autoreceptor activity inhibiting neurotransmitter release when activated, suggest mGlu7 plays a key role in modulating neuronal transmission, especially under high activity conditions. While mGlu7 is postulated to be an attractive therapeutic target for numerous neurological and psychiatric disorders, including anxiety and stress disorders (Fendt 2008, Flor 2012, Kalinichev 2013, Golubeva 2016)^{1,2,3,4} schizophrenia (Cieřlik, 2018)⁵, depression (Wierońska 2019)⁶, and pain (Moloney 2015)⁷, there has been a chronic lack of potent, selective and brain-penetrant ligands to allow an appropriate characterisation of mGlu7 function and its potential as treatment for indications with high unmet medical need. Here we describe the profile of NTX94827, a novel mGlu7 negative allosteric modulator (NAM) identified using Neurosterix' unique allosteric modulation drug discovery platform. NTX94827 demonstrated potent mGlu7 inhibition in 3 different in vitro pharmacological readouts (Table 1), a high degree of selectivity and a PK profile showing it is metabolically stable, bioavailable and brain penetrant. In preclinical models of anxiety and PTSD, it showed a dose-dependent efficacy following oral administration (Figure 1), in good alignment with measured concentration in the brain. Taken together, these data help understanding the function of mGlu7 receptors in anxiety neurocircuitry and validate the NAM approach as a potential new treatment for stress-related disorders. This work has been seminal for the identification of the first-in-class clinical candidate which is currently progressing towards IND-enabling studies.

Table 1 *in vitro* pharmacological profile

Compound	FLIPR Ca ²⁺ Assay		cAMP Assay
	Hum mGlu7	Rat mGlu7	Hum mGlu7
	IC ₅₀ – nM (% Inh vs. L-AP4 max)	IC ₅₀ – nM (% Inh vs. L-AP4 max)	IC ₅₀ – nM (% Inh vs. Glu max)
ADX71743	51 ± 13 (86)	88 ± 9 (101)	22 ± 4 (124)
MMPIP	20 ± 6 (65)	42 ± 7 (41)	41 ± 4 (134)
ADX94827	37 ± 6 (95)	20 ± 4 (137)	79 ± 3 (90)

1. Fendt et al. *Mol. Psychiatry* **2008**, 13, 970–979.
2. Flor, et al. *Neuropharmacology* **2012**, 62, 1619–1626.
3. Kalinichev et al. *J. Pharmacol. Exp. Ther.* **2013**, 344, 624–636.
4. Golubeva et al. *Curr. Drug Targets* **2016**, 17, 538–616
5. Cieslik et al. *Drugs. Front. Mol. Neurosci.* **2018**, 11, 316.
6. Wierońska, J.M.; Pilc, A. *Pharmacol. Ther.* **2019**, 193, 75–82.
7. Moloney et al. *Neurobiol. of Stress* **2015**, 28.

Figure 1: Number of entries into the open arms

Graph shows mean ± s.e.m. of n=12 rats
 * $p \leq 0.05$, as compared to CCK4/Vehicle group
 *** $p \leq 0.001$, as compared to CCK4/Vehicle group

Disclosures: **C. Mombereau:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **J. Paparin:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **C. Ferry:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **A. Javault:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **D.S. Dupuis:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **L. Park:** A. Employment/Salary (full or part-time): Naason Science. **J. Rocher:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **M. Kalinichev:** A. Employment/Salary (full or part-time): Neurosterix Pharma. **R. Lutjens:** A. Employment/Salary (full or part-time): Neurosterix Pharma.

Poster

PSTR259: All About GPCRs

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.10/B30

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NSERC Grant RGPIN-2019-04989

Title: Neurosteroids modulate muscarinic acetylcholine responses in the mouse medial prefrontal cortex

Authors: **D. ZENG**, G. LEE, *C. D. BAILEY;
 Dept. of Biomed. Sci., Univ. of Guelph, Guelph, ON, Canada

Abstract: The neurotransmitter acetylcholine acts within the rodent medial prefrontal cortex (mPFC) to support cognitive functions and goal-directed behaviour. Acetylcholine fulfills this role by activating its nicotinic and muscarinic classes of receptors, which influence the function of neurons that send efferent signals from this brain region. Recent work in non-neuronal expression systems has shown that progesterone-based neurosteroids, which are formed within the brain *de novo*, allosterically modulate muscarinic receptor (mAChR) function. It is not known whether these neurosteroids modulate the function of native mAChRs in living neurons. We addressed this question using whole-cell electrophysiology in mPFC layer V pyramidal neurons of juvenile age mice to determine whether progesterone (100 nM and 1 μ M) and its metabolite allopregnanolone (ALLO; 10 nM and 100 nM) modulate mAChR-induced neuron excitability responses. Progesterone had no effect at 100 nM but potentiated mAChR responses at 1 μ M in males only. ALLO potentiated mAChR responses at 10 nM and 100 nM in both sexes. The ability of ALLO to potentiate mAChR responses persisted in the presence of antagonists to GABA_A, AMPA, kainate, and membrane progesterone receptors, suggesting that it does not act indirectly through any of these receptors. Radioligand binding assays showed that neither progesterone nor ALLO potentiate agonist binding to mAChRs, suggesting that they modulate mAChR responses via mechanisms downstream of the receptor itself. Overall results from this study demonstrate that progesterone and ALLO potentiate mAChR responses in mPFC output neurons, suggesting that these endogenous neurosteroids may act at this site to influence acetylcholine-dependent cognitive functions.

Disclosures: D. Zeng: None. G. Lee: None. C.D. Bailey: None.

Poster

PSTR259: All About GPCRs

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Program #/Poster #: PSTR259.11/B31

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Impact of 5-HT₇ receptor antagonist treatment on the BTBR mouse model of autism spectrum disorder

Authors: *D. A. AMODEO¹, P. FLORES³, M. R. GONZALES⁵, A. GUTIERREZ⁴, R. HERNANDEZ^{1,2}, M. JURADO², O. ROMERO DIAZ²;
²Psychology, ¹California State Univ. San Bernardino, San Bernardino, CA; ⁴Psychology,
³California State University, San Bernardino, San Bernardino, CA; ⁵Cal State Univ. San Bernardino, San Bernardino, CA

Abstract: The 5-HT₇ receptor has become a target for novel therapeutics. The current study aims to address these gaps within the literature by examining the effects of the 5-HT₇ antagonist SB 269970 on core features of autism spectrum disorder. Specifically, we aim to examine how 5-HT₇ receptor antagonist treatment impacts restricted, repetitive behaviors (RRBs) and social impairments present in a mouse model of ASD. To do so, male and female C57BL/6J and BTBR

mice received systemic injections of either 0, 1, or 5 mg/kg SB269970 treatment before measures of repetitive self-grooming and the social interaction task. Although there were no significant differences between the two doses, mice treated with 1.0 SB269970 expressed lower levels of repetitive self-grooming. Neither dose impacted social impairment found in BTBR mice.

Disclosures: D.A. Amodeo: None. P. Flores: None. M.R. Gonzales: None. A. Gutierrez: None. R. Hernandez: None. M. Jurado: None. O. Romero Diaz: None.

Poster

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Program #/Poster #: PSTR259.12/B32

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: CNPq
Capes
Faperj
INNT

Title: Adenosine A1 receptors modulate different signaling pathways by transactivating Epidermal Growth Factor receptors in the early developing retina

Authors: M. GUIMARÃES-SOUZA¹, L. G. R. XIMENES¹, J. PEREIRA-DE-LIMA², A. DOS SANTOS-RODRIGUES³, E. GIESTAL-DE-ARAÚJO⁴, *R. PAES-DE-CARVALHO²;

¹Program of Neurosciences, ²Neurobio., Fluminense Federal Univ., Niterói, Brazil; ³Program of Neurosciences and Dept. of Neurobio., Fluminense Federal Univ., Niterói, Brazil; ⁴Program of Neurosciences and Dept. of Neurobio., Fluminense Federal Univ., Niterói, Brazil

Abstract: Adenosine receptors are expressed in the chicken embryo retina and changes in cyclic AMP triggered by these receptors are observed during development, regulating several signaling pathways and transcription factors, including CREB, which is involved in developmental processes such as neuron survival and differentiation. Here we studied the regulation of CREB as well as ERK and AKT phosphorylation by A1 adenosine receptors in the developing chicken retina. Retinas from 10-day-old chicken embryos (E10), an early developmental stage, and E16, a more developed stage, were dissected and stimulated in Hank's saline with the adenosine A1 receptor agonists cyclohexyladenosine (CHA, 100nM), or cyclopentyladenosine (CPA, 100 nM), or with the Gi activator Mastoparan (MAST, 50µM). After the incubation period, retinas were lysed and analyzed by western blotting using antibodies against phospho-CREB, ERK and AKT and respective total proteins. Adenosine A1 receptors are negatively coupled to adenylyl cyclase but, surprisingly, the A1 agonist CHA stimulates CREB phosphorylation in E10 retinas in a way dependent on Src kinase, PKC and MEK/ERK. Interestingly, MAST, a peptide capable of directly activating the Gi protein, also increases the phosphorylation of CREB, as well as ERK and AKT, suggesting that inhibition of cAMP accumulation plays an important role in this effect.

On the other hand, CHA promotes an inhibition of CREB, ERK and AKT phosphorylation in E16. The effect of CHA on CREB activity in E10 is blocked by the A1 antagonist DPCPX or by Thyrphostin AG 1478, an inhibitor of Epidermal Growth Factor receptor activity, suggesting a transactivation of this receptor mediated by A1 receptors in this stage of development. Our results indicate that adenosine A1 receptors differentially modulate CREB, ERK and AKT activity in developing retinas. The A1 receptor modulates CREB activity in E10 retinas in an EGF receptor transactivation-dependent manner and through a pathway involving SRC/PKC/ERK. In E16 retinas, the A1 receptor promoted an inhibitory effect on CREB, ERK and AKT, suggesting a shift in signaling pathways during retinal development.

Disclosures: M. Guimarães-Souza: None. L.G.R. Ximenes: None. J. Pereira-de-Lima: None. A. dos Santos-Rodrigues: None. E. Giestal-de-Araujo: None. R. Paes-de-Carvalho: None.

Poster

PSTR259: All About GPCRs

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Program #/Poster #: PSTR259.13/B33

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH Grant F32MH136662

Title: Decoding the logic of neuromodulation: Functional diversity among G protein coupled receptors co-expressed in hippocampal neurons

Authors: *J. X. HE, M. VON ZASTROW;
Psychiatry, Univ. of California San Francisco, San Francisco, CA

Abstract: While neuroscience historically focused on anatomical connections between neurons, we now recognize that there is an enormous amount of chemical communication layered over synaptic networks mediated by G protein-coupled receptors (GPCRs). In fact, recent single cell transcriptomic studies make clear that individual neurons co-express a huge diversity of such receptors, suggesting that individual neurons have the potential to respond to a greater diversity of neuromodulatory ligands than previously recognized. The complexity of this “neuromodulatory network” is staggering and opens the question - how does a neuron ‘know’ which of its many co-expressed receptors was activated? I hypothesize that an individual neuron can detect a multitude of neuromodulatory ligands and can distinguish between neuromodulators despite their cognate GPCRs coupling to similar transduction pathways. Using primary cultures of hippocampal neurons, where there’s been a great diversity of GPCRs described, I expressed biosensors for cAMP and PKA and assessed fluorescence increases upon bath application of selective agonists for Gs-coupled receptors. I found that essentially all neurons in the culture show cAMP responses to agonists of 4 different Gs-coupled receptors (B1-AR, D5R, VIPR1, and CRHR1) indicating that they are functionally co-expressed in these cells.

At the level of PKA activity, the receptors begin to diverge, with D5R producing less global PKA and almost no nuclear PKA activity compared to the other receptors. One of the functional outcomes of Gs-coupled receptor signaling is to increase gene transcription, so I developed a transcriptional reporter assay where GFP is produced following phosphorylation of the transcription factor CREB. Despite producing similar cAMP responses, stimulation of VIPR1 drives a dramatic increase in GFP expression, over twice as much as stimulation of the other receptors. I am presently working on determining what makes VIPR1 uniquely such a strong transcriptional regulator.

These data confirm my hypothesis that not all Gs-coupled receptors are made equal, showcasing the depth of information the neuromodulatory network can communicate and the substantial role that a single neuromodulator can have on long term changes in the neural circuit.

Disclosures: J.X. He: None. M. von Zastrow: None.

Poster

PSTR259: All About GPCRs

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR259.14/B34

Topic: B.01. Transmitters, Transporters, and Other Signaling Molecules

Support: NIH niddk R01 120824

Title: Tonic Inhibition of Colorectal Afferent Mechanotransduction by Group III Metabotropic Glutamate Receptor mGluR7

Authors: *N. DEIERLEIN¹, B. FENG², J. LIU²;

¹Univ. of Connecticut Biomed. Engin. Dept., Storrs, CT; ²Biomed. Engin., Univ. of Connecticut, Storrs, CT

Abstract: Chronic visceral pain, which characterizes the prevalent disorders of gut-brain interactions (DGBI), is typically triggered by mechanical distension of hollow visceral organs like the distal colon and rectum (colorectum). All extrinsic afferents innervating the colorectum are glutamatergic excitatory neurons expressing a full spectrum of ionotropic and metabotropic glutamate receptors (iGluR and mGluR) at their sensory endings in all layers of the intestinal wall except the serosa. In addition, colorectal glutamate signaling is strongly indicated by the presence of glutamatergic myenteric neurons that project to virtually every myenteric ganglion. In this study, we focused on the role of mGluR7, a Gi-coupled inhibitory receptor with the highest expression level among all glutamate receptors in colonic afferent neurons. By conducting extracellular single-fiber recordings in an ex vivo colorectum-nerve preparation, we showed that intracolonic MMPIP, a selective mGluR7 antagonist, not only triggered spontaneous spiking of colorectal afferents but also enhanced the neural encoding of colorectal stretch and distension. In addition, blocking the L-type calcium channels with nifedipine abolished MMPIP's effect in triggering spontaneous spiking of colorectal afferents but did not completely negate its

effect in sensitizing the afferents to colorectal stretch. On isolated longitudinal colorectal strips, MMPIP increased the frequency and magnitude of spontaneous muscle contractions. These compelling findings strongly indicate the tonic inhibition of colorectal afferents by inhibitory glutamatergic signaling via mGluR7, partly through the inhibition of longitudinal smooth muscle tone. Targeting mGluR7 could offer a promising therapeutic approach for managing visceral pain in DGBI.

Disclosures: N. Deierlein: None. B. Feng: None. J. Liu: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.01/Web Only

Topic: B.04. Synaptic Transmission

Title: Comparisons in the responses to reduced extracellular Ca^{2+} on membrane potential and hyper excitability of neurons among various model preparations

Authors: *M. A. E. DE CASTRO¹, Y. KIM³, J. KIM⁴, R. L. COOPER²;

¹Biol. Dept., ²Dept Biol, Univ. of Kentucky, Lexington, KY; ³Gatton Acad., Richmond, KY;

⁴Model Lab. Sch., Richmond, KY

Abstract: Membrane potential provides a driving force for movement of ions such as K^+ , Na^+ , and Ca^{2+} , thus allowing membranes to be excitable. A steady state or resting membrane potential is maintained by many factors such as ionic leak channels, pumps, and exchangers. Many cells have a resting membrane potential directed toward the equilibrium potential of the K^+ ion (E_K) due to a high permeability to K^+ but are usually slightly depolarized due to a small Na^+ leak into cells through the Na^+ leak channel (NALCN). At rest, membranes generally have a low permeability to Ca^{2+} ; thus, the permeability to Ca^{2+} likely has a minor role contributing to the resting membrane potential. However, extracellular Ca^{2+} blocks the NALCN channel, thus driving the membrane potential into a hyperpolarized state. Removal of extracellular Ca^{2+} results in a depolarized state since Na^+ leak is enhanced. In each cell type, and even along the membrane of one cell, the distribution and density of NALCN channels can vary, thus presenting local differences in the effects of Ca^{2+} on the membrane potential. To compare the effects of reduced Ca^{2+} on membrane potential, membrane potential was measured in the crayfish (*Procambarus clarkii*) opener muscle, larval *Drosophila melanogaster* neuromuscular junction, and blue marine crab. The motor neurons of both preparations appeared to depolarize and caused an increase in spontaneous quantal synaptic events in lowered extracellular Ca^{2+} . However, raised extracellular Ca^{2+} had a greater effect on the membrane potential in *Drosophila* than in crayfish ($p < 0.05$, t-test). Sensory neurons from a marine crab and freshwater crayfish also become hyperexcitable with lowered extracellular Ca^{2+} ($p < 0.05$, t-test). This is likely due to the neurons being depolarized. However, it appears that neurons do not acutely recover well after being exposed to zero extracellular Ca^{2+} . Computational simulations assessing the effect of extracellular Ca^{2+} on

the summed permeability for NALCN channels are aiding in developing quantifiable measures for models based on empirical data.

Disclosures: M.A.E. de Castro: None. Y. Kim: None. J. Kim: None. R.L. Cooper: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.02/Web Only

Topic: B.04. Synaptic Transmission

Support: Beckman Fellow (KB)
Chellgren Fellow (KN)

Title: Modulation of enhancing synaptic transmission by serotonin or by lipopolysaccharides (LPS) at glutamatergic synapses appears to use different mechanisms.

Authors: *K. NEGLIA¹, J. BIDROS¹, M. DUNN², R. MCINTOSH¹, K. BROCK¹, R. L. COOPER³;

¹Univ. of Kentucky, Lexington, KY; ²Univ. of Kentucky, Union, KY; ³Dept Biol, Univ. of Kentucky, Lexington, KY

Abstract: Recent evidence demonstrated that lipopolysaccharides (LPS) can have direct effects on membrane potential of cells independent of an immune response. It now appears that direct action of LPS (≥ 250 $\mu\text{g/ml}$) on glutamatergic synapses at the crayfish neuromuscular junction (NMJ) primarily enhances evoked synaptic transmission but not spontaneous vesicle fusion events. It is well known that serotonin (5-HT, 100 nM to 1 μM) increases spontaneous vesicle fusion and evoked transmission at the crayfish NMJ. 5-HT increased evoked and spontaneous transmission ($p < 0.5$, Paired t-Test; $n = 6$) but LPS only enhanced evoked when the concentration exceeded 100 $\mu\text{g/ml}$ and in a dose -dependent manner ($p < 0.5$, Paired t-Test; $n = 6$ and Sign test non-parametric). It appears 5-HT is mediated via 5-HT 2A-like receptors and then activating an IP3 induced secondary messenger cascade to promote vesicle docking as well as increase the occurrence of spontaneous quantal events. While LPS is activating a K2P - potassium channel, hyperpolarizing the presynaptic nerve terminal and potentially removing residue voltage-gated Na⁺ channels inactivation. Thus, promoting a larger driving gradient for Ca²⁺ and Na⁺ ions during depolarization of the terminal during evoked action potentials. The presence of Cd²⁺ (1 mM) blocks the evoked transmission without retarding the frequency of spontaneous quantal events. The frequency of quantal events still increases in the present of 5-HT and Cd²⁺. The frequency of quantal events before and after LPS with or without Cd²⁺ is not significantly altered.

Disclosures: K. Neglia: None. J. Bidros: None. M. Dunn: None. R. McIntosh: None. K. Brock: None. R.L. Cooper: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.03/B35

Topic: B.04. Synaptic Transmission

Support: NIH R01MH118258

Title: Ghrelin increases glutamate release at the perforant path-dentate gyrus granule cell synapses by cross-activating dopamine D₁ receptors

Authors: C. ORAEGBUNA, *S. LEI;
Univ. of North Dakota, Grand Forks, ND

Abstract: Ghrelin is a multifunctional peptide hormone with receptors present in various brain tissues including hippocampus and has been associated with neuroprotection, neuromodulation, and memory processing. Ghrelin is an endogenous ligand for growth hormone secretagogue receptor (GHSR1a). Here we studied the roles and mechanisms of ghrelin in glutamatergic transmission at the perforant path (PP)-granule cells (GCs) in the dentate gyrus by recording AMPA EPSCs in hippocampal slices cut from both male and female C57BL/6J mice. Our results showed that ghrelin concentration-dependently elicited a persistent enhancement of AMPA EPSCs. The ghrelin-induced augmentation of glutamatergic transmission was mediated by increasing presynaptic glutamate release as supported by the following results; ghrelin decreased the coefficient of variation (C. V.) and paired-pulse ratio; ghrelin increased NMDA EPSCs as well; ghrelin enhanced the frequency with no effect on the amplitude of mEPSCs. Ghrelin enlarged the size of readily releasable pool and increased release probability. Ghrelin-elicited increases in glutamate release did not require the functions of G α q-phospholipase C pathway and G α i, but was dependent on G α s and cAMP/EPAC/PI3K pathway. Our results further indicated that ghrelin-induced enhancement of glutamate release was mediated by heterodimerization with dopamine D₁ receptors.

Disclosures: C. Oraegbuna: None. S. Lei: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.04/B36

Topic: B.04. Synaptic Transmission

Support: NIH R00 MH118425
NARSAD BBRF Young Investigator Award
T32MH119049
T32NS04554016
UCI - Eugene Cota-Robles Fellowship

Title: Assessing the function of the putative neuropsychiatric disorder-associated protein MDGA1 in the mouse brain

Authors: *M. SANDOVAL, G. SANDOVAL, G. FRANCHET-SCHAER, M. LU, J. DÍAZ-ALONSO;
Anat. & Neurobio., UC Irvine, Irvine, CA

Abstract: The MAM domain-containing glycosylphosphatidylinositol (GPI) anchor protein 1 (MDGA1) is a completely extracellular, GPI-anchored cell adhesion molecule. Human MDGA1 gene mutations have been associated with neuropsychiatric disorders including major depressive disorder. In mice, global MDGA1 knockouts (KO) show impaired excitation/inhibition balance and defects in cognitive function. However, despite the growing research interest in MDGA1 synaptic function, precise investigation of MDGA1 function within specific brain subregions has been limited. Previously, we have identified MDGA1 immunoreactivity within the lateral habenula (LHb), a region that shows hyperactivity in individuals with depression. Here, we assess behavioral changes with Cre-mediated MDGA1 region-specific KO of adult MDGA1^{f/f} mice. Importantly, the LHb contains majority excitatory neurons on which multifarious neuromodulatory, excitatory, and inhibitory projections across the prefrontal cortex and other subcortical regions impinge. We hypothesized that deletion of MDGA1 from LHb would lead to effects associated with depressive-like states in mice. To this end, in the absence of a general LHb Cre-driver line, MDGA1 was significantly reduced in LHb neurons of MDGA1^{f/f} mice infected with AAV-delivered Cre (AAV Cre-GFP) compared to GFP-containing control AAVs (AAV GFP) (n = 5 mice/virus). To assess MDGA1 function behaviorally and in association with depressive-like states in mice, we subjected both adult (2-3 months old) AAV Cre-GFP and AAV GFP control groups to two behavioral paradigms: i) tail suspension test (TST) and ii) forced swim test (FST). The removal of MDGA1 from LHb resulted in a significant decrease in the latency to immobility on the FST compared to AAV GFP infected mice but no change in the overall time immobile (n = 5 mice/virus). Surprisingly, both measures were unchanged during TST. To understand cellular activation as a result of MDGA1 KO, we assessed cFos expression in LHb of both sets of mice. AAV Cre-GFP infected mice showed both significantly higher cFos⁺ cells, and a significant increase in the overlap between GFP⁺ and cFos⁺ cells in the LHb. Taken together, these results suggest that removal of MDGA1 leads to the cell autonomous hyperexcitaiton of LHb neurons and therefore increased propensity to despair in adult mice. Ongoing experiments seek to address the underlying electrophysiological changes associated with LHb MDGA1 KO-mediated cellular hyperactivation via whole cell patch clamp in acute brain slices in adult mice. Overall, our results present a novel possible molecular mechanism in which MDGA1 may protect against depression susceptibility.

Disclosures: M. Sandoval: None. G. Sandoval: None. G. Franchet-Schaer: None. M. Lu: None. J. Díaz-Alonso: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.05/B37

Topic: B.04. Synaptic Transmission

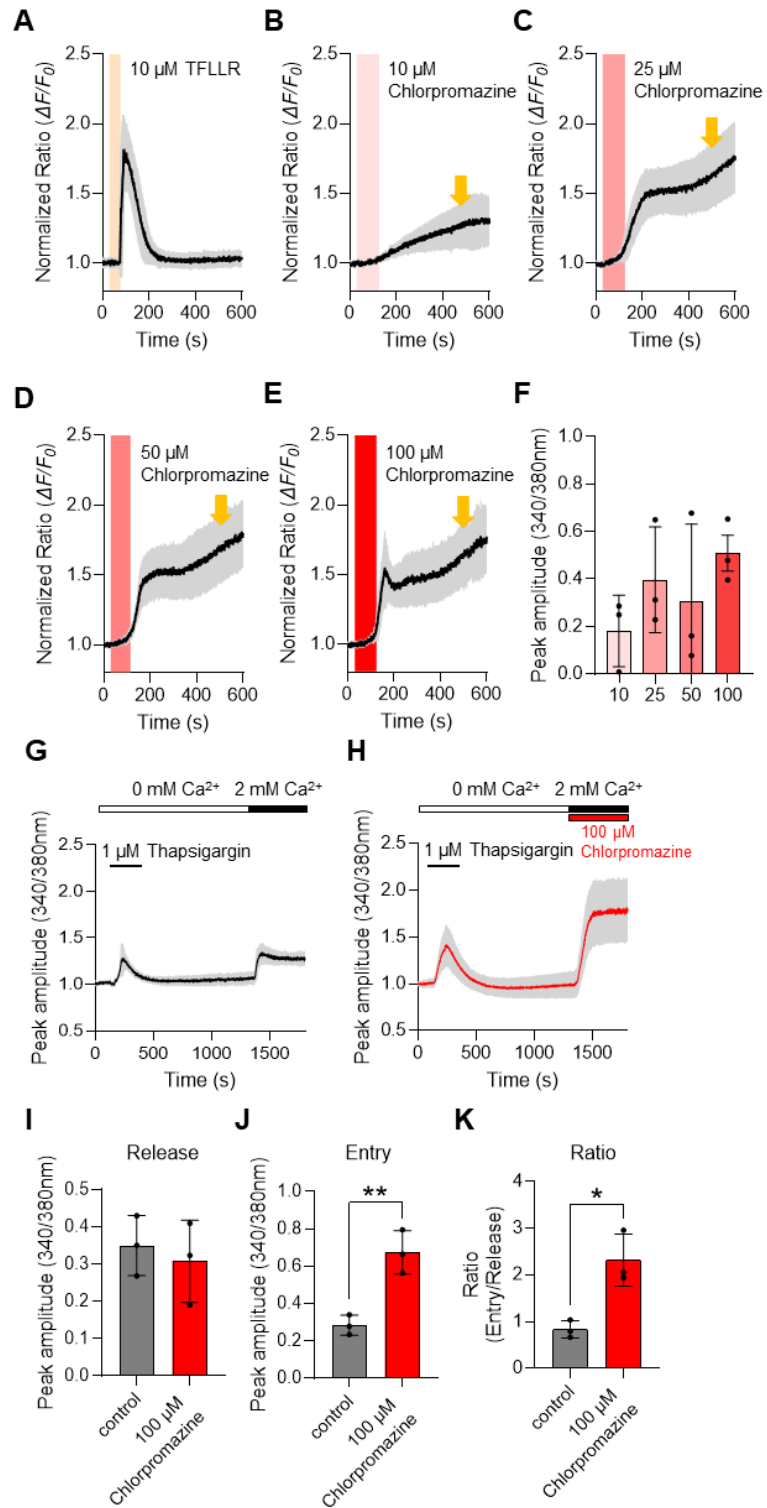
Support: NST Grant No. GTL24021-400

Title: Chlorpromazine increases the frequency of EPSC by non-canonical Ca^{2+} elevation of astrocytes

Authors: *H.-W. LIM;

Korea Inst. of Toxicology, Daejeon, Korea, Republic of

Abstract: Chlorpromazine, a widely used antipsychotic for schizophrenia, exerts its effects primarily via dopamine D2 receptor antagonism. However, the precise mechanism by which chlorpromazine acts in the brain is not fully understood. In this study, we investigated the effects and mechanisms of chlorpromazine on synaptic transmission. To investigate the effect of chlorpromazine on the glutamatergic synaptic transmission, we performed spontaneous excitatory postsynaptic currents (sEPSC) in rat primary cortical neurons and miniature EPSCs (mEPSC) in hippocampal CA1 pyramidal neurons. Chlorpromazine increases the frequency of sEPSC and mEPSC. The increase in mEPSC frequency is closely associated with astrocyte. We utilized fura-2 imaging to determine that astrocytic Ca^{2+} is responded to chlorpromazine. Chlorpromazine showed Ca^{2+} elevation by concentration. SKF96365, a TRPC channel inhibitor, and HC030031, a TPRA1 antagonist, were used, and Ca^{2+} levels increased in astrocytes. Additionally, chlorpromazine increased astrocytic Ca^{2+} in external 0 mM Ca^{2+} , suggesting that chlorpromazine increases intracellular Ca^{2+} level of extracellular Ca^{2+} . We utilized store-operated calcium entry (SOCE) to see whether chlorpromazine relates to SOCE. Chlorpromazine showed the additional Ca^{2+} elevation on SOCE, suggesting that chlorpromazine-increased Ca^{2+} is irrelevant to ER Ca^{2+} . After ER Ca^{2+} depletion, Ca^{2+} entry by chlorpromazine was increased when external 0 mM Ca^{2+} , demonstrating that chlorpromazine-increased Ca^{2+} elevation does not require extracellular Ca^{2+} . Chlorpromazine did not change the frequency with NPS2390, group I mGluR1/5 antagonist. These findings suggest that astrocytic glutamate release by chlorpromazine-mediated non-ER Ca^{2+} elevation enhance the presynaptic glutamate release through the activation of group I mGluR1/5. Our results provide the strategy for reduction of side effect of chlorpromazine.



Disclosures: H. Lim: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.06/B38

Topic: B.04. Synaptic Transmission

Title: Sva-containing lncrna ak057321 regulates synaptic development and transmission in ipsc-derived glutamatergic neurons

Authors: *J. LENTINI¹, Y. HUO^{3,2}, K.-Z. DUAN², Y. NONG¹, F. RANDI¹, Y. BAI¹, N. NEGRON¹, A. FOX¹, N. ALESSANDRI-HABER², S. D. CROLL², M. P. ANDERSON⁴; ²Neurosci., ¹Regeneron Pharmaceuticals, Tarrytown, NY; ³Biol., Boston Univ., Boston, MA; ⁴Pathology, Harrington Discovery Inst., Cleveland, OH

Abstract: Despite the 98.7% shared coding sequence between humans and chimpanzees, profound species differences exist (The Chimpanzee Sequencing and Analysis Consortium, *Nature* 2005). In particular, humans have a larger brain, sophisticated cognitive abilities, improved fine-motor skills and extended lifespans (Hrvovj-Mihic et al., *Front Hum Neurosci* 2013). Expansion of the human cortex may underlie humans' enhanced cognitive capabilities, at least in part due to the delayed maturation of the human brain (Lindhout et al., *Nature* 2024). In addition, human neurons have specialized complex processes, increased synapse numbers and greater connectivity (Lindhout et al., *Nature* 2024; Sherwood et al., *Cerebral Cortex* 2020). However, the mechanisms underlying the complexity of the human brain circuitry remains elusive. In humans, many functionally important genes underlying proper neurodevelopment harbor intronic Sine-VNTR-Alu (SVA) retrotransposable elements. SVA elements arose within the hominid lineage along evolution and further expanded in the human genome to create novel, human-specific elements within numerous key neurodevelopmental genes (Nadler et al. *Commun Biol* 2023). Nadler et al. (2023) identified a unique hominid-specific regulatory mechanism whereby human-specific intronic SVA elements within neurodevelopmental genes inhibits gene expression until an upregulation of an SVA-containing lncRNA, *AK057321*, de-represses the inhibition. This mechanism has been demonstrated to effectively upregulate several key SVA-containing genes during early neuronal differentiation in N-Tera2 cells (Nadler et al. *Commun Biol* 2023). Our current work seeks to understand the role of SVA-containing lncRNA *AK057321* in human neural development. We found that lncRNA *AK057321* is expressed in human glutamatergic and GABAergic iPSC-derived neurons, but not in other neural cell types. After administering a lentiviral shRNA to reduce the expression of *AK057321* in developing glutamatergic neurons, we found a decrease in colocalized puncta of pre- and post-synaptic markers without an overall change to dendritic morphology. The synaptic deficits were further demonstrated by a reduction in the frequency of spontaneous excitatory post-synaptic currents in a human iPSC-derived neuron-astrocyte co-culture without concomitant changes in neuronal membrane properties. Our work highlights a novel role of a unique SVA-lncRNA in synaptic development and transmission in human neurons.

Disclosures: J. Lentini: A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent

holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **Y. Huo:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **K. Duan:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **Y. Nong:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **F. Randi:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **Y. Bai:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **N. Negron:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **A. Fox:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **N. Alessandri-Haber:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **S.D. Croll:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **M.P. Anderson:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.07/B39

Topic: B.04. Synaptic Transmission

Support: NIH intramural support for Dr. Lovinger

Title: Spinal glycine receptor $\alpha 1$ subunits coordinate startle response through a cell-type specific mechanism

Authors: *S. WEI¹, J. XU¹, Y. XU², D. M. LOVINGER¹, L. ZHANG¹;

¹NIAAA/NIH, Rockville, MD; ²Anesthesiol., Univ. Pittsburgh Sch. Med., Pittsburgh, PA

Abstract: Glycine receptor $\alpha 1$ subunits (GlyR $\alpha 1$ s) are the most abundant inhibitory ligand-gated ion channels expressed in the spinal cord and brainstem at the adult stage. These receptors mediate the majority of inhibitory neurotransmission. Numerous naturally occurring point-mutations in GlyR $\alpha 1$ result in loss of function and exaggerated startle response in human and animals. While brainstem GlyR $\alpha 1$ function is the primary focus of startle research, the role of spinal GlyR $\alpha 1$ in exaggerated startle response remains unreported. To address this question, we crossed GlyR $\alpha 1$ flox mice with choline acetyltransferase (ChAT)-Cre, glycine transporter2 (GlyT2)-Cre, or calmodulin kinase II alpha (CamKII α)-Cre lines to selectively delete GlyR $\alpha 1$ subunits in a cell-type specific manner. Deletion of GlyR $\alpha 1$ from each cell type specifically reduced the colocalization of GlyR $\alpha 1$ subunit with these respective cells, as revealed by RNAscope measurements. ChAT expression was restricted to in spinal ventral horn, specifically within laminae IX and VII. On the other hand, CamKII α overlapped with GlyT2 positive cells, which were widely distributed across spinal gray matter. Deletion of GlyR $\alpha 1$ subunits from ChAT-positive neurons resulted in exaggerated startle response induced by both acoustic sound and tactile stimuli, consistent with increased the startle response observed in global GlyR $\alpha 1$ deficient mice. In contrast, mice lacking GlyR $\alpha 1$ in either GlyT2-positive or CamKII α -positive neurons exhibited reduced startle responses to the same stimuli. Acoustic startle stimulation induced cFos expression in ChAT-positive neurons. Selective deletion of GlyR $\alpha 1$ in ChAT- or GlyT2-positive cells produced opposing effects on cFos expression. ChAT-specific GlyR $\alpha 1$ deletion enhanced cFos activation, whereas GlyT2-specific GlyR $\alpha 1$ deletion suppressed it. Finally, we performed intraspinal injection to deliver either cytomegalovirus (CMV)-Cre or CamKII α -Cre virus into GlyR $\alpha 1$ flox mice. Mice receiving CMV-Cre virus exhibited an enhanced startle response, while CamKII α -Cre-virus injected mice displayed a reduced startle response. Collectively, these findings demonstrate that spinal GlyR $\alpha 1$ subunits regulate startle response through a coordinated cell-type specific mechanism.

Disclosures: S. Wei: None. J. Xu: None. Y. Xu: None. D.M. Lovinger: None. L. Zhang: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.08/Web Only

Topic: B.04. Synaptic Transmission

Support: NIH Grant R01DC011099

Title: In-vivo imaging of a vertebrate electrical synapse reveals an active gap junction channel turnover

Authors: *D. MARTINEZ CASTILLO, A. E. PEREDA;
Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Electrical synaptic transmission is mediated by intercellular channels made of connexins proteins, which cluster at cell-cell structures known as gap junctions (GJs). While providing continuous communication, GJs are dynamic structures at which channels actively turnover. Club Endings (CE) are a group of identifiable mixed, electrical and chemical, synaptic terminals located on the lateral dendrite of Mauthner cells, a pair of reticulospinal neurons that mediate tail-flip escape responses in fish. Because of their size, experimental access and functional identifiability, these contacts are considered a model for exploring vertebrate electrical transmission. Structural/functional correlation in goldfish using EM and manipulation of exo-endocytosis suggested the presence of a fast turnover of GJs channels at these terminals. This turnover was required to maintain electrical transmission, indicating its modulation might underlie plastic changes in synaptic strength. To directly expose this phenomenon, we took advantage of the amenability of zebrafish larvae (5-8 days post fertilization) for in-vivo imaging. We used a transgenic line in which connexin 34.1, one of the connexins present at these terminals, is tagged with GFP. In order to directly monitor and quantify the active turnover of channels at CEs we performed FRAP with the help of a 2-photon microscope. Using this approach, we obtained recovery rates which on average yield a time constant of 5.89 hours (n=18), corresponding to a half-life of 4.08 hours, a value consistent with our previous estimates in goldfish and those observed for GJs in general. Remarkably, this approach also revealed a wide variability in recovery rates among different CEs, suggesting GJs at these terminals might co-exist at different functional states. Thus, our in-vivo imaging of an identifiable electrical synapse confirms that channels at neuronal GJs actively turnover. The fact that this phenomenon can exist at varying rates, suggests it might be used to control the number of GJ channels present at electrical synapses and potentially underlie plastic regulation of their synaptic strength.

Disclosures: D. Martinez Castillo: None. A.E. Pereda: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.09/B40

Topic: B.04. Synaptic Transmission

Support: R01DC011099

Title: Association of dendritic mitochondria to auditory mixed synapses in the Mauthner cell

Authors: *J. KALRA, A. E. PEREDA;

Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Mitochondria at synaptic terminals are essential in supporting neurotransmitter release and maintaining the presynaptic vesicle cycle. However, less is known about the role of dendritic mitochondria in regulating synaptic transmission. We therefore explored the distribution of mitochondria in the Mauthner cells, a valuable model for studying the properties of synaptic

transmission of identifiable synaptic inputs. For this purpose, we generated a transgenic zebrafish line in which mitochondria in the Mauthner cells are fluorescently tagged. This fish allowed us to expose the presence, number, and spatial distribution of mitochondria within this cell's processes. Rather than being homogeneously distributed, dendritic mitochondria were found to be clustered, suggesting their association with specific synaptic inputs. We explored this possibility at the lateral dendrite by co-labeling mitochondria with labeling for the neuronal gap junction channel protein Cx35, a marker of excitatory mixed (electrical and glutamatergic) synapses, or glycine receptors as a marker of inhibitory synapses. While glycine receptor labeling was uniformly distributed across the somatodendritic compartment, most mitochondria were found associated with areas additionally containing labeling for Cx35. This distribution was most noticeable at the distal portion of the lateral dendrite, where an identifiable group of ~20 large mixed terminals, known as "Club Endings" (CEs), are tightly segregated. Expansion microscopy of sections of the lateral dendrite containing these terminals allowed examining the volume and shape of mitochondria near CE contacts, which revealed the predominance of small-sized mitochondria. Because small mitochondria are thought to be linked to high energy requirements, this finding would suggest a higher energy need in the area. While the energy requirement of electrical transmission remains unknown, the observed association would be consistent with the highly dynamic turnover of channels at gap junctions observed at these terminals, which likely demands energy support. Consistent with a high number of mitochondria nearby, CEs contain an unusually high number of gap junctions (~35 per terminal, totalling 12,400 channels each) covering 80% of the synaptic contact area. Electrical and glutamatergic transmission at CEs undergo parallel activity-dependent potentiation, a phenomenon that results from interactions between these two forms of transmission. Because these interactions occur postsynaptically, dendritic mitochondria may also provide coordinated energy support for these plastic changes.

Disclosures: J. Kalra: None. A.E. Pereda: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.10/B41

Topic: B.04. Synaptic Transmission

Support: NIH Grant 1U01NS128537

Title: Repurposing naturally occurring bistable Gq opsins as optogenetic tools to modulate neuronal excitation

Authors: *D. KANKANAMGE¹, O. BERMAN², R. GOWRISHANKAR³, R. KUMAR¹, V. KALYANARAMAN¹, K. KOTHAPALLI¹, A. BERNDT⁴, M. SHALEV-BENAMI², O. YIZHAR², M. R. BRUCHAS³, B. A. COPITS¹;

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Abstract: G protein-coupled receptors (GPCRs) modulate neuronal activity by coupling to diverse downstream signaling cascades to tune behavioral states. As the most prominent family of druggable targets in the human genome, pharmacological approaches have revealed key roles for numerous GPCR families in human psychiatric conditions, including pain and substance-use disorders. However, traditional pharmacological approaches still lack spatial and temporal precision for understanding how GPCRs impact neural circuit dynamics to influence behavior. Optogenetic techniques enable more precise spatial and temporal control but have been mainly limited to ion-based tools in the brain. The generation of optoXR chimeras between the light-sensitive protein rhodopsin and GPCRs of interest has enabled optical control of neuromodulatory signaling cascades. However, their use for studying neural circuit dynamics can be limited due to irreversible activation of the bleaching pigment rhodopsin. Here, we developed and characterized two naturally occurring bistable opsins and showed their ability to excite neuronal circuits. Also, we showed that these opsins are photoswitchable opsins that mainly couple to Gq signaling cascades and are rapidly activated by pulsed violet/blue light, switched off with amber light, and effective for repeated, prolonged, and reversible activation.

Disclosures: D. Kankanamge: None. O. Berman: None. R. Gowrishankar: None. R. Kumar: None. V. Kalyanaraman: None. K. Kothapalli: None. A. Berndt: None. M. Shalev-Benami: None. O. Yizhar: None. M.R. Bruchas: None. B.A. Copits: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.11/B42

Topic: B.04. Synaptic Transmission

Support: NIH Grant MH131317

Title: Oxytocin modulates synaptic transmission and plasticity at entorhinal to dentate granule cell synapses.

Authors: *A. MARRON^{1,2}, D. H. BRAGER²;

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Abstract: Social memory is essential for navigating the world, as humans rely on social imitation to efficiently learn new skills. The neuropeptide oxytocin, implicated in social behaviors like pair bonding and emotional recognition, plays a key role in social memory via signaling in hippocampal area CA2. Since the dentate gyrus of the hippocampus provides direct excitatory input to CA2, it is important to understand how the dentate gyrus and oxytocin signaling contribute to social memory processing. Although the dentate gyrus' role in pattern

separation is well-defined, its involvement in social memory, including whether social input processing differs from other stimuli, remains unclear. Synaptic transmission from the entorhinal cortex to dentate gyrus is critical for pattern separation and may underlie social recognition memory, yet research investigating if oxytocin modulates dentate granule (DG) cell synaptic transmission is limited. To address this, we used current clamp and voltage clamp to investigate the effect of oxytocin signaling on synaptic transmission and plasticity at perforant path to dentate granule cell synapses. We found bath application of the oxytocin receptor agonist Thy⁴, Gly⁷-oxytocin (TGOT, 400 nM) increases excitatory post synaptic potential (EPSP) slope and decreases paired pulse ratio, suggesting a presynaptic mechanism of oxytocin action. Surprisingly, theta-burst pairing long-term potentiation, which is expressed postsynaptically, occludes the effect of oxytocin on EPSP slope. These results indicate oxytocin may exhibit mixed pre- and postsynaptic effects. To better separate the pre- and postsynaptic effects of oxytocin, we measured miniature excitatory postsynaptic currents (mEPSCs) before and after application of TGOT. TGOT increased the frequency, but not amplitude, of mEPSCs supporting a presynaptic effect of TGOT on synaptic transmission. These findings highlight a previously unexplored role for oxytocin in dentate gyrus transmission, suggesting a broader function for oxytocin in modulating hippocampal circuits involved in social memory.

Disclosures: A. Marron: None. D.H. Brager: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.12/B43

Topic: B.04. Synaptic Transmission

Support: Japan Society for the Promotion of Science Grants-in-Aid for Scientific Research (KAKENHI) (grant number JP24K12381)

Title: Mechanisms of 3-nitrotyrosine-mediated excitatory modulation in the rat spinal dorsal horn

Authors: *Y. KAIMOCHI¹, M. YAMANAKA², W. TANIGUCHI³, N. NISHIO³, Y. MATSUYAMA⁶, T. UENO⁴, R. MIYAKE⁴, T. NAKATSUKA⁷, H. YAMADA⁵;

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Abstract: Introduction:

3-Nitrotyrosine (3-NT) is recognized as a biomarker of nitrosative stress. Although elevated levels of 3-NT have been observed in various pain-associated conditions, there are no reports

indicating that 3-NT directly induces pain.

Objective:

To elucidate the electrophysiological effects and underlying mechanisms of 3-NT on spinal dorsal horn neurons.

Methods:

Whole-cell patch-clamp recordings were performed on spinal cord slices prepared from male Sprague-Dawley rats. Spontaneous excitatory postsynaptic currents (sEPSCs) and miniature inhibitory postsynaptic currents (mIPSCs) were measured in dorsal horn neurons. 3-NT and pharmacological agents were administered via perfusion in artificial cerebrospinal fluid.

Results:

Perfusion with 3-NT significantly increased both the frequency and amplitude of sEPSC. These effects were completely abolished by CNQX, an AMPA/kainate receptor antagonist, and by tetrodotoxin (TTX), which inhibits axonal conduction. 3-NT had no effect on mIPSC frequency but significantly reduced its amplitude. This reduction persisted in the presence of strychnine, a glycine receptor antagonist, but was abolished in the presence of bicuculline, a GABA receptor antagonist.

Discussion:

The 3-NT-induced enhancement of sEPSCs was eliminated by both CNQX and TTX, suggesting that 3-NT facilitates glutamate release via activation of excitatory interneurons. The decrease in mIPSCs amplitude in the presence of strychnine but not bicuculline indicates that 3-NT acts post-synaptically to inhibit GABA receptors. This inhibition may disinhibit excitatory interneurons, resulting in enhanced glutamate release.

Conclusion:

3-NT inhibits GABA receptors in the spinal dorsal horn, leading to reduced mIPSCs amplitude. Additionally, by disinhibiting excitatory interneurons through GABA receptor blockade, 3-NT enhances glutamate release, thereby increasing both the frequency and amplitude of sEPSCs. These findings suggest a dual mechanism by which 3-NT contributes to the modulation of excitatory transmission in the spinal dorsal horn.

Disclosures: Y. Kaimochi: None. M. Yamanaka: None. W. Taniguchi: None. N. Nishio: None. Y. Matsuyama: None. T. Ueno: None. R. Miyake: None. T. Nakatsuka: None. H. Yamada: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.13/B44

Topic: B.04. Synaptic Transmission

Support: NIH R15 DA062968

Title: A forward genetic screen to identify novel genes involved in the mechanism of action of amphetamine

Authors: *H. ALSHAREEF¹, K. POQUETTE², L. CARVELLI^{1,3,2};

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Abstract: Amphetamine (Amph) is a psychostimulant broadly prescribed for neurological conditions such as attention deficit hyperactivity disorder (ADHD) and narcolepsy. However, when taken at high doses and for prolonged periods, Amph can lead to addiction due to its ability to elevate extracellular dopamine (DA) levels in the brain's reward system. Given the complexity of the mammalian dopaminergic system, we use mutated *Caenorhabditis elegans* (*C. elegans*) strains from the Million Mutation Project (MMP) to identify novel genes that encode proteins involved in the mechanism of action of Amph. In wild-type *C. elegans*, exposure to Amph triggers swimming-induced paralysis (Swip), a behavior linked to overstimulation of the D2-like DA receptors (DOP-3), following an increase of extracellular DA. By performing a forward genetic screen, we show that the MMP strain VC20048 displayed an abnormal hyper-response to Amph, suggesting that one or more mutated genes in this strain is/are involved in the abnormal Amph-induced behavior. The VC20048 strain has 48 randomly mutated genes, including *lev-1*, which encodes for a subunit of the nicotinic acetylcholine receptor. We found that worms lacking only the *lev-1* gene showed similar behavioral changes to those observed in VC20048, including DA-dependent basal Swip and DA-independent Amph-mediated Swip. Our data suggest that *lev-1* plays a key role in the mechanism of action of Amph. To validate this claim, we are currently generating double mutants lacking expression of both *lev-1* and the tyrosine hydroxylase *cat-2* genes, or *lev-1* and *dop-3* gene. Because *cat-2* and *dop-3* are required to generate Amph-mediated and DA-dependent Swip, our experiments will determine whether *lev-1* functions independently from DA to generate Amph-induced Swip.

Disclosures: H. Alshareef: None. K. Poquette: None. L. Carvelli: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.14/B45

Topic: B.04. Synaptic Transmission

Support: NIH grant NS116852
NIH grant NS112538

Title: Optically probing the spectrum of GABAergic effects in the epileptic hippocampus

Authors: *T. SHIU¹, K. P. LILLIS³, K. J. STALEY²;

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Abstract: Hippocampal GABAergic synapses are typically associated with inhibitory function. However, our previous study demonstrated that individual interneurons could evoke GABA_A

responses with variable reversal potentials (E_{GABA}) onto the same pyramidal neuron. This was observed by puffing glutamate onto GABAergic interneurons, and simultaneously using whole-cell patch-clamp to record the E_{GABA} from neighboring cell. Notably, 12.5% interneurons exhibited E_{GABA} values more depolarized than the resting membrane potential, indicating a potentially excitatory GABA_A effect. Despite this observation, the extent and functional significance of E_{GABA} heterogeneity remain poorly characterized. Additionally, glutamate puffing lacks temporal precision and is labor intensive. To overcome these limitations, we combined optogenetics with whole-cell patch-clamp recordings to assess how individual interneurons influence nearby pyramidal neurons in organotypic hippocampal slice cultures (OHSC), an ex vivo model in which slicing-induced trauma leads to spontaneous epileptiform activity. To selectively target interneurons, we delivered a Cre-dependent AAV encoding st-CoChR, an excitatory light-gated cation channel that can be activated at 920nm, to OHSCs from P6-P8 DLX-Cre pups on the day of preparation. Recordings were performed at or after day in vitro 7. We first validated st-CoChR activation by optically stimulating and simultaneously recording from st-CoChR-expressing interneurons. Pulses of 5 ms, 10 ms, and 20 ms at 920 nm were tested; a 5 ms pulse reliably evoked single action potentials (n=3 cells). In separate experiments, we optically stimulated st-CoChR-expressing interneurons while recording from neighboring st-CoChR-negative pyramidal neurons. These recordings revealed robust IPSPs with variable amplitudes, suggesting heterogeneity in inhibitory synaptic strength and/or E_{GABA} . Future experiments will focus on stimulating individual st-CoChR-expressing interneurons in CA1 and DG while recording IPSPs and E_{GABA} from neighboring cells using gramicidin perforated-patch techniques, which preserve intracellular chloride concentrations for more accurate E_{GABA} measurements. To distinguish developmental effects from those driven by seizures, we will compare inhibitory connectivity in CA1 and DG at DIV2 and DIV10, with a subset of slices treated with kynurenic acid to suppress epileptiform activity. These studies aim to characterize the heterogeneity of inhibitory synapses and how seizures alters inhibitory signaling. Understanding this variability may inform strategies to restore inhibitory function in the epileptic brain.

Disclosures: T. Shiu: None. K.P. Lillis: None. K.J. Staley: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.15/B46

Topic: B.04. Synaptic Transmission

Support: NIH Grant R01NS116051
NIH Grant R21AG086934

Title: The protein interactome of the neuron specific gene family reveals novel pathway involvement in neuronal function

Authors: *A. SERRANO RODRIGUEZ¹, L. VEGA², S. J. WILSON³, M. OVERBY⁴, H. K. MULLER⁵, J. P. WEICK³;

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Abstract: Members of the neuron-specific gene family (NSG1-3) are small endolysosomal proteins that have been implicated in the trafficking and processing of multiple receptors and signaling proteins specifically in neurons. While much is known about how NSG1-3 interact with, and alter the cycling of AMPA receptors between intracellular endosomes and the plasma membrane during plasticity, the specific mechanisms by which NSGs regulate protein trafficking and synaptic signaling remain poorly understood. To address this, we employed proximity-dependent proteomic approach using the promiscuous biotinylating enzyme TurboID to identify the interactomes of NSG1, NSG2, and NSG3 in human stem cell-derived neurons. Our proteomic analysis revealed distinct yet overlapping protein networks that confirmed known functions of NSGs in synaptic transmission and protein trafficking. In addition, our data revealed novel pathway involvement for all family members. For example, the NSG3 interactome implicated involvement in protein processing pathways in the ER, RNA binding, and protein translation, while the largely shared interactomes of NSG1 and NSG2 were associated with axon-guidance, presynaptic membrane, GABA-ergic synaptic function. Intriguingly, our findings suggest NSGs may bind to each other in a heteromeric complex, and co-immunoprecipitation assays detected novel interaction of all NSGs with Amyloid precursor protein. Finally, we used electrophysiology to confirm the involvement of NSG1-2 in inhibitory synaptic transmission. These findings suggest a more complex role for NSGs in orchestrating protein trafficking and synaptic modulation, with implications for neurodegenerative disease mechanisms.

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Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.16/B47

Topic: B.04. Synaptic Transmission

Support: NIMH R00MH117235
Sloan Research Fellowship
BBRF Young Investigator Award

Title: The adhesion GPCR latrophilins are essential for inhibitory synaptic inputs onto D2 MSNs that integrate and process responding to acute amphetamine injections

Authors: *S. WATSON¹, B. A. GRUETER², R. SANDO¹;

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Abstract: The neuron's ability to form precise, subcellular synaptic connections with their target of interest is crucial to the generation of functional neural circuits. To this end, increasing evidence emphasizes the vital role of pre- and post-synaptic cell adhesion molecules that conduct bidirectional, trans-synaptic signaling responsible for synaptic development. In particular, latrophilin (Lphn) is a postsynaptic adhesion G protein-coupled receptor known to promote initial synapse formation in hippocampal circuits. To expand on this knowledge, we explore the role of Lphn1-3 in the dorsal mouse striatum to investigate how Lphn deletion may alter striatal synaptic connectivity and function. We generated custom L123^{flxed}-D1cre and L123^{flxed}-A2acre conditional knockout (cKO) mouse lines to explore the effects of Lphn deletion on dopamine-1 (D1) and dopamine-2 (D2) striatal medium spiny neurons (MSNs), respectively. Using 8-10-week-old mice, we discovered that L123f-A2a^{cre+} mice exhibit severe ambulatory locomotor deficits following 1mg/kg and 3mg/kg intraperitoneal injection of amphetamine, suggesting alterations in D2 MSN signaling. We then utilized wild-type A2acre-Ai14 and L123f-A2a-Ai14 mice to conduct *ex vivo* D2 MSN electrophysiology via Ai14+ D2 MSNs. We found that D2 MSNs showed a slight, but significant increase in spontaneous excitatory post synaptic current (sEPSC) frequency, suggesting a slight increase in excitatory tone. Moreover, we found a substantial increase in inhibitory spontaneous post synaptic current (sIPSC) frequency. Taken together, we conclude that Lphns are critical for inhibitory synaptic inputs onto striatal D2 MSNs, and that the alterations in D2 MSN signaling generate severe motor deficits upon the administration of amphetamine. Thus, we conclude that Lphns play an important role in mediating specific inputs onto striatal D2 MSNs, where the loss of these aGPCRs increases inhibitory connectivity and leads to significantly larger inhibitory tone. These findings provide novel insight into the importance of Lphns in striatal development and function.

Disclosures: S. Watson: None. B.A. Grueter: None. R. Sando: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.17/B48

Topic: B.04. Synaptic Transmission

Title: Glun2d-containing nmda receptors in pv basket cell axons gate circuit excitability

Authors: *V. J. OLAH¹, T. G. BANKE⁴, I. WITTEVEEN⁵, Y. YOOK⁶, R. E. PERSZYK⁷, T. T. NGUYEN⁷, F. G. GHINGER⁸, S. J. MYERS¹⁰, J. Y. ZHANG⁸, D. LIOTTA¹, H. YUAN⁹, S. F. TRAYNELIS², M. J. ROWAN³;

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Abstract: According to the classical Hebbian learning model, postsynaptic NMDA receptors modulate the impact of neuronal signals by detecting coincident depolarization of the synaptic partners. However, recent evidence suggests that NMDA receptors are also expressed presynaptically in subsets of cortical excitatory synapses, thereby influencing synaptic communication via a wholly distinct mechanism. As NMDA receptors are involved in a wide range of normal functions ranging from development to plasticity and are considered high confidence therapeutic targets for both neurological disease and neuropsychiatric indications, a complete mechanistic understanding of these receptors is paramount. Glutamate- and glycine-activated NMDA receptors are tetrameric assemblies of two GluN1 and two GluN2 subunits, of which there are four types (GluN2A,B,C,D) that control channel properties and synaptic time course. Among the GluN2 subunits, GluN2D has unique properties (much slower kinetics, higher glutamate affinity) and is the least studied subtype. Here we show multiple lines of evidence including that GluN2D-containing NMDA receptors are functionally localized at the presynaptic basket terminals of ‘fast-spiking’ parvalbumin (PV) interneurons in the mature hippocampus. Further, we examined putative mechanisms by which they are trafficked to presynaptic boutons in PV basket terminals. We show that the basal activity of GluN2D-containing NMDA receptors functionally controls stochastic GABA release and short-term plasticity. We also demonstrate that GluN2D-containing receptors specifically on PV interneurons mediate the inhibitory synaptic and behavioral responses to low dose ketamine, suggesting that this neuron-type-specific target has therapeutic potential for treatment-resistant depression and a host of other brain disorders.

Disclosures: V.J. Olah: None. T.G. Banke: None. I. Witteveen: None. Y. Yook: None. R.E. Perszyk: None. T.T. Nguyen: None. F.G. Ghinger: None. S.J. Myers: None. J.Y. Zhang: None. D. Liotta: None. H. Yuan: None. S.F. Traynelis: None. M.J. Rowan: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.18/B49

Topic: B.04. Synaptic Transmission

Support: NIH Grant R01MH124934

Title: Serotonin transiently increases the excitatory synaptic drive of a subpopulation of extratelencephalic neurons in the adult mouse prelimbic cortex

Authors: A. S. ABERRA, *A. T. GULLEDGE;
Geisel Sch. of Med. at Dartmouth, Hanover, NH

Abstract: The prefrontal cortex receives dense serotonergic innervation that contributes to a broad array of cognitive and behavioral processes, including decision making, learning, and regulation of mood. Two broad non-overlapping populations of pyramidal neurons provide

output from local cortical circuits: extratelencephalic (ET) neurons that extend ipsilateral corticofugal projections and intratelencephalic (IT) neurons that project bilaterally within the telencephalon. Serotonin (5-HT) selectively enhances excitability of IT neurons (via 5-HT_{2A} receptors) but inhibits ET neurons (via 5-HT_{1A} receptors), but the presynaptic effects of 5-HT on excitatory afferents to ET and IT neurons is less well understood. Here, we studied the effects of 5-HT on spontaneous excitatory drive in ET and IT neurons in the adult mouse prelimbic cortex. Whole-cell current-clamp recordings were made from pairs of pyramidal neurons in layer 5 that were identified from their electrophysiological profiles. Bath application of 5-HT (40 μ M) induced a transient (~3 min) increase in the amplitude and frequency of spontaneous excitatory post synaptic potentials (sEPSPs) in a subset of pyramidal neurons. ET neurons experienced a larger and more variable increase in sEPSP frequency across all recordings (n = 58 ET, 40 IT). Part of the variability in responses could be explained by the baseline sEPSP frequency, which was inversely correlated with the 5-HT-induced change in sEPSP frequency in ET neurons ($R^2 = 0.20$, $p < 0.0001$), but not IT neurons. At the same time, the onset of the increase in sEPSP amplitude and frequency coincided with a consistent and sustained 5-HT_{1A}-receptor-mediated outward current that was prominent in ET, but not IT, neurons. 5-HT also increased the number of synchronous sEPSPs in ET-ET pairs, but not IT-IT pairs, relative to chance, suggesting that 5-HT may directly excite a yet-unknown subpopulation of ET-targeting cortical neurons.

Disclosures: A.S. Aberra: None. A.T. Gullledge: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.19/Web Only

Topic: B.04. Synaptic Transmission

Support: Fondecyt 1241173
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Anid grant for doctoral students
FIB-Universidad de Valparaiso grant for doctoral students

Title: Disrupted temporal integration in hippocampal ca1 neurons during pentylentetrazole-induced epileptogenesis.

Authors: *F. AGUILAR¹, F. GUIFFA³, S. VAN BUUREN³, J. AHUMADA⁴, C. CERNA⁵, R. SOTOMAYOR-ZÁRATE², M. FUENZALIDA⁶;

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Abstract: Background: Epilepsy, a neurological condition characterized by recurrent seizures, affects approximately 1% of the global population. In many cases, the condition is accompanied by memory and learning deficits. Approximately 30% of patients develop pharmacoresistance, highlighting the critical need to understand the mechanisms underlying epileptic network formation. The present study investigated how temporal integration of excitatory inputs onto hippocampal CA1 pyramidal neurons — which are essential for memory processing — is altered during epileptogenesis. **Methods:** Intraperitoneal administration of pentylenetetrazole (PTZ, 35 mg/kg), a GABA-A receptor antagonist, was used to induce an excitatory/inhibitory (E/I) imbalance in mice. Whole-cell patch-clamp recordings were performed on CA1 pyramidal neurons to assess synaptic transmission and temporal integration properties. **Results:** PTZ treatment produced a three-fold increase in evoked excitatory current amplitude compared to vehicle controls. The excitation-inhibition index (calculated from co-recorded currents at -40 mV) was significantly elevated in PTZ animals (1.98 ± 0.30 , $n=12$ vs. 0.82 ± 0.10 , $n=9$; $P=0.0039$). This E/I imbalance coincided with increased presynaptic glutamate release probability, evidenced by higher spontaneous EPSC frequency (PTZ: 1.29 ± 0.21 Hz, $n=10$ vs. vehicle: 0.70 ± 0.14 Hz, $n=8$; $P=0.0394$) and reduced paired-pulse ratio (PTZ: 1.02 ± 0.06 , $n=17$ vs. vehicle: 1.52 ± 0.12 , $n=13$; $P=0.0007$). Critically, current-clamp recordings revealed enhanced temporal summation of excitatory inputs separated by 15 ms. While vehicle neurons generated action potentials in $49.04 \pm 6.84\%$ of trials ($n=7$), PTZ neurons fired in $75.15 \pm 3.58\%$ of trials ($n=10$; $P=0.0030$), demonstrating a broadened temporal summation window. Intrinsic neuronal properties remained unchanged, suggesting primarily presynaptic mechanisms. Interestingly, while miniature IPSCs increased (PTZ: 1.77 ± 0.20 Hz, $n=8$ vs. vehicle: 0.88 ± 0.25 Hz, $n=7$; $P=0.05$), spontaneous IPSCs were unchanged ($P=0.08$), indicating complex alterations in interneuron function. **Conclusions:** PTZ-induced epileptogenesis profoundly disrupts temporal integration within the hippocampus, through enhanced presynaptic glutamate release and altered excitatory and inhibitory (E/I) balance. These findings provide mechanistic insights into epileptic network dysfunction and may inform therapeutic strategies targeting temporal coding deficits in epilepsy. <!--EndFragment-->

Disclosures: F. Aguilar: None. F. Guiffa: None. S. van Buuren: None. J. Ahumada: None. C. Cerna: None. R. Sotomayor-Zárate: None. M. Fuenzalida: None.

Poster

PSTR260: Pre- and Postsynaptic Modulation I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR260.20/B50

Topic: B.05. Synaptic Plasticity

Support: NIH R15MH129932-01

Title: Sex differences in endocannabinoid-modulation of hippocampal CA1 synaptic plasticity during adolescence

Authors: C. G. REICH¹, *V. R. SMITH², D. MERCADO², G. PETROVA³;

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Abstract: Work in our lab demonstrated a robust sex difference of rat endocannabinoid signaling at hippocampal dendritic GABAergic synapses (Ferraro, 2020). This is consistent with the literature on perisomatic eCB signaling in females. Importantly, these effects are not reported in similar studies of eCB signaling in males. We hypothesize that this sexual divergence across the somatodendritic axis in adolescent hippocampal pyramidal cells translates into differences in eCB-mediated synaptic plasticity. Field excitatory post-synaptic potentials (fEPSPs) were recorded from CA1 in male and female Sprague-Dawley rat (40-55 days old) hippocampal slices. Following a 10 min baseline, two weak (normally non-LTP inducing) stimulations (30 Hz, 0.5 sec and 50 Hz, 0.5 sec) were applied with 25 min between each stimulation. Then, a stronger LTP-inducing stimulation (100 Hz x 2, 0.5 sec apart) was applied and responses were allowed 30 min to recover. For drug-based experiments, slices were pre-incubated (>1 hr.) with the drug, which was bath applied throughout the experiment. All sample sizes were n=5 or greater, the size needed for 80% statistical power. We observed significant sex differences in LTP induction thresholds. During the 30 Hz stimulation, male slices exhibited ~115% increase in response magnitude, whereas female slices increased ~150% over baseline values. Male slices required the 100 Hz stimulation to reach response magnitudes of 150% over baseline values. In female slices, blocking CB1 receptors with AM251 (3 μ M) impaired LTP induction with 30 Hz stimulation. In male slices, enhancing tonic levels of 2-AG via inhibition of MAG-lipase (JZL184, 100 nM) enhanced LTP induction with 30 Hz stimulation to ~140% compared to baseline levels. Whereas, in females application of JZL184 to slices led to a reduction in LTP magnitude (~115% over baseline values), indicating an occlusion effect of tonic 2-AG signaling. Furthermore, enhancing AEA levels via FAAH inhibition (URB597, 1 μ M) blocked LTP induction (~102% over baseline values), indicating an occlusion effect of tonic AEA levels. In females, inhibiting AEA synthesis (NAPE-PLD inhibitor, 10 μ M) reduced 30 Hz LTP induction (~130% over baseline values). The CB1-dependent lower LTP thresholds in female slices is consistent with enhanced CB1-mediated suppression of GABAergic neurotransmission, thus resulting in greater excitatory drive in female CA1. Furthermore, the decrease in male LTP threshold via an increase in 2-AG tone suggests a lower excitatory drive in male CA1. Our preliminary data support the hypothesis that sex differences in tonic eCB activity are observed in eCB-mediated synaptic plasticity.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.01/B51

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support:

NIH Grant R01NS132293
The Harry T Mangurian Jr. Foundation, Inc

Title: Pff-induced lewy-like pathology disrupts executive function and modulates both intrinsic membrane properties and cortical excitability in mice

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Abstract: Lewy body dementias (LBD) are a group of neurodegenerative disorders characterized by the accumulation of α -synuclein aggregates, leading to cognitive, motor, and psychiatric symptoms. Among these manifestations, executive dysfunction stands out as a prominent and debilitating feature, significantly impacting the daily functioning and quality of life of affected individuals. Despite the hallmark executive function deficits in patients with LBD, the role of α -synuclein aggregation in executive dysfunction remains poorly understood. One brain area potentially contributing to executive function impairments is the prefrontal cortex, which is a nexus for executive function, and an area that exhibits robust Lewy pathology. To investigate the involvement of α -synuclein aggregation in the prefrontal cortex in LBD symptoms, we utilized a combination of pharmacological, electrophysiological, and behavioral approaches in the pre-formed fibril (PFF) mouse model of LBD at 6-weeks, 3-months and 1-year post-PFF injection. At each time point, we examined the effects of pathological α -synuclein on layer V pyramidal neurons in the prelimbic medial prefrontal cortex (PL-mPFC). To distinguish neurons with and without α -syn inclusions, we used a novel autofluorescent aggregate binder, enabling real-time identification of Lewy-like inclusion-positive (LLI+) and inclusion-negative (LLI-) neurons during patch-clamp recordings. At 3 and 12 months post-injection, LLI+ neurons exhibited decreased rheobase and increased excitability, while LLI- neurons showed the opposite—elevated rheobase and reduced excitability—suggesting α -syn inclusions drive hyperexcitability in affected cells and potentially cause compensatory hypoactivity in surrounding neurons, contributing to PL-mPFC circuit dysfunction. To assess executive function, we used a pairwise visual discrimination (PVD) task with reversal. At 6 weeks, both monomer- and PFF-injected mice performed similarly, indicating no early deficits. By 3 months, PFF-seeded mice showed impairments in both task acquisition and reversal accuracy, suggesting an aggregate burden-related decline in cognitive flexibility. Together, these findings provide new insights into how α -syn pathology disrupts prefrontal circuits and executive function in LBD, offering potential targets for future therapeutic intervention.

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Poster**PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I**

Location: SDCC Halls B-H

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Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: RM1MH133065

Title: Integrative multimodal profiling of human iPSC-derived neurons using Patch-seq

Authors: ***J. CHEN**¹, J. LIU², S. DIAZ DE LEON-GUERRERO³, P. GOWDA⁵, M. ELACHWAH¹, J. DUAN⁶, A. KREIMER⁷, Z. P. PANG⁴;

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Abstract: Advancing genetic discoveries in neurodevelopmental and psychiatric disorders (NPD) toward clinically actionable biology remains challenging due to limited understanding of gene function. To address this, we established a high-throughput multimodal Patch-seq pipeline to systematically characterize human neurons derived from induced pluripotent stem cells (iPSCs). Our approach integrates electrophysiological, morphological, and transcriptomic analyses at single-cell resolution to support predictive modeling and functional annotation of neuronal subtypes and disease-associated genetic variants. To date, we have performed whole-cell patch-clamp recordings on over 300 healthy-control iPSC-derived neurons to measure intrinsic neuronal properties, including resting membrane potential, action potential threshold, firing patterns, and spontaneous synaptic currents. Following electrophysiological recording, neurons were filled with Alexa Fluor™ 568 for morphological tracing. Morphometric analyses—including Sholl analysis and quantification of neurite length, branching complexity, and arborization—are ongoing. Neuronal nuclei were aspirated post-imaging for single-cell RNA sequencing with an ~80% sequencing success rate, yielding high-quality transcriptomic data that demonstrate substantial cellular heterogeneity. Preliminary integration of these multimodal datasets reveals considerable functional and morphological diversity, validating the feasibility of predictive modeling approaches linking gene expression with electrophysiological and morphological phenotypes. This integrative Patch-seq multimodel, planned to exceed 3,000 neurons, including genetically modified disease models, will establish an essential reference dataset to facilitate robust analyses of biological convergence and divergence across disorders, significantly advancing mechanistic understanding of NPD genetic risk.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Topic: B.06. Intrinsic Membrane Properties and Signal integration

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Title: Electrophysiological maturation of hIPSC-derived dopaminergic neurons

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Abstract: Dopaminergic (DA) neurons are critically involved in the control of voluntary movement, reward processing, and cognition. The vulnerability of some of them in Parkinson's disease underscores the need for reliable human-relevant *in vitro* models. Human iPSC-derived DA neurons can bridge this gap, but a detailed understanding of their electrophysiological maturation is essential for translational use. The aim of this study was to characterize the temporal dynamics and defining features of electrophysiological maturation in hIPSC-derived DA neurons, with a focus on pacemaking activity. Differentiation of DA neurons was performed using either the GM38 or a mCherry-tagged TH-reporter iPSC line and conventional methods (Stathakos et al., 2019). Neuronal identity and DA phenotype were confirmed via immunocytochemistry (β III-tubulin, TH). Cell-attached and whole-cell patch-clamp recordings were carried out at multiple time points to assess action potential generation, I_h current amplitude, spontaneous firing patterns, and D2-mediated autoinhibition. Temperature was $\sim 32^\circ\text{C}$. DA neurons were identified by their slow, regular firing rate (0.5-5 Hz), broad action potentials (>1.35 ms half-width), presence of I_h , and hyperpolarizing response to $100\ \mu\text{M}$ dopamine. TH expression started to be observed by day 10 and was found in 64% of the neuronal cells by day 30 of terminal differentiation. Electrophysiologically, neurons began firing evoked action potentials between days 10-14, with I_h current detectable in some cases. Spontaneous activity emerged around day 20, initially irregular, and transitioned to stable pacemaking by day 40. These observations are consistent with previous data (Borgs et al., 2016 and others). Dopamine application at this stage induced a reversible hyperpolarization (>5 mV) via somatodendritic D2 receptor activation, confirming functional autoinhibitory feedback. The low coefficient of variation ($\text{CV} \sim 0.18$) in firing pattern further indicated a mature DA phenotype. Interestingly, some presumably non-DA neurons also fired spontaneously around day 40-50, but their firing was irregular ($\text{CV} \sim 0.65$) and was not inhibited by dopamine. We conclude that, by day 40, hIPSC-derived putative DA neurons exhibit hallmark features of mature electrophysiological function, positioning them as a powerful *in vitro* model for neurophysiological and pharmacological studies. The nature of non-DA neurons remains to be determined. Their presence may be problematic in the perspective of DA neuron-based grafts in PD patients.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Title: Coupled beta - high-frequency oscillations in local field potentials emerges through synchronized bursting in a simple reduced model of the subthalamic nucleus

Authors: *H. SHEHEITLI^{1,2,3}, K. J. O'NEILL, III², J. WANG², J. E. AMAN², L. A. JOHNSON², J. L. VITEK, MD, PHD²;
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Abstract: The classic hallmark of Parkinson's Disease (PD) is the elevation of local field potential (LFP) oscillatory power in the beta frequency range (13-30Hz). More recently, it has been reported that phase-amplitude coupling (PAC) between beta and high frequency oscillations (HFO) (>200Hz) is found to be linked to symptom severity in PD. While numerous models of basal ganglia electrophysiology have been put forward with a focus on the generation of the beta oscillation, much less attention has been given to the coexistence and interaction of slow and fast rhythms in basal ganglia LFPs. Our purpose here is twofold, first to demonstrate the emergence of synchronized bursting in a reduced model of the subthalamic nucleus (STN) consisting of a single population of excitatory quadratic integrate-and-fire neurons with slow adaptation and recurrent synapses, and second to elucidate the association of these population states to the beta-HFO PAC observed in the LFP. We numerically simulate a population of heterogeneous Izhikevich neurons with recurrent conductance-based excitatory synapses; we demonstrate that as the strength of recurrent synapses increases, the population undergoes a transition from an asynchronous firing regime to a synchronized bursting regime; in the latter, the average inter-burst interval matches the beta frequency range and the intra-burst average firing rate matches the HFO frequency range. The beta-HFO PAC is seen to emerge in this synchronized bursting regime concurrently with the emergence of an HFO peak in the power spectrum of the LFP. While the proposed model system is not intended to be a precise model of the subthalamic nucleus, it serves to demonstrate how beta-nested HFO can emerge from the interplay of fast spiking, slow adaptation currents and recurrent excitatory synapses. This simple model captures a dynamical mechanism that may partially explain the emergence of coupled beta-HFO oscillatory activity in PD and can be used to better understand and predict the effect of interventions such as deep brain stimulation for which the STN is a common target; it can also be seen as a computationally efficient building block in future models of the larger basal ganglia thalamocortical network that aim to account for the distributed multiscale oscillatory alterations manifesting in the PD state across different brain regions.

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Poster

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Topic: B.06. Intrinsic Membrane Properties and Signal integration

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Title: Cell-type specific noradrenergic modulation in the mouse paraventricular nucleus of the hypothalamus

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Abstract: The paraventricular hypothalamus (PVH) is a central regulator of autonomic functions, integrating signals from various pathways, including noradrenergic inputs, to maintain processes like energy balance. It receives input from the arcuate nucleus and other modulatory centers. Our long-term goal is to understand how the noradrenergic system influences feeding behavior by investigating the properties of PVH neurons and their modulation by noradrenaline (NA).

We first characterized the intrinsic electrophysiological properties of PVH neurons using perforated patch-clamp recordings in mice, aligning our findings with previous studies in rats. Three distinct neuron types were identified: Type I neurons, corresponding to magnocellular (MC) neurons, displayed delayed spike onset and slow spike-frequency adaptation. Type II neurons (putative parvocellular, PC) fired repetitive spikes without delay, while Type III neurons (putative preautonomic, PA) showed spike bursts and rapid adaptation. Morphologically, Type III neurons had more complex dendritic structures, while Type I had the largest somata. These distinct neuron types were distributed differently across PVH regions, with Type I neurons located anteriorly and Type III neurons more posteriorly.

NA differentially modulated the activity of these neuron types. In Type I neurons, around 80% showed increased firing in response to NA, while the remaining 20% were inhibited. Type II neurons mostly decreased their firing (60%), although 35% showed excitation. In Type III neurons, 65% were inhibited by NA. This demonstrates that NA modulates PVH neurons in a cell-type-specific manner.

To analyze the neuron type-specific composition of AR subtypes mediating the differential NA responses, we performed cell type-specific transcriptomics and confirmed the functional relevance of these AR subtypes using functional pharmacology. As a prerequisite, we first addressed the challenge that electrophysiologically defined PVH neuron Types I-III cannot currently be linked to distinct genetic markers. To circumvent this, we investigated AR expression profiles in PVH neuron populations defined by the expression of key modulatory peptides: oxytocin (Oxt, predominantly expressed in Type I neurons), arginine vasopressin (AVP, also predominantly Type I), corticotropin-releasing hormone (CRH, represented across all

three types), and thyrotropin-releasing hormone (TRH, predominantly Type II). In conclusion, our findings reveal that NA differentially modulates PVH neurons through distinct AR subtypes, suggesting a complex regulatory mechanism for autonomic processes such as energy balance.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Topic: B.06. Intrinsic Membrane Properties and Signal integration

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Title: Distinct electrophysiological properties and dopaminergic modulation of medium spiny neurons across divisions of the tail striatum and in comparison to the dorsolateral striatum.

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Abstract: The tail striatum (TS) is a unique striatal domain with predominantly sensory functions. Recent evidence supports that abnormal dopamine transmission in the TS contributes to sensory symptoms of dopaminergic diseases, like hallucinations, by dysregulating TS medium spiny neurons (MSNs). Importantly, the ventral TS is segregated into three divisions (medial, intermediate, and lateral), in which TS-MSNs express D1 and D2 dopamine receptors unevenly, but the functional significance of this organisation is not yet understood. We used whole-cell patch clamping in rat (P45-55, both sexes, n=62) brain slices to carry out the first in-depth electrophysiological characterisation of TS-MSNs (n=102) and compared them across TS divisions (n=26-46) to gain insight into their organisation. The adjacent dorsolateral striatum (DLS) domain, which is well-characterised, was also included for comparison. Bath-application of 5-100 μ M dopamine was used to investigate dopaminergic modulation of MSNs in each region. We found TS-MSNs to be electrophysiologically distinct from DLS-MSNs (n=28), reflecting the established functional differences between these domains. TS-MSNs were more depolarised than DLS-MSNs at rest (-74 ± 1 mV vs -85 ± 2 mV), had lower capacitance (39 ± 3 pF vs 66 ± 6 pF), and higher EPSC frequency (3.8 ± 0.2 Hz vs 1.4 ± 0.2 Hz). We also found differences in excitability (rheobase) between TS divisions (lateral: 130 ± 11 pA, intermediate: 176 ± 12 pA), supporting their functional heterogeneity. Unexpectedly, TS-MSNs were unaffected by dopamine < 100 μ M, and only lateral division TS-MSNs responded to 100 μ M dopamine, exhibiting reduced excitability. DLS-MSNs were strongly suppressed (no firing) by 100 μ M

dopamine, consistent with previous studies. These data suggest TS-MSNs are less sensitive to dopamine than DLS-MSNs, particularly in the medial and intermediate divisions where D1 and D2 are expressed atypically, which could have implications for dopaminergic disease. Our preliminary results (n=4 rats) from imaging the TS after injection of fluorescently tagged viral vectors into the primary visual cortex show diffuse innervation of the whole ventral TS, indicating that the TS divisions share common visual inputs, which could be important if they function cooperatively. Next, we will map primary visual and auditory cortex input to the TS by recording optogenetic photocurrents in TS-MSNs of all divisions to investigate whether their input is heterogeneous. Overall, our findings advance our understanding of TS-MSNs and the TS divisions and provide novel insight into how TS-MSNs are affected by high striatal dopamine in disease.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Topic: B.06. Intrinsic Membrane Properties and Signal integration

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Title: Morphological and electrophysiological properties of hypothalamic A11 dopamine neurons

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Abstract: Descending pain modulation systems are key targets in understanding the transition to chronic pain. Descending dopamine pain modulation originates from hypothalamic A11 dopamine neurons that project to the dorsal horn of the spinal cord and modulate nociception. However, very little is known about the basic properties of A11 dopamine neurons. Thus, we characterized these neurons using brain slice immunohistochemistry and electrophysiology in male and female mice.

Using immunohistochemistry, we characterized the expression of enzymes required for monoamine synthesis in A11. A11 dopamine neurons were identified by expression of tyrosine hydroxylase (TH), the rate-limiting enzyme in dopamine synthesis. Consistent with prior reports, we found that 95% of TH+ A11 neurons expressed amino acid decarboxylase (AADC), the final enzyme in dopamine synthesis, but not enzymes required for serotonin or noradrenaline

synthesis. Thus, A11 neurons are likely capable of synthesizing dopamine. Next, using immunohistochemistry, we determined the specificity and penetrance of Cre expression in A11 in Cre lines available to target dopamine neurons. We used TH-Cre-1, TH-IRES-Cre, and TH-2A-CreER mice crossed with Ai14 reporter mice, which produced cytosolic tdTomato fluorescence in Cre⁺ neurons. TH-2A-CreER mice labeled A11 dopamine neurons with the highest specificity (70%), whereas TH-Cre-1 and TH-IRES-Cre demonstrated specificity below 50%. All TH-Cre lines demonstrated high penetrance (~90%) for A11 dopamine neurons. Using fluorescence-guided patch-clamp electrophysiology in acute brain slices from TH-IRES-Cre::Ai14 and TH-2A-CreER::Ai14 mice, morphology and the intrinsic and active membrane properties of A11 dopamine neurons were determined. After recording with an internal solution containing Neurobiotin, brain slices were fixed and immunostained for TH and with fluorescent streptavidin to visualize Neurobiotin-filled neurons. We found that both TH⁺ and TH⁻ neurons in the A11 region were small, high membrane resistance, and had a simple proximal dendritic arbor. Additional electrophysiological properties of A11 neurons will be presented. A11 dopamine neurons are important in the modulation of pain. However, little was previously known about their electrophysiological properties. This report will provide foundational information necessary to develop high quality studies of these neurons and advance the study of descending dopamine modulation in chronic pain.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.08/B58

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Title: In Vivo Whole-Cell Recording from Lateral Habenular Neurons

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Abstract: Major depressive disorder (MDD) is a common psychiatric condition characterized by symptoms such as impaired concentration, persistent sadness, and recurrent suicidal thoughts. Recent studies have increasingly implicated the lateral habenula (LHb) as a key neural substrate in the pathophysiology of MDD. Suppressing LHb activity alleviates depressive symptoms, suggesting that its hyperexcitability drives depressive states (Yang et al., 2018). However, the precise functional role of the LHb *in vivo* remains unclear due to its deep anatomical location, which complicates access via traditional electrophysiological methods. Standard approaches, such as extracellular recordings and *in vitro* patch-clamp assays, do not capture subthreshold membrane potential dynamics under naturalistic conditions. Furthermore, discrepancies between *in vitro* and *in vivo* data underscore the urgent need for high-resolution intracellular recordings in

awake, behaving animals. This study aims to investigate the temporal dynamics of LHb neuronal activity during the development of depressive-like states and during recovery following antidepressant treatment in mice. By characterizing the progression of electrophysiological changes across these states, we aim to identify the neurophysiological adaptations underlying symptom onset and remission. This could reveal circuit-level signatures of vulnerability or resilience to MDD. Our methodological approach combines long-term local field potential (LFP) recordings using Neuropixels probes with *in vivo* whole-cell patch-clamp techniques optimized for deep-brain access. We have developed a customized experimental protocol that enables stable intracellular recordings from LHb neurons by minimizing tissue damage through the targeted aspiration of overlying structures. The recording pipette is positioned within 0.5 mm of the LHb, and precautions are taken to prevent contamination during insertion. This approach allows for precise monitoring of population-level activity and single-cell membrane potential dynamics. This research will yield new insights into the electrophysiological properties of LHb neurons across affective state transitions and establish a robust framework for examining the temporal neurophysiology of mood disorders. By elucidating how LHb activity evolves along the depressive trajectory, our findings may facilitate the development of early diagnostic tools and therapeutic interventions for MDD. Our ultimate goal is to advance understanding of the neurobiological mechanisms underlying depression and improve clinical outcomes.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

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Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: NIH P01DA047233

Title: Characterization of lateral habenula neurons activity patterns

Authors: *M. ISHIKAWA, A. CORONA, P. J. KENNY;
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Abstract: The lateral habenula (LHb) is thought to play an important role in stress-related behavioral disorders such as major depression. Stress induces hyperexcitability of LHb neurons, which facilitates the expression of depressive-like behaviors. Conventionally, the firing pattern of LHb neurons is categorized by three major modes of intrinsic activity: silent, tonic, and bursting. However, it is empirically known that more subtypes of firing pattern exist in the LHb, but the details of the subtypes remain unknown. In this study, we recorded nearly 400 neurons to

investigate the pattern of action potential firing in LHb neurons. As reported previously, we obtained similar percentages of three firing patterns of spontaneous action potential in the LHb: silent 50.4%, tonic 34.4% and burst 8.0%. Some populations of neurons showed mixed or combined with tonic and burst firing; we defined this group as mixed firing 7.2%. In addition, we applied hyperpolarized stimulation during spontaneous action potential firing recording and checked the firing pattern in LHb neurons. Since it is well known that LHb neurons show rebound burst firings by hyperpolarization, as expected, we found more burst firing neurons but mostly mixed patterns were observed by the stimulation. Moreover, some of the burst firing neurons were composed by repetitive burst firings followed by large after hyperpolarization potential, which was rarely seen in spontaneous burst firings. These findings suggest that there are at least two subtypes of burst firing in LHb neurons. We are investigating the mechanisms of the two types of burst firing patterns.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Title: Social isolation stress increases burst-firing and disrupts microglia in the lateral habenula

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Abstract: Social isolation (SI) increases vulnerability to mood disorders by disrupting neural circuits involved in emotion regulation. Stress increases the propensity of lateral habenula (LHb) to fire high-frequency bursts of action potentials, an adaptation contributing to behavioral deficits associated with mood disorders. Effects of SI on LHb burst firing have not been explored. Using current-clamp recordings in LHb slices, we detected a striking increase in the proportion of LHb neurons that burst-fire in mice after a prolonged period of SI. Burst-firing LHb neurons in SI mice exhibited more frequent burst-firing events and hyperpolarized resting membrane potential compared to controls. Microglia are emerging as key regulators of neural circuit homeostasis. Using Cx3cr1-Cre::Ai14 reporter mice, we detected a marked decrease in

the numbers of microglia in the LHb of SI mice relative to controls. LHb microglia in SI mice had increased process endpoints, a morphological feature associated with enhanced neuronal surveillance, relative to controls. To investigate the role of microglia in gating LHb burst-firing, mice were permitted to consume regular chow (control) or chow containing the CSF-1R antagonist PLX3397 (lesioned), which depletes microglia from the brains of mice. The microglia-lesioned mice demonstrated a striking increase in the proportion of LHb neurons that engaged in burst-firing relative to controls. This shows that microglia exert inhibitory homeostatic control over the burst-firing activity of LHb neurons. Together, these data suggest that social isolation remodels LHb microglia, resulting in increased burst-firing of LHb neurons, which is a core cellular adaptation thought to drive vulnerability to mood disorders.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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22K19362

Title: Networks between neuron and nonneuronal cells are involved in chronic stress-dependent electrophysiological changes of lateral habenular neurons

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Abstract: The lateral habenula (LHb) is known to downregulate dopaminergic and serotonergic systems via direct or indirect projections, and its dysfunctions are considered to participate in the pathophysiology of psychiatric disorders. While hyperactivation of LHb neurons is implicated in depression, the detailed electrophysiological basis for LHb hyperactivity remain unclear. Here,

we used a chronic social defeat stress (CSDS) mouse model to elucidate underlying mechanisms for the stress-induced electrophysiological changes in LHb neurons. Whole-cell recordings revealed that rebound depolarizing potentials (RDPs) were evoked after the offset of the hyperpolarizing pre-pulse in LHb neurons. We classified neurons exhibiting RDPs lasting longer than 400 ms as long-RDP neurons and the others with shorter ones as short-RDP neurons. In CSDS-susceptible mice, the proportion of long-RDP neurons was significantly reduced compared to CSDS-resilient and CSDS-naïve mice. Both short and long RDPs were triggered by T-type Ca^{2+} channels and terminated by SK channels. Meanwhile, the plateau depolarizing potential in long-RDPs was mediated by cyclic nucleotide-gated (CNG) channels. Notably, application of carbenoxolone or meclofenamic acid, gap-junction blockers, shortened RDP durations, suggesting that CNG channels are activated in electrically-coupling cells. As expected, dual whole-cell recordings revealed existence of electrical coupling between neurons and non-neuronal cells, but not among neurons. Furthermore, dye-coupling experiments revealed couplings between neurons and oligodendrocyte-lineage cells. RNA-seq data showed the reduced expression of *Cnga4* in CSDS-susceptible mice. AAV-based CRISPR-Cas9 local knockout of *Cnga4* in the LHb led to a significant decrease in the proportion of long-RDP neurons. These findings suggest that CNG channels and neuron-glia networks play crucial roles in stress-related LHb plasticity, which may offer novel therapeutic targets for depression.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.12/C2

Topic: B.06. Intrinsic Membrane Properties and Signal integration

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T2241002
2024YFF0507601

Title: Ionic mechanisms and physiological functions of complex spike burst in lateral septum neurons

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Abstract: Neuronal information processing depends on the precise control of firing patterns. Complex spike burst (CSB), a unique firing pattern, consists of 2-6 action potentials (APs) with progressively declining amplitude and increasing duration that ride on top of a slow depolarizing

potential. CSBs in hippocampal CA1 pyramidal neurons are involved in the activity-dependent synaptic plasticity and the experience-dependent remapping of place fields. CSBs are also observed in neurons of the lateral septum (LS), a key limbic structure involved in the regulation of emotion, motivation, and social behaviors. However, the ionic mechanisms and physiological functions of CSBs in LS neurons (LSNs) remain unclear. We conducted whole-cell patch-clamp recording in LSNs of brain slices obtained from adult male mice. We found that the majority of the recorded LSNs showed CSBs, which depend on the generation of Ca^{2+} spikes. In the presence of tetrodotoxin, positive current injections could activate the Ca^{2+} spikes in an intensity-dependent manner. Bath application of Cd^{2+} , a non-selective calcium channel blocker, completely abolished CSBs. Consistently, two-photon imaging demonstrated that CSBs were accompanied by large Ca^{2+} transients, underscoring the pivotal role of Ca^{2+} dynamics in mediating the generation of CSBs. Interestingly, CSB-associated increase in Ca^{2+} transients were confined to the somatodendritic compartments. Paired recording from LSNs revealed that only APs, but not the slow depolarization potential, during the CSBs could evoke postsynaptic responses, suggesting that the slow depolarization potential may exert its effects solely in somatodendritic compartments. Indeed, pairing CSBs, but not similar number of APs (without the slow depolarization), with simultaneous or preceding synaptic inputs produced long-term synaptic potentiation. Together, our results indicate that the generation of CSBs depends on Ca^{2+} influx and membrane potential depolarization. While APs can conduct along the axon and trigger synaptic transmission, the slow depolarization potential during CSBs plays a crucial role in producing synaptic plasticity at the somatodendritic compartments of LSNs.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.13/C3

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Title: Using cognitive demand to uncover sex-specific EEG microstate patterns and hormonal modulation of mental fatigue

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Abstract: Individual differences account for a substantial proportion of variability in baseline EEG measures. However, certain task-evoked conditions can amplify these differences. Here, we report that microstate dynamics reveal sex- and age-related differences only under cognitive demand, highlighting the potential for task-based EEG markers in diagnostic applications. Using open-source EEG data (dataset accession number: ds005385, <https://doi.org/10.18112/openneuro.ds005385.v1.0.2>), we found that sex differences in microstates B, E, F, and G were evident only after cognitive task performance [Wilk's $\Lambda =$

0.787, $F(7, 52) = 2.008$, $p = 0.072$, $\eta^2 = 0.213$]. This task-evoked emergence of microstate differences suggests that cognitive effort or fatigue may interact with underlying biological factors. Based on this, we hypothesized that mental fatigue may be modulated by gonadal hormone levels, with particular interest in whether hormonal fluctuations across the menstrual cycle contribute to differential susceptibility. Data collection is ongoing in an electroencephalogram (EEG) study designed to examine interactions between self-reported fatigue, microstate dynamics, and gonadal hormone fluctuations. Participants were asked to complete a questionnaire, along with EEG measures, two cognitive tasks, and collection of a saliva sample. Microstate analysis completed from post task eyes-closed EEG measures was used to investigate potential relationships between self reported questions and the various microstates. Saliva samples were used to measure relative estradiol, progesterone, and testosterone levels to then be compared to microstate dynamics and self reports. Initial analysis in self reports of fatigue revealed a significant relationship between perceived fatigue and microstates F and G following cognitive tasks. Specifically, when participants reported mental fatigue there was a negative correlation in the occurrence and coverage of microstate F [($r = -0.777$, $p = 0.014$), ($r = -0.791$, $p = 0.011$)] and G [($r = -0.733$, $p = 0.025$), ($r = -0.768$, $p = 0.011$)]. Previous research has demonstrated that microstate F is associated with personal information processing, while microstate G is linked to somatosensory network activity. The observed reduction in these microstates among individuals reporting higher levels of mental fatigue may reflect diminished self-related processing during fatigue states. Ongoing work will assess whether self-reported fatigue interacts with microstate dynamics and gonadal hormone fluctuations.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.14/C4

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: NIH Grant 1R01HL162575
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Title: Investigating the mechanisms underlying hypoxia-evoked increases in vasopressin cell firing frequency in the context of salt-evoked inverse neurovascular coupling

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Abstract: We recently showed an inverse neurovascular coupling (iNVC) response in which salt-induced vasopressin (VP) cell activation in the hypothalamic supraoptic nucleus (SON) resulted in dendritic release of VP, leading to localized vasoconstriction and hypoxia. Additionally, we showed that hypoxia in turn increases VP firing, suggesting that iNVC mediates a positive feedback loop to optimize VP activation during homeostatic challenges (Roy et al., Cell Reports 2021). Still, the mechanism by which iNVC-hypoxia increases firing in VP neurons has not yet been explored. We hypothesized that hypoxia-induced acidosis, via activation of acid-sensitive ion channels (ASICs) in this region, is a key mechanism contributing to VP cell excitation during iNVC. To test this, we first performed 2-photon imaging in an in vivo anesthetized rat preparation in which the SON is exposed, and showed that in addition to hypoxia, an acute salt challenge (1 mL, 2 M NaCl, infusion rate 2 mL/hr) induced local acidosis, as shown by an increase fluorescence intensity of the pH sensitive dye pHRhodo ($p < 0.0001$, $n = 22$, paired t-test). Using cell-attached electrophysiology ex vivo in eGFP-VP transgenic rats we find that VP firing frequency was significantly increased over time when the slice was exposed to an 8-minute hypoxic challenge (95% O₂ to 20% O₂) ($n = 19$, $p < 0.0001$, RM ANOVA). This effect persisted in the presence of ASIC-blocker amiloride (100 μ M) ($n = 9$, $p < 0.0001$, RM ANOVA), and, contrary to our expectations, the peak change in firing was significantly larger than in the control group ($n = 19$ control, $n = 9$ amiloride, $p < 0.05$, unpaired t-test). To determine the effects of exogenous acidosis per se on SON VP cell firing, we recorded from slices perfused with HEPES-buffered solutions at pH 7.25 (control) or pH 6.3 (acidosis). Extracellular pH was monitored via a micro-pH probe, and intracellular pH was monitored via pHRhodo fluorescence. We observed a mixed effect of a 5-minute acidosis challenge on firing, with a predominantly inhibitory response ($n = 7/16$ cells, $p < 0.05$, RM ANOVA) aligned with changes in intracellular pH. This effect was blocked in the presence of amiloride (100 μ M) ($n = 9$, $p = 0.38$, RM ANOVA). Taken together, these findings indicate that in addition to hypoxia, an acute salt challenge also induces local tissue acidosis within the SON. However, and contrary to our initial hypothesis, we show that acidosis is not a key intermediary by which hypoxia excites VP neurons. Instead, our data suggests that acidosis may constrain firing frequency and prevent overexcitation during local hypoxia.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

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Program #/Poster #: PSTR261.15/C5

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: AMX-20-CE-06
ANR-16-CONV-0001

Title: Serotonergic modulation of excitability in visual cortical pyramidal neurons across layers and areas

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Abstract: The predictive processing framework posits that perception arises from a dynamic comparison between sensory input and internally generated predictions, with prediction error (PE) signals guiding internal model updates. While PE signals have been described in layer 2/3 (L2/3) pyramidal neurons of the mouse primary visual cortex (V1), we hypothesise that layer 5 extratelencephalically projecting (L5ET) neurons in secondary visual cortex (V2M) also participate in PE computations and active inference. PE computations are proposed to be gated by neuromodulators, which can shift the balance between sensory and predictive information streams. While 5-HT has been implicated in altered brain states and visual hallucinations, its effect on the computational repertoire and intrinsic excitability of visual cortical neurons remains poorly understood.

To address this, we investigated whether 5-HT modulates the intrinsic excitability of pyramidal neurons in V1 and V2M. Using in vitro whole-cell current-clamp recordings in L2/3 and L5 neurons under synaptic blockade, we found that 5-HT produced layer- and area-specific modulation of somatic excitability, including changes in input resistance, resonance frequency, firing rate-current (f/I) relationships, firing patterns, and after-hyperpolarisation amplitude. In L5 neurons, 5-HT also reduced dendritic excitability. To probe the basis of inter-neuronal variability, we are constructing computational models with varying apical dendritic length and conductance profiles. Finally, we are using multivariate analysis of excitability features to uncover distinct functional clusters across layers and areas.

These findings demonstrate how serotonergic modulation shapes the computational properties of visual cortical pyramidal neurons, providing a potential mechanism for neuromodulatory control over PE computations and signalling.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

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Program #/Poster #: PSTR261.16/C6

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: NIH Grant NS027881 (CSL)
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Title: Orexin (Ox) and Alpha 1 adrenergic (Adra1) receptor activation enables a novel apamin-insensitive slow-afterhyperpolarization (ai-sAHP) mediated by the Ca^{2+} -dependent closure of ionotropic delta glutamate receptors (GluD) in serotonergic dorsal raphe ($\text{DR}^{5\text{-HT}}$) neurons

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Abstract: Serotonin release from DR^{5-HT} neurons modulates arousal, mood and reward and is a primary target for treating anxiety and depression. Orexin and norepinephrine inputs are important regulators of DR^{5-HT} neuron excitability acting via OX, and Adra1 GPCRs to produce a noisy depolarizing cation current and a greatly enhanced sAHP with two distinct components (Ishibashi et al. 2016 PMID: 27683906). The first is mediated by apamin-sensitive SK channels ($\tau \sim 0.5$ s) while the second is slower ($\tau \sim 5$ s) and apamin-insensitive (ai-sAHP). Here we used whole-cell patch clamp recordings in mouse brain slices to identify the channels underlying this ai-sAHP. Our evidence suggested the ai-sAHP results from the transient closure of the cation channels activated by orexin-A (OxA) or the Adra1 agonist phenylephrine (PE). Critically, as shown for the PE-evoked cation current (Gantz et al. 2020 PMID:32234214), we found that 1-Naphthylacetyl spermine (Naspm, 100 μ M), a blocker of ionotropic GluD receptors, also blocked the OxA-induced inward current and the associated ai-sAHPs following OxA or PE. Thus, metabotropic OX and Adra1 receptors gate GluD channels and the transient closure of these channels produces the ai-sAHP. To determine how these channels are closed, we investigated the sensitivity of this GluD current to both extracellular and intracellular $[Ca^{2+}]$. Elevating $[Ca^{2+}]_o$ attenuated both the inward current and the ai-sAHP while lowering $[Ca^{2+}]_o$ augmented both. Moreover, strong buffering of $[Ca^{2+}]_i$ (10 mM BAPTA) completely blocked the ai-sAHP, without reducing the depolarizing current. To test if elevating $[Ca^{2+}]_i$ is sufficient to elicit the ai-sAHP, we added photolyzable caged- Ca^{2+} (DM-Nitrophen) to the patch solution and uncaged Ca^{2+} before and after application of OxA or PE. This produced ai-sAHP-like currents only after the OxA or PE-mediated depolarizing current had developed and both the depolarizing current and ai-sAHP were blocked by subsequent application of Naspm. Finally, to test if this novel sAHP augments spike-frequency adaptation (SFA) like the classical sAHP, we tested if OxA altered the ability of DR^{5-HT} neurons to follow pulse trains at different frequencies (0.5 - 10 Hz). We found that OxA produced a delayed reduction in firing fidelity even at low frequencies and that this effect was blocked by application of Naspm. Collectively, our findings indicate: 1) GluD channels underlie the OxA and PE induced depolarizing current, 2) Ca^{2+} plays a previously unknown role in regulating GluD current, 3) Ca^{2+} influx suppresses the GluD current to produce a novel ai-sAHP and 4) this ai-sAHP enhances SFA like the classical slow-AHP.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

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Program #/Poster #: PSTR261.17/C7

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: NIGMS-SURE (R16-GM145475)
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NIGMS-RISE R25-GM061838
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5R25-GM061151-21

Title: Reward-associated cues reduce h-current amplitude in midbrain dopamine neurons

Authors: *C. C. CALO¹, K. Y. BOSQUE CORDERO⁴, J. CAPELLA², K. LABOY⁵, R. VAZQUEZ-TORRES⁶, O. VÉLEZ ACEVEDO⁷, D. CONSUEGRA GARCIA⁸, C. A. JIMENEZ-RIVERA³;

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Abstract: Cocaine is a psychoactive substance that targets brain regions involved in motivation and reward learning. Ventral tegmental area (VTA) dopamine (DA) neurons play a critical role in these processes, yet their intrinsic modulations during drug and non-drug self-administration or reward-associated cue learning remain poorly understood. This study employed an Intermittent Access (IntA) cocaine model, both contingent and non-contingent, to examine how reward delivery and associated cues modulate the hyperpolarization-activated cyclic nucleotide-gated (HCN) current (I_h), an intrinsic property regulating neuronal physiology. Rats were divided into saline and cocaine IntA groups, with yoked controls receiving non-contingent cocaine infusions either paired (Yoked + cue) or unpaired (Yoked - cue) with a light cue to control for contingency and cue exposure. A parallel sucrose self-administration cohort served as a non-drug reward control. Whole-cell patch-clamp recordings in VTA slices revealed reduced I_h amplitude in the cocaine IntA and Yoked-cocaine + cue groups, accompanied by a hyperpolarizing voltage shift in all cocaine-treated animals. Cocaine IntA increased synaptic integration, while both IntA and Yoked-cocaine + cue groups exhibited reduced membrane capacitance (C_m). Notably, similar I_h reductions were observed in sucrose IntA and Yoked-sucrose + cue groups. These learning-associated changes may enhance DA neurons' ability to signal reward anticipation or saliency. We propose that I_h modulation in VTA DA neurons maintains intrinsic excitability, improves signal-to-noise ratio, and facilitates learning of reward-salient cues—processes essential for motivation toward drug and non-drug rewards. This hypothesis provides insight into how intrinsic plasticity in VTA DA neurons shapes reward learning.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.18/C8

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Title: Investigating Ketamine's Impact in Neuronal Excitability and Synaptic Activity for Translational Neuroscience Applications

Authors: *M. HE, S. GRIGORYEV, D. LIU, C. PETROSKI, R. E. PETROSKI;
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Abstract: Ketamine, a dissociative anesthetic with rapid-acting antidepressant effects, has demonstrated clinical efficacy in major depressive disorder (MDD) and other neuropsychiatric conditions. While its role as a noncompetitive NMDA receptor antagonist is well established, the systems-level mechanisms by which ketamine modulates neural circuits remain poorly defined. At therapeutic doses, ketamine selectively blocks NMDA receptors, reducing excitatory neurotransmission. Ketamine is thought to preferentially inhibit NMDA receptors on GABAergic interneurons, leading to disinhibition of excitatory neurons. Over time, this increased glutamate in the synaptic cleft and enhanced AMPA receptor activity are thought to drive synaptic plasticity—including elevated BDNF release, mTOR signaling, dendritic spine formation, and synaptogenesis—particularly in the prefrontal cortex. However, identifying a quantifiable and reproducible functional endpoint of ketamine's action remains essential for advancing both basic research and clinical translation. In this study, we performed whole-cell patch-clamp recordings to assess spontaneous action potentials and miniature excitatory postsynaptic currents (mEPSCs) in cortical neurons across a range of ketamine concentrations. These studies should provide insights into the mechanism of ketamine on network activity and excitatory-inhibitory balance. Such metrics are critical for evaluating therapeutic efficacy, optimizing dosing strategies, and understanding how drugs reshape cortical circuit dynamics in health and disease.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Title: Comparative electrophysiological characterization of micromagnetic and electrical stimulation in ex vivo bullfrog sciatic nerve

Authors: *M. S. HOPPER¹, D. TONINI¹, I. ULLAH², Y. YASSIN², R. SAHA¹, W. C. LOW³, D. LIAO², J.-P. WANG¹;

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Abstract: Micromagnetic stimulation (μ MS) is an emerging modality for evoking neural responses using time-varying magnetic fields generated by microcoils. This technique enables stimulation without an electrochemical interface and offers orientation-dependent selectivity via directional induced electric fields, but its effects on peripheral nerve activation dynamics remain underexplored. In this study, we characterize compound action potential (CAP) responses evoked by magnetic and electrical stimulation in the *ex vivo* bullfrog (*Rana catesbeiana*) sciatic nerve using custom solenoidal microcoil devices (“MagPen”) and standard stainless steel electrodes. Stimuli were delivered in a nerve chamber optimized for electrophysiological recording. We assessed responses to sinusoidal waveforms with varying phase angles and measured CAPs with downstream electrodes. Magnetic stimulation evoked reproducible CAPs with shorter stimulus-to-response delays and increased amplitudes relative to electrical stimulation at matched thresholds, suggesting distinct recruitment dynamics. The strength-duration relationship for magnetic stimulation exhibited broader pulse-width dependence. Recovery cycles showed delayed excitability restoration under magnetic stimulation, with reliable CAP re-emergence by 10 ms. Waveforms at intermediate phase angles (e.g., 90° and 270°) produced stronger responses than pure 0° or 180° sinusoids, likely due to sharper onset/offset transitions that enhance induced field strength. These results demonstrate that μ MS activates peripheral nerves with distinct spatiotemporal and waveform-dependent properties compared to electrical stimulation. This supports its potential for orientation-selective stimulation that avoids electrochemical interfaces and enables detailed investigation of nerve conduction dynamics.

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Poster

PSR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Program #/Poster #: PSTR261.20/C10

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Title: Investigating dopaminergic modulation of subpopulations in entopeduncular nucleus

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Abstract: Dysregulated midbrain dopamine (DA) signaling is central to the pathophysiology of Parkinson's Disease (PD). Nonetheless, the role of extrastriatal pathways, particularly the entopeduncular nucleus (EP), remains underexplored. The EP, the rodent homolog of the human globus pallidus internal segment (GPi), is a key output nucleus of the basal ganglia. In PD, the loss of dopaminergic neurons in the substantia nigra pars compacta (SNc) leads to altered firing frequency, pattern, and spike-time entropy in EP neurons. DA modulates input from both the striatum and the external globus pallidus (GPe) onto EP neurons, integrating direct and indirect pathway information. Disruption of this modulation likely contributes to PD pathology, though the cellular mechanisms remain unclear. To investigate how DA directly influences EP activity, we performed ex vivo whole-cell current-clamp recordings to characterize the intrinsic properties of EP neurons. Recordings were collected under baseline conditions and during bath application of DA to assess changes in spontaneous and evoked activity. To further examine effects in distinct EP subpopulations, we employed a transgenic and viral strategy targeting projection-defined and molecularly distinct cell types. Specifically, we used PV-Cre mice with GFP expression in EP neurons and injected a retrograde tracer into the lateral habenula (LHb) or motor thalamus. This allowed us to label LHb-projecting neurons and thalamic-projecting GABAergic neurons thought to form distinct EP limbic and sensorimotor circuits. This approach enabled cell-type-specific analysis of DA modulation in functionally distinct EP circuits to better characterize dopamine modulation of EP subsets. Understanding how dopamine modulates these two populations will contribute to our understanding of how dysregulated dopamine signaling contributes to limbic and motor symptoms of PD.

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Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

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Program #/Poster #: PSTR261.21/C11

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: NIH Grant R01-DK138834

Title: The membrane excitability properties of GUCY2C-high neuropod cells in the gut-brain axis

Authors: *K. J. KENNEDY¹, A. LONDREGAN², S. WALDMAN², M. L. COVARRUBIAS³;
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Philadelphia, PA

Abstract: A subtype of small intestine neuropod cells (NPCs) expressing high levels of membrane bound guanylyl cyclase 2C (GUCY2C-high) play a key role in visceral pain (VP) modulation by clinically used GUCY2C agonists, such as linaclotide, and may form chemical synapses with submucosal sensory afferents of the dorsal root ganglion (DRG). Our previous work showed that DRG neurons co-cultured with intestinal crypts become hyperexcitable when in contact with GUCY2C-high NPCs in the absence of GUCY2C ligands. Furthermore, upon exposing the co-culture to linaclotide, these DRG neurons become hypoexcitable. This modulation is specifically dependent on GUCY2C expression in GUCY2C-high NPCs. Towards elucidating how GUCY2C ligands modulate GUCY2C-high NPCs and their interaction with DRG neurons, we are characterizing the excitable membrane properties of GUCY2C-High NPCs derived from adult mouse crypts from the small intestine, dissociated and plated on Matrigel-coated coverslips. Whole-cell current-clamp recordings demonstrate that GUCY2C-high NPCs (n=8) have healthy resting membrane potentials (-63.3 ± 4.3 mV) and, relative to typical neurons, they exhibit high input resistance (5.7 ± 1.6 G Ω), low capacitance (3.9 ± 0.5 pF), low rheobase (16 ± 5.6 pA), and generally fire single action potentials that are relatively short (73.4 ± 3.1 mV). Under whole-cell voltage-clamp conditions (n=8), GUCY2C-high NPCs exhibit a fast inactivating early inward current ($I_{\max} = -99 \pm 32.5$ pA at 10 mV) and a delayed outward current ($I_{\max} = 1005.3 \pm 154.3$ pA at 90 mV) which can be eliminated upon replacing Cs for K in the internal solution (n=5), leaving a robust fast inactivating inward current ($I_{\max} = -1124.8 \pm 164.8$ pA at 10 mV). Overall, these results demonstrate that GUCY2C-high NPCs display neuron-like excitability and the expected Na and K membrane conductance. Supporting the importance of GUCY2C-high NPCs' membrane excitability, preliminary experiments also showed the optogenetic hyperpolarization of the GUCY2C-high NPCs membrane potential with archaerhodopsin recapitulates the silencing effect of linaclotide on DRG neurons.

Disclosures: K.J. Kennedy: None. A. Londregan: None. S. Waldman: None. M.L. Covarrubias: None.

Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.22/C12

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: NIH Grant MH131857
NIH Grant MH122810
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Child Neurology Society Philip R. Dodge Young Investigator Award

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Title: Hcn currents distinguish mediodorsal thalamic subnuclei

Authors: G. J. HECKLER¹, G. J. ORDEMANN², K. SPRINGER³, F. DRIVER⁴, A. HARRIS⁶, A. C. JACKSON⁵, *A. C. BRUMBACK⁷;

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Abstract: The mediodorsal thalamus (MD) coordinates higher-order functions including working memory, cognitive flexibility, and sleep regulation. The MD can be subdivided into medial (MD-M), central (MD-C), and lateral (MD-L) regions based on anatomical location, cytoarchitecture, and synaptic connectivity. We recently found that MD-M neurons projecting to medial prefrontal cortex (mPFC) in adult mice have decreased hyperpolarization-activated cyclic nucleotide-gated (HCN) channel activity compared to MD-L neurons, which causes differential intrinsic excitability between these two subregions.

Here, we used patch-clamp electrophysiology, fluorescence in situ hybridization, and immunohistochemistry to investigate how HCN function and subunit expression impact oscillatory activity across MD-M, MD-C, and MD-L. We found HCN-related differences across all three subnuclei in both current and voltage-clamp recordings. Clustering neurons by HCN properties alone accurately assigned neurons to their correct subnucleus with >95% accuracy. Greater HCN activity correlated with decreased input resistance, reduced action potential firing, and increased resonant frequency. These findings link network rhythmicity to intrinsic cellular properties that shape circuit dynamics within the prefrontal thalamocortical network.

Disclosures: G.J. Heckler: None. G.J. Ordemann: None. K. Springer: None. F. Driver: None. A. Harris: None. A.C. Jackson: None. A.C. Brumback: None.

Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR261.23/C13

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: R01CA268125

Title: Does OX induce spontaneous firing of motoneurons by increasing intrinsic excitability?

Authors: *K. BUI¹, P. NARDELLI¹, N. HOUSLEY¹, T. C. COPE²;

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Abstract: Chemotherapy-induced peripheral neuropathy is a common, dose-limiting side effect of platinum-based drugs like oxaliplatin. Acute symptoms occurring as early as one day after oxaliplatin (OX) infusion include muscle cramping associated with spontaneous fasciculations. In preclinical study of rats, we find that OX-induced fasciculations arise from spontaneous firing originating in motoneurons (MNs) at sites within the central nervous system. The present study tested for biophysical expressions of increased excitability in central regions of MN action potential generation. We performed *in vivo* intracellular recordings from spinal motoneurons 24 hours after IP injection of OX (30 mg/kg). MNs corresponding to triceps surae muscle movement were targeted for current-clamp recordings. Recordings were obtained from seven MNs, five of which exhibited spontaneous activity. MNs with spontaneous activity ($n = 5$) had an average rheobase of 19.6 ± 6.6 nA, while those without spontaneous activity ($n = 2$) had a similar rheobase of 17.5 ± 11.0 nA. Five MNs generated an evoked response following a 6 nA inward current injection above rheobase. Among these, MNs with spontaneous activity ($n = 3$) demonstrated an average firing rate of 96.4 ± 58.2 pulses per second (pps) and a spike count of 329.3 ± 109.7 . In comparison, MNs without spontaneous activity ($n = 2$) had an average firing rate of 80.0 ± 33.8 pps and a spike count of 211.5 ± 89.8 . In summary, acute OX treatment elicited spontaneous activity in the majority of recorded MNs, with 5 out of 7 exhibiting such activity. These MNs show no changes in rheobase values or altered excitability in response to current injection, as evidenced by similar firing rates and spike counts compared to MNs without spontaneous activity. These early findings suggest that OX-induced spontaneous activity may be driven by extrinsic factors, such as increased synaptic input, rather than changes within the MNs themselves. Increasing sample size should provide sufficient confidence regarding these preliminary indications that acute OX treatment does not affect the intrinsic excitability of motoneurons in segments residing within the central nervous system.

Disclosures: K. Bui: None. P. Nardelli: None. N. Housley: None. T.C. Cope: None.

Poster

PSTR261: Intrinsic Properties and Modulation of Neuronal Firing I

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Program #/Poster #: PSTR261.24/C14

Topic: B.06. Intrinsic Membrane Properties and Signal integration

Support: R24MH114785
Sabbatical Program at JHU/APL

Title: Leveraging Connectomics to Map the Trophospongium, a Putative Morphological Marker for the Spike Initiation Zone, across *Drosophila* Neurons

Authors: *P. K. RIVLIN, J. ELLIS-JOYCE, J. MATELSKY, B. A. WESTER;
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Abstract: Comprehensive computational models of brain function require accurate representations of neuronal anatomy and physiology to simulate sensory processing and inter-regional information flow. A key parameter in such models is whether a neuron transmits signals via action potentials (spikes) or graded potentials. While electrophysiological recordings remain the gold standard for determining spiking behavior, many *Drosophila* neurons are either experimentally inaccessible or lack electrophysiological characterization, creating significant gaps in our understanding. To address this limitation, we aim to develop a machine learning (ML) framework to predict spiking behavior based on anatomical features extracted from connectomic data. As a first step, we are generating a database of neurons that contain a trophospongium (TPS), a glial specialization associated with the axon hillock of locust motor neurons (Hoyle et al. 1986), which we propose as a morphological marker for the spike initiation zone (SIZ) of *Drosophila* neurons. In the *Drosophila* hemibrain connectome (Scheffer et al. 2020), the TPS can be recognized by its distinct morphology and co-localizes with electrophysiologically identified spike initiation zone (SIZ) of the DM1 neuron in the antennal lobe (e.g., Gowens & Wilson 2009), supporting its potential role as a structural correlate of neuronal excitability. By systematically identifying TPS-containing neurons and correlating this anatomical marker with electrophysiological data, when available, our approach aims to enable scalable classification of spiking versus non-spiking neurons in the *Drosophila* brain. We present our progress in mapping TPS-containing neurons in the hemibrain and other *Drosophila* connectomes amenable to this analysis, laying the groundwork for data-driven prediction of neuronal signaling at scale.

Disclosures: P.K. Rivlin: None. J. Matelsky: None. B.A. Wester: None.

Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Program #/Poster #: PSTR262.01/C15

Topic: B.08. Epilepsy

Support: F31AA031916A (PA-23-271)
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2R01AA015614-16
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5R00AA025120-04

Title: Epileptiform discharges in the retrosplenial-hippocampal circuit following third-trimester alcohol exposure

Authors: *A. MYRICK¹, S. A. MCKENZIE², C. F. VALENZUELA³, D. N. LINSENBARDT⁴;

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Abstract: PURPOSE: Third trimester alcohol exposure (TTAE) damages the hippocampal-diencephalic-cingulate (HDC), a critical neural circuit for learning and memory. Changes in the function of the HDC are therefore speculated to drive differences in learning and memory observed in certain individuals with prenatal alcohol exposure. Because an anatomical center within the HDC network known as the retrosplenial cortex is thought to be a key mediator of memory consolidation, the current work assessed the impact of TTAE on electrophysiological events associated with memory consolidation during sleep known as sharp-wave ripples. In the process of this work, we discovered epileptiform discharges occurring through the retrosplenial cortex and hippocampus; large amplitude local field potential events generally associated with neurodegeneration or epilepsy. METHODS: Male and female TTAE and control (saline) mice were produced using a single day, double alcohol injection model (2.5 g/kg) at postnatal day 7. After postnatal day 60, a silicone electrode was implanted into the retrosplenial cortex and hippocampus. Following recovery, home cage electrophysiological recordings were collected in 24-hour epochs following a daily learning and memory task called the Barnes maze. RESULTS: We indeed observed sharp-wave ripples during epochs of sleep, however, we were extremely surprised to also observe frequent, large-amplitude events resembling epileptiform discharges; events which occurred at a much higher rate among TTAE mice. Much like sharp-wave ripples, these epileptiform discharges occurred more frequently during non-rapid eye movement sleep, and were observed in the hippocampus as well as the retrosplenial cortex. In addition to significantly more epileptiform discharges in the TTAE group, the average amplitude of these events within the dentate gyrus region of the Hippocampus were significantly lower in TTAE than in the saline group. Additionally, within the TTAE group alone, the dentate gyrus events had significantly lower average amplitude compared to the corresponding events in the retrosplenial cortex. CONCLUSIONS: These results identify and characterize HDC epileptiform discharges occurring at a high rate among TTAE mice. The occasional event being detected in the saline group may indicate an endogenous event that is amplified by TTAE. Together, this data represents a potential mechanism for learning and memory differences experienced by individuals exposed prenatally to alcohol which may be useful for developing therapeutic and supportive clinical routes.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Topic: B.08. Epilepsy

Support: UC Irvine Chancellor's Postdoctoral Fellowship to J.M.Y.
A.P Giannini Postdoctoral Research Fellowship to J.M.Y.
NIH R01 1R01NS128222-01 to L.A.E.

Title: Preliminary evidence that spontaneous generalized seizures following PTEN deletion in the adult dentate gyrus are preceded by bouts of abnormal circuit activity

Authors: *J. M. YONAN¹, B. L. BOUBLIL², L. A. EWELL³, O. STEWARD⁴;

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Abstract: Overactivation of the mechanistic target of rapamycin (mTOR) pathway during development is associated with intellectual disability and seizures in humans. Previous reports from our lab reveal that deletion of phosphatase and tensin homolog (PTEN), an upstream negative regulator of the mTOR pathway, in the adult dentate gyrus initiates dramatic growth of mature neurons. This growth is characterized by progressive enlargement of granule cell bodies, apical dendrites, and mossy fiber axons over a period of 6 months following PTEN deletion, suggesting greatly increased neuronal connectivity with PTEN-expressing pre- and post-synaptic partners. Moreover, mice with unilateral PTEN deletion develop spontaneous seizures with a latency of approximately 2 months. Here, we report initial findings using high density single unit recordings in freely moving mice to explore circuit activity during the latent period preceding spontaneous seizures. Male and female PTEN^{tf/f};Rosa^{tdTomato} mice received intra-dentate injections of AAV-Cre at 2 months of age. Cre expression deletes PTEN and simultaneously induces tdTomato expression in transduced cells. Control mice received similar injections of AAV-GFP. 3-4 weeks post injection, mice were implanted with a tetrode drive targeted to the dentate gyrus within the area of PTEN deletion. Tetrode recordings from the hippocampus were performed daily for two weeks during open field foraging. Recordings revealed prolonged bouts of abnormal neuronal activity including interictal spikes and pathological high frequency oscillations during periods of both rest and exploration in PTEN-deleted mice. No abnormal neuronal activity was observed in mice injected with a control vector. These results suggest that pathological hypersynchronous activity occurs within the hippocampus coinciding with periods of neuronal growth and circuit remodeling due to vector-mediated PTEN deletion in the adult dentate gyrus prior to the onset of spontaneous seizures. Ongoing studies are analyzing whether place maps are altered during epileptogenesis in this model of adult-onset epilepsy.

Disclosures: J.M. Yonan: None. B.L. Boubilil: None. L.A. Ewell: None. O. Steward: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); OS is a co-founder, scientific advisor, and has economic interests in the company Axonis Inc, which is developing novel therapies for spinal cord injury and other neurological disorders..

Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR262.03/C17

Topic: B.08. Epilepsy

Support: NIH Grant 1R01NS112538 (Lillis)
CURE Taking Flight (Lau)

Title: In vivo neuronal resolution imaging of hippocampal neuronal activity in the intrahippocampal kainate mouse model of temporal lobe epilepsy

Authors: *L. A. LAU¹, P. O'GORMAN¹, K. J. STALEY², K. P. LILLIS³;
²Neurol., ¹Massachusetts Gen. Hosp., Boston, MA; ³Neurol., Harvard Med. School, MGH, Charlestown, MA

Abstract: Epilepsy affects 1% of the population and is characterized by recurrent spontaneous seizures. To gain new insights into the cellular drivers of seizure initiation will require high resolution recordings of neuronal activity during seizures. We have previously employed calcium imaging with the genetically encoded calcium indicator GCaMP to capture spontaneous seizure-like activity in an in vitro model of post-traumatic epilepsy. This work revealed that seizures spread along repeatable trajectories but were stochastic at the level of individual neurons. To extend these findings to an in vivo model of epilepsy, we are now performing in vivo calcium imaging in the intrahippocampal kainate (IHK) mouse model of temporal lobe epilepsy. IHK produces a focal injury near the site of kainic acid injection, with seizures reliably initiating near the site of kainic acid injection. To study neuronal activity in the relevant ictogenic region, we have employed a method for large field of view, high resolution calcium imaging of hippocampal neurons in vivo. This method combines hippocampal injection of AAV soma-GCaMP8, with the implantation of a 3 mm wide imaging cannula into an area of aspirated cortex. This method has allowed for 2-photon imaging of calcium activity of hundreds of individual CA1 hippocampal neurons, in an awake, head-fixed mouse. This method will significantly advance our ability to study seizure generating mechanisms in an in vivo model of chronic epilepsy.

Disclosures: L.A. Lau: None. P. O'Gorman: None. K.J. Staley: None. K.P. Lillis: None.

Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR262.04/C18

Topic: B.08. Epilepsy

Support: Colorado State University

Title: Mouse behavioral seizures and the use of machine learning models for their detection and analysis

Authors: *H. KIM¹, H. SEO², R. LEE¹, S.-H. LEE¹, S. KIM¹;
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Abstract: Epilepsy is a widespread brain disease impacting all age groups and affects nearly 50 million people according to the World Health Organization. Consequently, significant experimental and theoretical research attempts to find the mechanisms underlying the abnormal (and transient) brain electrical activity caused by epilepsy to facilitate the development of therapeutic solutions. With no appropriate treatments for seizures, understanding epileptogenesis is essential for therapeutic development. Seizures are caused by abnormally synchronous brain activity that can result in changes in muscle tone. These behavioral seizures are detected by visual inspection. The Racine scale, the most widely used seizure scoring systems in preclinical models, in rodents, is used to quantify seizure intensity scores. However, visual inspection is time-consuming, low-throughput, and partially subjective, and there is a need for rigorously quantitative approaches that are scalable. Here, we used supervised machine learning approaches to develop automated classifiers to detect behavioral seizures directly from video and movement data. Using the kainate (10 mg/kg)-induced seizure model in mice, we trained video and movement data from a 3-axis accelerometer to detect seizure events. Our findings demonstrate that supervised machine learning approaches can be used to accurately quantify behavioral seizure events videos and 3-axis movement data of mice in their home cage. These results enable high-throughput, noninvasive, and standardized seizure scoring to further understand epileptogenesis and develop therapeutic discovery.

Disclosures: H. Kim: None. H. Seo: None. R. Lee: None. S. Lee: None. S. Kim: None.

Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR262.05/C19

Topic: B.08. Epilepsy

Title: Integrated in vitro and in vivo Strategy for Accelerated Drug Discovery for Epilepsy

Authors: *X. GUO;

ICE Biosci., South San Francisco, CA

Abstract: Effective translation of anti-seizure compounds is hindered by the low predictive value of traditional single-model screens. We present an NIH/NINDS-aligned, industry-ready capability that integrates target-based assays, neural-network phenotyping, and tiered in-vivo validation to accelerate discovery of drug-resistant epilepsy therapies. **Methods:** small molecules were screened against ion channel such as KCNQ2/3 openers, Nav1.6 blockers and GABAA potentiators using automated patch-clamp or manual patch clamp assay. Hits (XC50 around the reasonable range, e.g. less than 10 nM) underwent parallel CNS-DMPK profiling (MDCK-MDR1 permeability, microsomal stability, rat Kp,uu) and brains slice recordings of cortical-hippocampal networks (Such as 4-aminopyridine-evoked model). Compounds reducing burst rate significantly without suppressing baseline activity advanced to acute seizure tests (MES, PTZ, 6-Hz model) followed by lamotrigine-resistant amygdala kindling and post-kainate

spontaneous-seizure models with continuous EEG recording. **Conclusions:** Combining orthogonal in-vitro, ex-vivo, and rigorous in-vivo assays enriches for brain-penetrant, mechanistically diverse anti-seizure leads and lowers attrition versus historical ETSP benchmarks. This scalable external-CRO platform, fully aligned with NIH priorities, offers partner programs a rapid route to next-generation therapies for refractory epilepsy.

Disclosures: X. Guo: None.

Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR262.06/Web Only

Topic: B.08. Epilepsy

Title: Temporal Lobe Epilepsy and Dissociative Phenomena: A Clinical and Neurobiological Review with Emphasis on Overlap with Dissociative Identity Disorder

Authors: *L. VASCONCELOS CAETANO¹, B. SOUZA², C. CALDERONI³, G. MARROQUES NOLETO⁴, T. SILVA⁵, L. OLIVEIRA PINHEIRO⁶, S. DA SILVA SÃO JOSE⁶, V. BASTOS⁷, R. PEREIRA⁸, C. DIAS PEREIRA⁹, E. DUARTE³, M. CORRÊA GONDIM BEZERRA RODRIGUES¹⁰, K. SANTIAGO DIAS¹¹;

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Abstract: Temporal Lobe Epilepsy (TLE), a focal epilepsy, is characterized by a complex clinical presentation and a broad range of psychiatric symptoms, including dissociative phenomena resembling Dissociative Identity Disorder (DID). This work consists of a systematic review based on PRISMA guidelines, conducted through the databases PubMed, Cochrane, Scopus, Embase, and Web of Science, using the MeSH/DeCS descriptors “Temporal Lobe Epilepsy” and “Dissociative Disorders.” Articles presenting evidence on the association between TLE and dissociative manifestations—such as depersonalization, multiple identities, and diagnostic overlap with dissociative disorders, especially DID—were included. Eligible articles were assessed using the Joanna Briggs Institute tool. Literature reviews were excluded. Thirty-eight articles were identified: 12 from Embase, 26 from Scopus, and none from other databases. Two duplicate articles and 31 others were excluded according to the criteria, resulting in five eligible studies. It was demonstrated that déjà vu, jamais vu, dream states, and out-of-body experiences may be related to ictal activity in medial temporal structures, such as the hippocampus, amygdala, and parahippocampal gyrus—areas involved in mesial temporal lobe

epilepsy (MTLE), the most common subtype of TLE. Among 1,245 patients, a longer time to diagnosis by video-electroencephalogram (video-EEG) was observed in cases with typical TLE symptoms, and a shorter time in patients with dissociative symptoms associated with depression, anxiety, and migraine. Furthermore, patients with refractory TLE showed poorer performance in prospective memory and executive functions, correlating frontal inhibitory deficits with dissociative presentations. Finally, a case initially interpreted as possession and later diagnosed as TLE was described, highlighting the risk of underdiagnosis in cultural contexts. In summary, there is significant clinical overlap between refractory TLE and dissociative phenomena, especially with MTLE and DID. However, typical TLE symptoms combined with dissociative symptoms may delay diagnosis, while depersonalization and multiple identities might facilitate it, emphasizing the importance of an intersystemic approach.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Taubman Institute Emerging Scholar Award - Kenneth Eisenberg
Emerging Scholar
UMMS Research Scouts Award
Dravet Syndrome Foundation Postdoctoral Fellowship

Title: Noradrenergic Circuit Response to Hyperthermia-evoked Seizures in the *Scn1a*^{+/-} Mouse Model

Authors: *M. ZHAO, A. NOOR, J. BARDEN, C. RANA, C. BURGESS, J. MATTIS;
Univ. of Michigan, Ann Arbor, MI

Abstract: Locus coeruleus (LC) neurons are the main source of norepinephrine (NE) released to most brain structures, including the seizure-prone neocortical and limbic regions, regulating numerous physiological progresses including arousal, attention, and memory. Previous evidence suggests that noradrenergic neurons may be involved in epilepsy and that their activation may be therapeutic for seizures. However, the activity of LC-NE network during seizures remains largely unclear. We directly investigated the response of noradrenergic networks to seizures by using *Scn1a*^{+/-} mice, a well-characterized pre-clinical model of Dravet Syndrome in which

seizures can be evoked with hyperthermia. We performed electrocorticography recordings concurrently with fiber photometry and two-photon recordings to measure (1) the activity of LC noradrenergic neurons and (2) downstream NE neurotransmission during seizures. We found that LC noradrenergic neurons were inhibited at the populational level during hyperthermia-induced seizures but, surprisingly, downstream NE release was dramatically elevated. In both datasets, we confirmed the absence of these signal changes in wild-type littermate control mice that also underwent hyperthermia, and in heating trials of *Scn1a*^{+/-} mice that failed to evoke seizures. To reconcile these discordant findings, we tested and validated the Glutamate Amplifies Noradrenergic Effects (GANE) theory, in which excitatory activity in downstream regions locally evokes NE release, independent of somatic LC activation. In sum, our result has identified an intriguingly discordant finding: seizures are associated with a decrease in LC-NE neuronal activity, but an acute increase in downstream NE levels, likely triggered by local neuronal activation of NE release. Future work will examine the functional implications of these seizure-associated disruptions to NE circuitry.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Topic: B.08. Epilepsy

Support: NIH NINDS R01NS133169

Title: Bnst seizure-activated neurons modulate breathing circuitry and fatality during sudep

Authors: *S. MULLOY¹, E. HONG², J. QIN³, E. XU⁴, W. P. NOBIS⁵;

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Abstract: Sudden Unexpected Death in Epilepsy (SUDEP) is the most common cause of death in refractory epilepsy patients, yet the underlying mechanisms are unclear. Our work has shown that the Bed Nucleus of the Stria Terminalis (BNST) increased c-fos expression after seizures with respiratory failure, and blocking BNST improves seizure mortality. Therefore, the BNST may play a critical role in seizure-related respiratory dysfunction through modulation of respiratory brainstem circuitry. Male and female DBA1 seizure primed mice were used in an audiogenic seizure model (AGS). Fiber photometry (FP) was used to monitor BNST calcium-dependent neural activity in vivo. BNST activity was recorded during hypercapnia (5 and 7% CO₂) and hypoxia (8% O₂) concurrently with whole body plethysmography (WBP) to measure hypercapnic and hypoxic ventilatory response (HCVR and HVR), and during seizure induced respiratory arrest. A virally mediated Targeted Recombination in Active Population (TRAP)

strategy was used to selectively target BNST neurons active during AGS induced respiratory arrest. The TRAP virus was paired with a cre-dependent viral vector expressing fluorescent synaptophysin to identify functional projections of seizure-activated BNST neurons. It was also paired with a cre-dependent channel rhodopsin to optogenetically modulate BNST terminals on neurons in brainstem respiratory regions. Intrinsic excitability of seizure-activated BNST neurons was recorded compared to non-activated neurons using whole-cell patch clamp. During gas exposure, mice showed HCVR and HVR with respiratory response correlated to FP. As measured by FP, BNST in vivo activity decreased during hypercapnia but showed no response during hypoxia (n=4 mice, 3 trials/mouse). BNST activity was drastically increased during AGS preceding respiratory failure and death. Seizure-activated BNST neurons formed synapses in the respiratory brain regions of the nucleus tractus solitarius (NTS), parabrachial nucleus of the pons (PBN), and periaqueduct gray (PAG) (n=4 mice). They also had higher resting membrane potential compared to non-activated BNST neurons. Optogenetic stimulation of BNST seizure-activated terminals on NTS neurons did not produce either inhibitory or excitatory currents (n=1 mouse, 5 cells), but work characterizing other respiratory brainstem regions is still ongoing. Overall, our work suggests that seizures activate BNST neurons that project to respiratory brainstem regions, and that BNST activity is altered by ventilatory challenges, suggesting this maybe a potential modulation target for SUDEP.

Disclosures: S. Mulloy: None. E. hong: None. J. Qin: None. E. Xu: None. W.P. Nobis: None.

Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR262.09/C22

Topic: B.08. Epilepsy

Support: R21NS111389
R01NS126418
LB692
G20RR024001

Title: Development and characterization of an autoresuscitation assay for preclinical SUDEP models

Authors: *J. HINMAN¹, S. IYER³, S. DRAVES¹, S. A. MATTHEWS², K. A. SIMEONE⁴, T. A. SIMEONE⁵;

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⁵Pharmacol., Creighton Univ. Sch. of Med., Omaha, NE

Abstract: Approximately 1:1000 people with epilepsy die of Sudden Unexpected Death in Epilepsy (SUDEP) annually. Cardiorespiratory dysfunction has been implicated in SUDEP, but the underlying mechanisms remain elusive. Terminal events have been reported to result from

prolonged apnea suggesting a failure to autoresuscitate, which involves gasping and return to eupnea. To investigate the efficacy of the autoresuscitation reflex and potential contribution to SUDEP, we adapted an anoxia-induced autoresuscitation challenge for use in the SUDEP field. The challenge was optimized to allow majority survival of wild-type mice regardless of age. We tested *Kcna1*^{-/-} mice, a SUDEP model, at two different ages corresponding to low-risk and high-risk SUDEP susceptibility. As SUDEP risk increased in *Kcna1*^{-/-} mice, autoresuscitation failure and mortality increased. During the challenge, *Kcna1*^{-/-} mice had increased breathing frequency, tidal volume, minute ventilation, and peak expiratory flow compared to wild-type mice. Once gasping was initiated, *Kcna1*^{-/-} mice had reduced gasp frequency and duration. Two distinct phases of gasping- Type I and Type II- were identified during the autoresuscitation response. *Kcna1*^{-/-} mice exhibited reduced latency, number, frequency, and time spent in Type II gasps, suggesting potential greater importance for this phase. These data indicate that *Kcna1*^{-/-} mice have enhanced chemosensitivity in response to anoxia, which may contribute to autoresuscitation failure.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR262.10/C23

Topic: B.08. Epilepsy

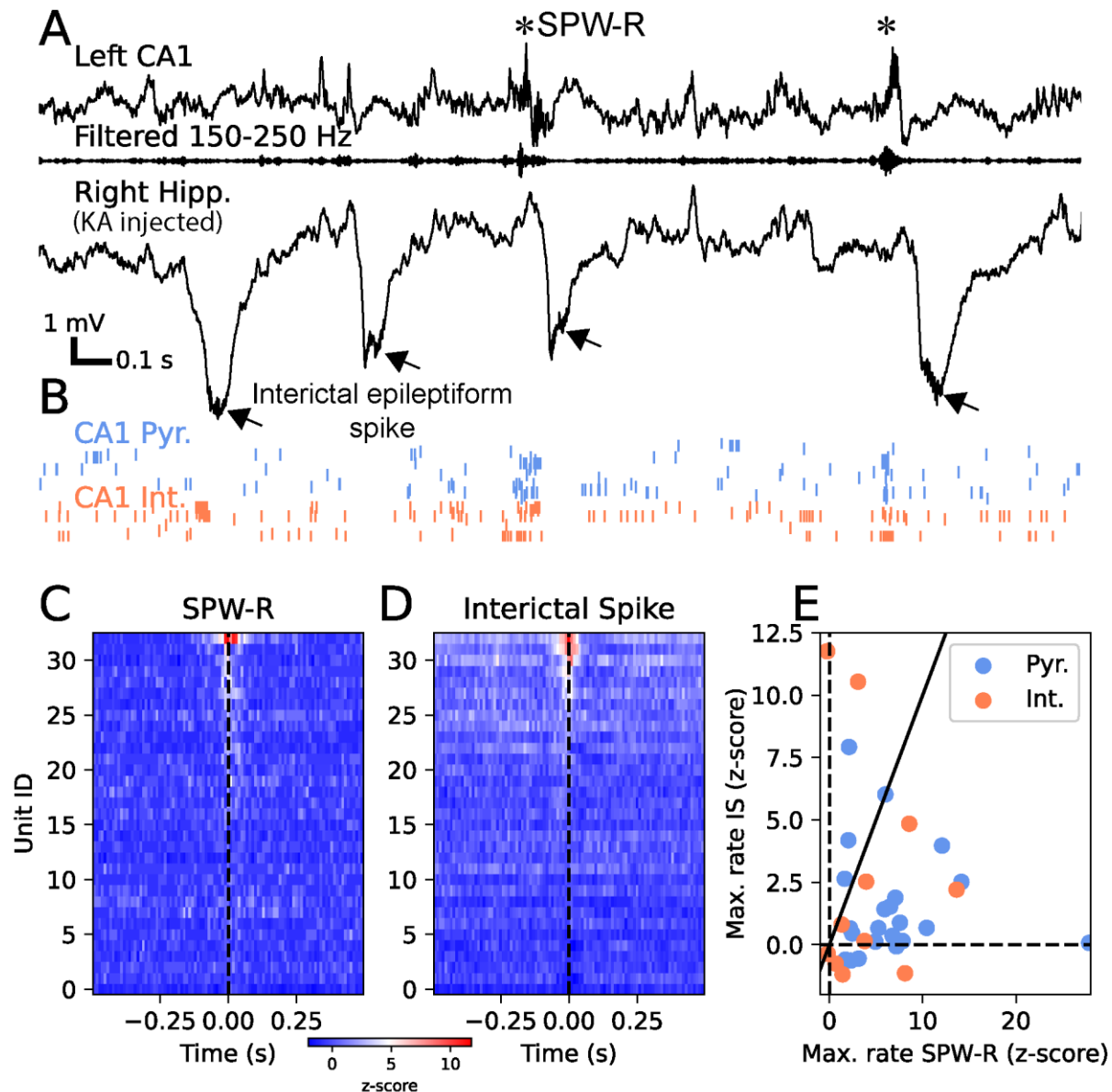
Support: NIH R01 1R01NS128222-01
American Epilepsy Society Grant 835029
VolswagenStiftung Freigeist Fellowship
NINDS T32 5T32NS045540-20

Title: Disrupted interhemispheric hippocampal communication in a mouse model of temporal lobe epilepsy

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Abstract: In rodents, nearly all sharp-wave ripples (SPW-R) are synchronized between the two hippocampal hemispheres. The significance of this synchronization is unclear, and furthermore, it is not known at all whether it is altered in disease states like temporal lobe epilepsy. Furthermore, in temporal lobe epilepsy, hippocampal sclerosis alters the circuitry of the hippocampus to become hyperexcitable, leading to epileptiform high-frequency discharges which are thought to be pathological forms of SPW-R. How these dynamics alter bilateral hippocampal communication is an open question. Using *in vivo* recordings from CA1 in mice

injected with kainic acid (n = 7) or saline (control, n = 6) above the right hippocampus, we observed that lags in SPW-Rs onset time between each hemisphere were distributed significantly differently in each condition (Anderson-Darling test, 2 groups, $A^2k = 159$, p-value < 1×10^{-6}). This finding is consistent with a loss of bilateral coordination of SPW-R timing in focal epilepsy. We also examine preliminary evidence that contralateral CA1 neurons are differentially recruited by contralateral fast oscillations. We use these recordings to explore place coding and replay properties of the contralateral hippocampus in temporal lobe epilepsy. In all, we hypothesize dysregulated interhemispheric communication between hippocampi may partially explain memory impairments seen in patients and animal models.



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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Support: NIH / NIMH R00 MH118425 to JDA
Brain and Behavior Research Foundation 30264 to JDA

Title: Neuron lineage-specific roles of the AMPA receptor GluA2 subunit in the regulation of the synaptic transmission, circuit excitability and seizures

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Abstract: AMPA receptors (AMPA), comprised by GluA1/4 subunits, mediate fast excitatory synaptic transmission throughout the CNS. AMPAR trafficking and function are strongly influenced by subunit composition. In mature mammalian neurons, the vast majority of GluA2 subunits undergo post-transcriptional mRNA editing, which confers specific biophysical properties to AMPARs, including making them calcium-impermeable (CI). In the mature, non-injured hippocampus, CI-AMPA are dominant, even exclusive, in most excitatory neurons. In contrast, CI-AMPA coexist with GluA2-lacking, calcium-permeable (CP) ones at some inhibitory interneurons (INs) (Liu SJ et al. 2007). Mutations in the GluA2 subunit can lead to intellectual disability and seizures in humans (Salpietro V et al., 2019). Similarly, GluA2 knockout mice exhibit increased mortality during the first three weeks of life and severe cognitive and motor deficits (Jia et al., 1996). Mutant mice with defective GluA2 editing present similar deficits and display spontaneous seizures (Konen et al., 2020). AMPARs are expressed in most neuron types and their subunit composition differs across neuron types (Lu et al., 2009). However, previous work has focused almost exclusively on excitatory neurons, which account for the vast majority of AMPAR expressed in the brain. To understand the impact of GluA2 deletion on inhibitory neurons vs principal neurons, in this study we generated GABAergic neuron (GAD2-GluA2 KO) and principal neuron-specific (CaMKII-GluA2 KO) GluA2-deficient mice. We confirmed the specificity of the GluA2 KO using immunofluorescence and synaptic AMPAR EPSC rectification assays. We then evaluated motor function and monitored hippocampal activity with in vivo electrophysiological recordings. Unexpectedly, GAD2-GluA2 KO, but not CaMKII-GluA2 KO mice showed severe motor symptoms, suggesting a necessary role of GluA2 in GABAergic neurons such as cerebellar Purkinje cells or striatal medium spiny neurons. More strikingly, loss of GluA2 in inhibitory neurons, but not in principal neurons, led to pathological interictal activity in the hippocampus and spontaneous generalized seizures. Ongoing work is aimed to pinpoint the specific GABAergic neuron populations and dissect the

molecular and cellular mechanisms underlying the lineage-specific contribution of GluA2 subunits to neuronal excitability and network stability. Altogether, our work identifies GABAergic neurons as key mediators of AMPAR GluA2 subunit function and its role in regulating brain activity.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Program #/Poster #: PSTR262.12/Web Only

Topic: B.08. Epilepsy

Support: SECIHTI (CF-2023-I-539)
VIEP-24-0397
HempMeds® México.

Title: Synergistic Effect of Cannabidiol and Cannabigerol on Seizures Induced by Pentylentetrazole

Authors: *A. PATRICIO-MARTÍNEZ^{1,2}, M. P. VALENCIA APANGO^{1,2}, F. PATRICIO MARTÍNEZ^{1,3}, I. D. LIMON PEREZ DE LEON¹;

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Abstract: Cannabidiol (CBD) and cannabigerol (CBG) are non-psychotropic compounds derived from the *Cannabis sativa* L. plant. CBD has demonstrated a protective effect by reducing seizures associated with excitotoxicity in epilepsy. However, despite its benefits in some patients, it does not fully suppress epileptic symptoms. In contrast, CBG exhibits anti-inflammatory and neuroprotective properties and is known to block voltage-gated sodium channels, suggesting its potential as an alternative therapeutic option for epilepsy. The objective of this study was to evaluate the synergistic effect of CBD and CBG on seizure intensity and frequency in a pentylentetrazole (PTZ)-induced seizure model in rats. Male Wistar rats (270-320 g) were randomly assigned to six experimental groups: VEH + VEH, CBG + VEH, CBD + CBG + VEH, VEH + PTZ, CBG + PTZ, and CBD + CBG + PTZ (n = 8 per group). Animals received daily intraperitoneal doses of CBD (15 mg/kg) and CBG (50 mg/kg) for 14 days; PTZ was administered on day 7 of treatment. Seizure activity was recorded for 45 minutes following PTZ administration, and both frequency and intensity were assessed using the Racine scale. Results showed a statistically significant reduction in the frequency and severity of tonic-clonic seizures in the CBD + CBG + PTZ group. In conclusion, the combination of CBD and CBG

appears to exert a synergistic anticonvulsant effect, potentially enhancing neuroprotection against excitotoxic damage and reducing seizure activity.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Program #/Poster #: PSTR262.13/C25

Topic: B.08. Epilepsy

Support: R01 NS099586-05A1

Title: Metabolic and Electrophysiological Insights into Absence Epilepsy from a Novel GLUT1 Deficiency Model

Authors: *G. NAIK¹, S. HE³, S. KILIANSKI², M. GERDING⁴, N. GUAGLIARDO⁴, B. BUCKLEY¹, M. P. BEENHAKKER², B. G. CONDRON³;
²Pharmacol., ¹Univ. of Virginia, Charlottesville, VA; ³Biol., ⁴Univ. of Virginia, Charlottesville, VA

Abstract: Absence epilepsy is a common pediatric seizure disorder characterized by disruptions in thalamocortical network activity and loss of consciousness. One of its most penetrant genetic causes is GLUT1 Deficiency Syndrome (GLUT1DS), a metabolic encephalopathy caused by mutations in SLC2A1, the gene encoding the glucose transporter GLUT1. Despite this well-defined metabolic cause, how impaired brain glucose transport drives seizure generation remains unclear.

We have developed a blood-brain barrier-specific knockdown model of GLUT1 Deficiency Syndrome (GLUT1DS) that enables direct investigation of brain-specific glucose dysregulation in epileptogenesis, an aspect that cannot be isolated in existing global GLUT1 DS models. Critically, our model reproduces the full range of seizure phenotypes observed in GLUT1DS patients, including not only hallmark spike-and-wave discharges (SWDs) associated with absence seizures but also spontaneous tonic-clonic seizures, which are typically absent in current models of G1DS. Our preliminary observations indicate that homozygous GLUT1 knockdown animals develop earlier and more severe spontaneous seizures, whereas heterozygous animals exhibit spontaneous seizures after a longer latency and with reduced severity. Alongside this EEG characterization, we are currently conducting PET imaging and cerebrospinal fluid (CSF) glucose analyses to further validate the model.

By leveraging GLUT1DS as a genetically and metabolically defined model, our goal is not only to elucidate the ictogenic mechanism of GLUT1DS-related epilepsy but also to use it as a lens through which to better understand the broader metabolic and network dysfunctions that drive absence epilepsy. Hence, future work will focus on electrophysiological analyses of

thalamocortical circuit dynamics underlying the observed absence seizures, and compare it with other absence models to identify shared and distinct pathophysiological mechanisms that can guide therapeutic advances.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

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Topic: B.08. Epilepsy

Support: NIH NINDS T32 NS115724
NIH NINDS K08 NS121464
Taubman Institute Emerging Scholar Award - Kenneth Eisenberg
Emerging Scholar
UMMS Research Scouts Award

Title: Endogenous suppression of nigral GABAergic neurons during hyperthermia-evoked seizures in *Scn1a* haploinsufficient mice

Authors: *C. RANA, L. ZHU, J. BARDEN, R. QURESHI, H. HABERLAND, J. DISLA, J. MATTIS;
Univ. of Michigan, Ann Arbor, MI

Abstract: The substantia nigra *pars reticulata* (SNr), a major output of the basal ganglia, has been identified as a seizure “choke point” given its ability to modulate seizures despite being situated outside the seizure onset zone. Previous studies across multiple different seizure types have demonstrated that SNr activity increases at seizure onset and that inhibition of this activity is anticonvulsant. Patients with Dravet Syndrome (DS), a severe developmental and epileptic encephalopathy, exhibit medication-refractory seizures and progressive motor deficits, but the role of the SNr in DS has not yet been established. In this study, we investigated SNr activity in the haploinsufficient (*Scn1a*^{+/-}) mouse model of DS, quantifying intrinsic cell properties and *in vivo* population activity of SNr parvalbumin-positive (PV⁺) neurons in *Scn1a*^{+/-} mice versus WT littermates: (1) Whole-cell patching revealed that the firing of SNr PV⁺ neurons was significantly decreased in *Scn1a*^{+/-} mice, with an accompanying increase in individual spike width. (2) To monitor the activity of SNr PV⁺ neurons *in vivo*, we recorded their population activity via fiber photometry using a virally expressed calcium indicator (GCaMP8s), in addition to simultaneous electrocorticography recordings to identify seizure onset and offset times. Similar to our findings at the single-cell level, the interictal activity of SNr PV⁺ neurons was significantly different between genotypes: calcium transients in *Scn1a*^{+/-} mice were less frequent, lower amplitude, and wider. (3) Surprisingly, we observed a profound *decrease* in SNr

PV+ activity during hyperthermia-evoked seizures, contrary to published literature in other models. However, when we used the chemoconvulsant pentylenetetrazol (PTZ) to trigger seizures in *Scn1a*^{+/-} mice, we observed *increased* SNr PV+ activity. Our findings reveal impairments in SNr PV+ neurons that may contribute to the motor phenotype of DS. The surprising finding that SNr PV+ cells are suppressed during hyperthermia-evoked seizures may be due to a combination of elevated temperature and *Scn1a* haploinsufficiency, possibly providing mechanistic insight into febrile seizures. Future experiments manipulating these neuronal populations during seizures will test the role of this circuit in seizure initiation and termination to assess its potential as a therapeutic target.

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Poster

PSTR262: *In Vivo* and Behavioral Analysis of Epilepsy

Location: SDCC Halls B-H

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Program #/Poster #: PSTR262.15/C27

Topic: B.08. Epilepsy

Support: NIH/NINDS grant R01-NS09697-09
NIH/NINDS grant R21-NS138525-01
Savoy Foundation of Canada Postdoctoral Fellowship

Title: High-throughput screening (HTS) utilizing a zebrafish model for *stxbp1* disorders

Authors: *P. WHYTE-FAGUNDES¹, S. C. BARABAN²;

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Abstract: Syntaxin-binding protein 1 (STXBP1) is a core component of the highly conserved SNARE protein complex that governs exocytosis. It plays a critical role in neurotransmitter release, synaptic plasticity, cognitive function, and neurodevelopment. Heterozygous *de novo* mutations in *STXBP1* lead to severe neurodevelopmental disorders, including epilepsy, global developmental delay, and intellectual disability. Recent longitudinal clinical data highlight gross motor delays as a prominent comorbidity, affecting nearly half of *STXBP1* patients (Xian et al., Brain 2023). Unfortunately, current treatments are limited to supportive care, including antiseizure medications, as well as physical, occupational, and speech therapies, underscoring the urgent need for disease-modifying interventions. To address this problem, we used CRISPR technology to generate a preclinical zebrafish model (*stxbp1a*^{-/-}) that recapitulates the movement disorder observed in this patient population (Grone et al., PLoS One 2017). Mutant larvae exhibit a 73% reduction in spontaneous swim activity compared to wild-type siblings (282 ± 136 mm vs. 1029 ± 625 mm, $p < 0.001$), closely resembling the hypomotility induced by exposure to the anesthetic tetracaine in WT larvae (301 ± 151 mm). We first evaluated the clinical candidate 4-

phenylbutyrate (4-PBA), currently in compassionate-use trials, and 18 AI-identified analogs, but observed no improvement in larval movement after a chronic 48-hour exposure ($p=0.87$). To identify potential lead compounds that reverse/rescue the motor disorder phenotype, we then performed phenotypic high-throughput screening (HTS) using FDA-approved ($n = 484$), GPCR ($n = 253$), MAP kinase ($n = 40$), PI3K-AT ($n = 117$) and Natural Products ($n = 221$) libraries. First-pass HTS using DanioVision tracking identified 17 lead candidates that significantly increased swim movement in mutant larvae. Ongoing second-stage validation, incorporating electrophysiology and high-resolution tracking with the Ramona Optics MCAM system, leverages advanced kinematic profiling to refine lead candidates (Whyte-Fagundes et al. Comm. Biol. 2025). Our results suggest that 4-PBA is unlikely to be effective for treating motor dysfunctions associated with *STXBPI* disorder, but novel candidates emerging from our HTS pipeline may offer therapeutic promise. This study highlights the utility of zebrafish as a high-throughput, *in vivo* platform for drug discovery targeting genetically defined neurodevelopmental disorders.

Disclosures: P. Whyte-Fagundes: None. S.C. Baraban: None.

Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.01/C28

Topic: B.09. Glial Mechanisms

Support: NIH R01NS115707
NIH R01NS105691
NSF CAREER CBET 1943906
Pitt Momentum
NIH R01NS129632
NIH R01Actf NS115707

Title: Intracortical Microstimulation Modulates Calcium Dynamics Differently in Oligodendrocyte Somas and Processes

Authors: *C. GARCIA PADILLA¹, T. D. KOZAI²;
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Abstract: Demyelination and oligodendrocyte (OL) dysfunction are features of many neurodegenerative diseases, yet mechanisms regulating OL function are poorly understood. OLs are the brain's primary myelinating cells and maintain neuronal health via structural and metabolic support. Disruption of OL function impairs neural communication and network integrity. Calcium (Ca^{2+}) signaling has emerged as a surrogate measure of intracellular activity in OLs, with Ca^{2+} transients correlating with processes like myelin formation. While many pharmacological and genetic strategies aim to modulate OL Ca^{2+} transients, few probe the effects

of direct intracortical microstimulation (ICMS) on OL Ca²⁺ dynamics.

We tested whether ICMS modulates OL Ca²⁺ signaling in vivo using PLPCreERT::Salsa6f mice, which express red fluorescence for OL morphology and a green cytosolic Ca²⁺ indicator. OL somas and processes were imaged in Layer 1, where their horizontal orientation supports high-resolution structural and functional analysis. To increase local cortical activity, 100 Hz or theta-burst stimulation (TBS) ICMS was delivered to Layer 2/3, based on evidence that neuronal activity influences OL function and myelination. Ca²⁺-linked fluorescence was recorded pre-, during-, and post- ICMS to quantify event amplitude, duration, and rates of rise and decay. Peak amplitude was significantly lower in OL somas ($0.132 \pm 0.016 \Delta F$) than processes ($0.165 \pm 0.031 \Delta F$) during the 100 Hz pre-ICMS period ($p = 0.007$), but not during ($p = 0.317$) or after ICMS ($p = 0.211$), suggesting 100 Hz ICMS may entrain the intensity of OL Ca²⁺ responses across compartments. Processes' decay time following 100 Hz ICMS increased from 0.698 ± 1.356 to 1.264 ± 1.136 sec ($p = 0.045$), an effect not observed with TBS ($p = 0.959$). This slower decay may reflect altered Ca²⁺ dynamics or sustained intracellular signaling after high-frequency stimulation. Additionally, both 100 Hz ($p = 0.014$) and TBS ($p = 0.025$) significantly increased the rate of rise in soma Ca²⁺ events post-ICMS, suggesting ICMS has a lasting effect on soma Ca²⁺ dynamics. Together, these findings indicate that ICMS paradigms can differentially shape OL Ca²⁺ dynamics across subcellular compartments, with 100 Hz ICMS promoting more prolonged responses.

These results suggest ICMS alters temporal features of OL Ca²⁺-linked fluorescence, which differ in somas and processes. While the mechanism (whether direct or indirect via neuronal or glial intermediates) remains unresolved, our findings identify a stimulation-responsive shift in OL Ca²⁺ dynamics potentially relevant to activity modulation to promote myelination.

Disclosures: C. Garcia Padilla: None. T.D. Kozai: None.

Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

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Program #/Poster #: PSTR263.02/C29

Topic: B.09. Glial Mechanisms

Title: Structural and Pharmacological Basis of Oxysterol Modulation of GPR17 in Human Oligodendrocytes

Authors: *J. C. SILBEREIS;
Biogen, Cambridge, MA

Abstract: GPR17 is a class A orphan GPCR that is predominantly expressed in the central nervous system where it plays an important role in oligodendrocyte development and myelination. Inhibiting GPR17 is a novel therapeutic approach for neurological indications featuring myelin damage such as multiple sclerosis and brain injury. Discovery of GPR17 inhibitors is challenged by both its orphan status and a rudimentary understanding of the

structural basis of GPR17 agonism and antagonism. A potential step towards GPR17 deorphanization was made by the recent discovery of select oxysterols as potential biological ligands of GPR17. To understand the structural pharmacology of GPR17 synthetic ligands and oxysterols, we solved novel cryo-EM structures of GPR17 with bound orthosteric antagonist or agonist/G protein heterotrimer. Modeling using these structural data predicted allosteric GPR17 binding sites at the cell membrane receptor interface, which was confirmed biochemically via competition binding assays of 24S-hydroxycholesterol with a cholesterol photoprobe. In vitro pharmacological characterization of oxysterols versus an orthosteric ligand in a novel human oligodendrocyte progenitor cell assay supported allosteric binding. Measurements of 24S-hydroxycholesterol taken from post-mortem human multiple sclerosis and control brain tissue suggest modulation of the GPR17 occurs at biologically relevant concentrations. Together these findings provide a structural basis for oxysterols as natural ligands of GPR17 and extend the pharmacological evidence to the native human cell type. Therefore, therapeutically targeting allosteric sterol interactions offers an alternative but as yet untested approach to GPR17 antagonism.

Disclosures: **J.C. Silbereis:** A. Employment/Salary (full or part-time): Employment, Biogen, Inc. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Biogen, Inc.

Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.03/C30

Topic: B.09. Glial Mechanisms

Title: Novel Insights at the Intersection of Copper Biology and Oligodendrocyte Development

Authors: ***J. WONG**¹, J. B. GRINSPAN³, K. L. JORDAN-SCIUTTO²;

²Dept Oral Med., ¹Univ. of Pennsylvania, Philadelphia, PA; ³Dept. of Neurol., Children's Hosp Philadelphia, Philadelphia, PA

Abstract: In the central nervous system, oligodendrocytes (Ols) synthesize myelin, a lipid-enriched membrane crucial for facilitating transmission of action potentials and trophic support to axons. Ols are generated from Ol progenitor cells (OPCs) which go through a well-documented maturation characterized by the extension of cell processes and the synthesis of critical myelin proteins such as myelin basic protein (MBP) which are then incorporated into the myelin membrane. The loss of myelin contributes to motor and neurocognitive dysfunction in demyelinating diseases such as multiple sclerosis (MS). Diseases involving an excess or deficiency in cytoplasmic copper (Wilson's and Menkes disease, respectively) also show impaired myelination in the CNS, suggesting oligodendrocytes may be keenly sensitive to copper deprivation. Consistent with this, feeding rodents the copper chelator cuprizone causes demyelination, primarily in the corpus callosum, and is commonly used as a model in

demyelination/remyelination studies. However, the exact mechanism by which cuprizone induces demyelination is still not clear. We treated cells in culture with cuprizone at 3 specific stages of differentiation: as OL progenitor cells (OPCs), differentiating OLs (DOLs), and mature OLs (MOLs). We found that only cultures allowed to differentiate for 3 days before being treated with cuprizone (MOLs) showed significant levels of oligodendrocyte death, beginning at approximately 100 μ M. We also found that MOLs contain more copper compared to OPCs and DOLs via inductively coupled plasma mass spectrometry (ICP-MS) analysis, and that MOLs express significantly more CTR1, the high affinity copper uptake protein and primary copper importer into cells. Additionally, MOLs treated with a nonlethal dose of cuprizone (50 μ M) exhibited significantly reduced myelin basic protein (MBP) stability compared to untreated MOLs. These data indicate that mature OLs are uniquely sensitive to cuprizone, suggesting that copper metabolism may be specifically critical to mature OLs and their stage-specific proteins such as MBP. We also found that activation of the endolysosomal cation channel TRPML 1 via Mucolipin synthetic agonist 1 (MLSA1) protected against the copper-chelating deficits in oligodendrocytes, possibly implicating lysosomal dysfunction in the actions of cuprizone. The results of this study will provide valuable insights into the effects of copper storage and transport on oligodendrocytes maturation and function.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

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Program #/Poster #: PSTR263.04/C31

Topic: B.09. Glial Mechanisms

Support: SNRI Pilot Pre-Clinical Imaging Grant

Title: A gut-derived metabolite, indoxyl sulfate, disrupts intact myelin structure and alters myelin macromolecule composition

Authors: *T. J. CONNERLY;

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Abstract: The field of gut-brain-axis research emerged from scientific discoveries demonstrating the microbiome's truly symbiotic relationship with human hosts. Microbes metabolize dietary and host-derived compounds into metabolites that can circulate throughout the body to directly or indirectly enact changes on the host. This surprising supply of bioactive molecules, termed gut-derived metabolites, are critical for the health of local systems, including the gut and epithelium, the two places we think of having the most bacteria. However, it is now increasingly recognized that gut metabolites can have direct targets in a thought-to-be isolated area of the body, the brain. A recent study within our lab demonstrated a gut-derived metabolite impairs oligodendrocyte maturation, reduces axonal myelination, and subsequently impairs neuron

function. This connection defining a gut-derived metabolite as a glial mediator establishes a precedent for investigating other bacterially derived products. Through a high-throughput screen of over 200 metabolites using a neuronal-glial co-culture, our lab identified indoxyl sulfate, a gut-derived metabolite, as a potent disruptor of oligodendrocyte maturation and myelination. Classified as a uremic toxin, indoxyl sulfate is usually eliminated by the kidneys. However, diseases such as multiple sclerosis, anxiety, dementia, chronic kidney disease with high cognitive impairment are associated with dramatic increases in serum indoxyl sulfate. This research will comprehensively investigate how indoxyl sulfate, a disease associated metabolite, interferes with oligodendrocyte development and function. In addition to our co-culture model for mechanistic investigation, we will utilize an in-vivo monocolonized mouse model selectively overexpressing (n=10) or ablating (n=10) production of the enzymes necessary to convert dietary tryptophan to indoxyl sulfate. Our work utilizes advanced magnetic resonance imaging techniques and macromolecule analysis to identify intact structural and macromolecule disturbances in myelin formation. Our early data indicates oligodendrocytes are myelinating, but with decreased levels of myelin associated proteins when assessed with western blotting. Follow-up experiments utilize transmission electron microscopy and full macromolecule analysis with western blots and LC-MS will further determine the impact of indoxyl sulfate exposure on myelination. By illuminating how a single gut-derived molecule impairs central nervous system integrity, this work has the potential to redefine therapeutic strategies for a wide range of central nervous system disease.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.05/C32

Topic: B.09. Glial Mechanisms

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Dr. Miriam and Sheldon G. Adelson Medical Research Foundation

Title: Proximity labeling and genome editing reveal the paranodal proteome in oligodendrocytes

Authors: *X. DING¹, J. CURTIS¹, Y. XING¹, Y. OGAWA², E. PELES³, M. N. RASBAND¹;
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Abstract: Proper central nervous system (CNS) function requires precise neuron-glia interactions. A key site of such interaction is the paranodal junction (PNJ), which is formed between myelinating oligodendrocytes (OLs) and axons. The PNJ flanks the nodes of Ranvier, anchors the myelin sheath to the axon, facilitates nodal Nav channel clustering, mediates

bidirectional communication between OLs and axons, and is essential for rapid action potential propagation. Notably, PNJ disruption is a pathological hallmark of various neurological disorders, including multiple sclerosis (MS). Despite its fundamental role as the largest cell-cell junction in vertebrates, the molecular organization of the PNJ remains poorly defined. To date, only a limited number of its components have been identified in OLs, including the adhesion molecule NF155, the cytoskeletal protein β II spectrin, and the scaffolding protein Ankyrin-G (AnkG). This critical knowledge gap reflects the technical challenges of protein isolation and identification posed by the lipid-rich, detergent-insoluble nature of the PNJ. Furthermore, subsequent functional studies in OLs typically require the generation of genetically modified animals for each candidate, a process that is time-consuming and labor-intensive. To overcome these obstacles, we employed a combination of mouse genetics, proximity proteomics, and AAV-mediated, cell-type-specific CRISPR/Cas9 genome editing, enabling rapid identification, validation, and functional dissection of novel PNJ components in OLs. We generated *Ank3^{LSL-TurboID}* mice, where the biotin ligase TurboID is conditionally fused to the C-terminus of endogenous AnkG following Cre-mediated recombination in OLs. This enables covalent labeling of paranodal proteins, followed by purification using streptavidin beads and identification via mass spectrometry. Our study uncovered a previously uncharacterized proteomic landscape at the PNJ, identifying novel PNJ-enriched candidates with potential roles in axoglial interactions. We validated selected candidates using AAV-delivered CRISPR genome editing to tag endogenous proteins, bypassing limitations associated with commercial antibody availability and specificity. In conclusion, our discovery platform provides a powerful and scalable strategy for accelerating the identification of PNJ components and advancing our understanding of axoglial interactions in both health and disease.

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Topic: B.09. Glial Mechanisms

Support: NIH NIA Intramural Research Program

Title: Deficiency of endocannabinoid synthase DAGLB in oligodendrocyte contributes to increased motor activity

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Abstract: 2-arachidonoyl-glycerol (2-AG), the most abundant endocannabinoid (eCB) in the brain, plays an important role in the nervous system. Deficiency of 2-AG has been closely

associated with a range of neurological symptoms, including memory impairment, motor coordination deficits and neuroinflammation. Daglb, as well as Dagla, is a key synthase of 2-AG. In contrast to the more abundant Dagla, our understanding of Daglb remains limited. Our previous research highlighted the importance of Daglb-mediated 2-AG biosynthesis in nigral dopaminergic neurons (DANs) in regulating dopamine release and motor skill learning. In addition to neuron, Daglb is also highly enriched in glia, particularly in oligodendrocytes. However, the role of Daglb in oligodendrocyte is poorly understood. Therefore, we generated mice with a conditional knockout (cKO) of Daglb in oligodendrocyte-lineage cells. In this study, we found Daglb cKO in oligodendrocytes increased locomotor travel distance and speed in the open field test, without affecting motor skill learning. Notably, administration of the 2-AG degradation inhibitor JZL-184 attenuated these behavioral alterations, indicating that the observed motor phenotype was attributed to the deficiency in 2-AG production. Consistently, gait analysis revealed a significant reduction in paw pressure of cKO mice compared to wild-type mice, while stride length, stride width, and other gait parameters showed no significant differences. Interestingly, further pathological analysis revealed that an enlarged myelin sheath lumen diameter of sciatic nerve in Daglb cKO mice. To probe downstream gene expression changes resulted from Daglb deletion, we performed RNAseq in primary cultures of oligodendrocyte-lineage cells. We found that loss of Daglb, without affecting Dagla expression, was associated with alterations in cell cycle regulation and myelination-related signaling pathways. Together, our results indicate that the loss of Daglb in oligodendrocytes may lead to alterations in myelin structure, potentially in motor neurons, which in turn could affect motor regulation in mice. By selectively deleting Daglb in oligodendrocyte-lineage cells, our study further investigates the role of Daglb, as a key enzyme in 2-AG synthesis, on motor behavior and neural function. These findings provide novel insights into the contribution of oligodendrocyte-derived eCB signaling to the regulation of the nervous system and suggest potential therapeutic targets for related neurological disorders.

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Poster

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Topic: B.09. Glial Mechanisms

Support: ERC Advanced Grant 694798
SNSF Grant 310030-182496
SNSF Ambizione Grant 208875

Title: Linking oligodendrocyte homeostasis with metabolic imbalances. Hints from Tmem117, a novel regulator of NCX activity.

Authors: M. ALAPPAT¹, M. MAZURKIEWICZ¹, A. PICARD², F. PRISCO³, A. KIPAR³, H. U. ZEILHOFER^{1,4}, B. THORENS², *S. GASPARI¹;

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Abstract: While the impact of metabolic imbalances on myelin content and integrity has been assessed in numerous studies, the reciprocal influence of central nervous system (CNS) myelin homeostasis on systemic metabolic health remains largely unexplored. In this study, we investigate the function of Tmem117, a previously uncharacterized protein, and demonstrate that transient disruptions in myelin homeostasis induce sex-specific effects on the counterregulatory response to hypoglycemia, glucose tolerance, and hepatic lipid accumulation in mice. We show that depletion of Tmem117 in mature oligodendrocytes leads to increased endoplasmic reticulum (ER) stress, dysregulation of key molecular pathways, and subsequent demyelination. Moreover, guided by evolutionary insights and in vitro analyses, we provide initial evidence suggesting that Tmem117 acts as a stabilizer of the sodium-calcium exchanger NCX1. Together, our findings reveal a critical role for CNS myelin integrity in maintaining whole-body metabolic homeostasis and identify Tmem117 as a potential cell-specific modulator of NCX1 activity.

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Poster

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Topic: B.09. Glial Mechanisms

Support: Swiss National Science Foundation grant 310030-182496
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Title: Clearly sweet and painful: Investigating the effect of western diet on myelin homeostasis

Authors: *M. ALAPPAT¹, M. MAZURKIEWICZ¹, F. PRISCO², A. KIPAR², B. THORENS³, H. U. ZEILHOFER^{1,4}, S. GASPARI¹;

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Abstract: Chronic pain is a debilitating disease affecting approximately 20% of the adult population. Its treatment remains highly challenging with efficacy and side effects varying substantially among individuals. A growing body of evidence suggests that lifestyle changes, particularly dietary habits and nutrient intake, strongly influence disease outcome, though the

mechanisms remain poorly understood. The Western diet (WD) has been implicated in the development of chronic pain, as well as in demyelinating diseases like multiple sclerosis, where it is thought to contribute through systemic inflammation and oxidative stress caused by WD-related metabolic imbalances. Unfortunately, there is still a knowledge gap on the underlying mechanisms that hinder the development of defined nutritional guidelines for disease management. Oligodendrocytes (OLs), the myelin-producing cells of the central nervous system (CNS), are highly responsive to metabolic imbalances and inflammation, and particularly vulnerable to oxidative stress. At the same time, their sole depletion triggers pain-like responses in mice. To investigate the complex interplay of WD, CNS myelination, and pain we utilize transgenic mouse models of both sexes and behavioural studies. Genetically-induced demyelination is combined with nociceptive testing to identify the affected modalities including thermal hyperalgesia, cold, and mechanical allodynia. Furthermore, a triple transgenic mouse model (PDGFR α -creERT^{tg/+}; Ai14^{fl/fl}; CNP-mEGFP^{tg/tg}) for fluorescent labelling of OLs/myelin and OPCs is employed to explore the effect of WD supplementation on CNS myelin morphology and OL dynamics. 12 weeks after WD onset, the CNS of mice are dissected, cleared with the SHIELD clearing method and subjected to imaging using the light-sheet mesoSPIM microscope. Our results so far indicate that the SHIELD transformation strategy for whole-CNS tissue clearing is suitable for our purposes. This protocol allows for the protection of the physiochemical properties and ensures proper fluorescent preservation of both brain and spinal cord tissue. Therefore, enabling us to image myelin dynamics throughout the CNS. This research seeks to advance our understanding of pDN and contribute valuable insights that may inform the development of novel and targeted treatment approaches.

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Poster

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Topic: B.09. Glial Mechanisms

Support: Swiss National Science Foundation Ambizione Grant 208875

Title: Identification of oligodendrocyte transcriptional signatures in Western diet-induced pain

Authors: *M. A. MAZURKIEWICZ¹, M. ALAPPAT¹, H. U. ZEILHOFER^{1,2}, S. GASPARI¹;
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Abstract: Chronic pain, whether considered a disease *per se* or a symptom of an existing disease, affects approximately 20% of the adult population. More than 1/3 of diabetic patients and obese individuals experience pain, while impaired glucose tolerance in non-diabetic subjects

has been correlated with an increase in the prevalence for neuropathic pain development. In line with these observations, adoption of a Western Diet (WD) triggers pain-like responses in preclinical rodent models. So does the sole depletion of oligodendrocytes (OLs), the myelin producing cells of the central nervous system (CNS). Furthermore, OLs are highly responsive to metabolic imbalances and inflammation, with particular vulnerability to oxidative stress, and as such, a promising candidate contributor to diet-induced myelin defects. In this project WD supplementation is used to induce metabolic imbalances in a transgenic mouse model, contributing to the development of mechanical hypersensitivity. Effects of the WD are investigated on the transcriptome level by performing single nucleus RNA sequencing (snRNAseq) on the whole spinal cord, with analysis focusing on oligodendrocyte lineage cells. Mice kept on a WD for 3 months separate into two distinct populations based on the mechanical sensitivity threshold: those that behave like regular diet control mice, displaying no sign of mechanical hypersensitivity and those that develop mechanical hypersensitivity that persists over several weeks. This division is present in both female and male mice. SnRNAseq results suggest that oligodendrocyte lineage cells comprise more than 47% of the cells in the mouse spinal cord and are highly affected by WD in both sexes. WD supplementation contributes to diet-related downregulation of transcripts associated with oxidative phosphorylation. Furthermore, we detect pain-related downregulation of core myelin genes (*Mbp*, *Mag*, *Mog*, *Cnp*, *Plp1*) and genes involved in mRNA processing. OLs are crucial for maintaining homeostasis of the CNS and normal pain perception. Thus, disruption of OLs and CNS myelin could be the missing link connecting metabolic imbalances with the development of pain.

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Poster

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Topic: B.09. Glial Mechanisms

Support: Shriners Hospitals for Children 85410-NCA-24
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Title: Targeting Poly(ADP-ribose) Glycohydrolase (PARG) to Promote Remyelination in Pediatric Multiple Sclerosis Models

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Abstract: Background: Multiple sclerosis (MS) is a chronic demyelinating disorder of the central nervous system (CNS), characterized by immune-mediated myelin loss and

neurodegeneration. Pediatric-onset MS accounts for approximately 5% of all MS cases and is associated with higher relapse rates and an increased risk of long-term disability. Currently, no therapies are approved that specifically promote remyelination in the pediatric population. Failure of remyelination is a major contributor to irreversible neurological decline. Poly(ADP-ribosyl)ation (PARylation), a reversible post-translational modification regulated by PAR polymerases (PARPs) and poly(ADP-ribose) glycohydrolase (PARG), has recently emerged as a key regulator of oligodendrocyte lineage progression and CNS myelin repair. **Objective:** To define the role of PARG in oligodendrocyte differentiation and remyelination, and to assess its therapeutic potential in a juvenile mouse model of demyelination relevant to pediatric MS. **Methods:** We employed a cuprizone-induced demyelination model in juvenile mice, combined with a tamoxifen-inducible Cre-loxP system to conditionally delete Parg in oligodendrocyte lineage cells. Mice were evaluated during the demyelination and remyelination phases for myelin integrity (MBP immunostaining), oligodendroglial differentiation (PDGFR α , Olig2, CC1), and behavioral outcomes. In parallel, we administered a brain-penetrant PARG inhibitor to test its translational potential in promoting myelin repair. **Results:** Parg deletion in oligodendrocytes had a dose-dependent effect on remyelination. Heterozygous Parg deletion enhanced oligodendrocyte maturation and significantly increased myelin restoration during recovery, as shown by elevated CC1⁺ cells and MBP levels. In contrast, homozygous Parg deletion impaired remyelination, suggesting that balanced PARylation is required for optimal oligodendrocyte function. Pharmacological inhibition of PARG promoted oligodendrocyte differentiation and remyelination without detectable adverse effects on astrocytic or microglial responses, supporting the feasibility of targeting PARG in vivo. **Conclusion:** These findings identify PARG as a critical regulator of myelin regeneration in the juvenile CNS. Both partial genetic deletion and pharmacological inhibition of PARG enhance remyelination, highlighting its potential as a therapeutic target for pediatric MS. Modulating PARylation dynamics through selective PARG inhibition may represent a promising strategy to restore myelin integrity and improve long-term neurological outcomes in children with demyelinating diseases.

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Poster

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Title: Deep Learning-based 3D Myelin Quantification Reveals Environmental-induced Neuroplasticity in Rat Prefrontal Subregions

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Abstract: Early-life social experience shapes myelination in the prefrontal cortex (PFC), yet quantitative evidence for fiber-level plasticity in discrete PFC subregions remains limited. We therefore created a topology-aware, three-dimensional deep-learning pipeline that segments individual myelin fibers in reflectance confocal microscopic z-stacks and returns direct measures of fiber density. Eight Long Evans rats were weaned at post-natal day 28 and housed either singly or in groups (n = 4 per condition) for four weeks. Coronal 40 μm sections including anterior cingulate, prelimbic, infralimbic, and orbital frontal cortices were processed with Nissl and Gallyas silver stains and imaged at $0.17 \mu\text{m} \times 0.17 \mu\text{m} \times 0.4 \mu\text{m}$ voxel resolution. Human-annotated volumes trained a modified U-Net equipped with topology-preserving loss functions that promote continuity of curvilinear structures. To provide an interim biological benchmark and validate the deep learning predictions, we extracted mean gray values from identical image stacks. A one-tailed independent-samples t-test indicated lower myelin signal in Infralimbic cortex of singly housed rats (M = 60.38, SD = 10.03) than in group-housed littermates (M = 78.51, SD = 12.11), $t(6) = -2.31$, $p = 0.031$, 90 % CI [-33.39, -2.87], Cohen's $d = 1.63$, supporting the deep learning segmentation measurements. These preliminary data are consistent with the hypothesis that reduced social interaction diminishes myelin content in PFC subregions involved in affective and social decision making. By delivering a scalable three-dimensional quantification framework and an open dataset, our study will enable precise mapping of environmentally driven myelin plasticity across PFC subregions. It will facilitate multimodal investigations of cortical maturation and pathology, such as coregistrative comparisons to MRI myelin measures.

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Support: NIH Grant R03 AG072218
NIH Grant R01 NS129632

Title: Hippocampal glutamate content and excitatory synaptic density in Fus^{OL}cKO hypermyelinating mice

Authors: *A. L. SCIUTTO¹, E. ROBBINS², T.-H. TUNG¹, S. WELLMAN³, T. D. KOZAI², T. CUI², F. CAMBI¹;

¹Neurol., ²Bioengineering, Univ. of Pittsburgh, Pittsburgh, PA; ³Bioengineering, Columbia Univ., New York, NY

Abstract: Oligodendrocytes and myelin play an important role in maintaining the integrity of neuronal circuits in addition to facilitating fast saltatory conduction. To study the impact of oligodendrocytes and myelin on neuronal circuits, our lab has generated a hypermyelinating mouse by conditionally knocking out Fused in Sarcoma (Fus) in oligodendrocytes (*Fus^{OL}cKO*). Electrophysiological recordings from microelectrodes implanted through the visual cortex to the CA1 subregion of the hippocampus show sustained neuronal activity in CA1 of *Fus^{OL}cKO* vs. wild type (WT) mice. While these data suggest a critical role of oligodendrocytes in maintaining neuronal signaling in the hippocampus, the biochemical basis of this firing remains unknown. To address this, custom glutamate-sensing microelectrodes were acutely implanted through the visual cortex and the underlying hippocampus. Glutamate content was measured at three locations, putative dorsal (d)CA1, medial (m)CA1, and ventral (v)CA1. Glutamate concentration in (d)CA1 was higher in the *Fus^{OL}cKO* vs. WT mice which is consistent with the enhanced neural activity in CA1 ($p=0.0317$). To contextualize this finding, the cellular environment was assessed via immunofluorescence. The probe tract was labeled with an anti-mouse IgG secondary antibody. No differences in activated astrocytes (GFAP+) and microglial density (Iba1+) were observed within 0-150 and 150-300 microns from the probe tract. Myelin density adjacent to the probe tract was assessed via immunofluorescence with markers of compact myelin (MOG and MBP) and was also not significantly affected by probe insertion. Given that these factors were not altered by probe insertion, we hypothesized that greater excitatory synaptic density in (d)CA1 of *Fus^{OL}cKO* mice underlies the greater glutamate concentration detected by the probe in (d)CA1 of *Fus^{OL}cKO* mice. To quantify excitatory synaptic density in recorded regions, excitatory presynaptic terminals were identified with VGLUT1 and the postsynaptic density with PSD95. The number of appositions of these two markers within 200nm of one another was the readout for excitatory synapses. Preliminary findings suggest that excitatory synaptic density in (d)CA1 of *Fus^{OL}cKO* mice is not significantly different than that of WT mice. ($p=0.4866$). To assess whether a decrease in inhibitory tone underlies the higher glutamate and firing rate, studies are underway to quantify inhibitory synapses. Overall, these data highlight the importance of oligodendrocytes and myelin in maintaining excitatory neuronal signaling in the hippocampus.

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Poster

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Topic: B.09. Glial Mechanisms

Title: In vitro model to study demyelinating disease using human OPC- and oligodendrocyte-like cells generated by deterministic cell programming

Authors: A. VETLEANU, M. HERRERA VAQUERO, T. TATAR OZKAN, S. HUSSAIN, L. FOULSER, S. MILDE, C. ARUTHAN, J. HUNDLING, H. GARNETT, N. JAMES, B. NEWMAN, V. YIANNI, ***I. FERREIRA**, K. FIRTH, G. MASTROGIOVANNI, T. OOSTERVEEN, W. BERNARD;
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Abstract: Oligodendrocyte precursor cells (OPCs) are proliferative progenitor cells that give rise to myelinating oligodendrocytes (OLs) during CNS development and in response to injury during adulthood. The primary function of OLs is the ensheathment of axons with myelin to enable action potential conduction. Impaired OPC differentiation, OL loss, and disrupted myelination are pathological features of demyelinating diseases, such as multiple sclerosis (MS). The development of therapies that promote remyelination is hampered by the poor translatability of animal models and a lack of robust, human-relevant in vitro models. Although human induced pluripotent stem cells (hiPSC) offer a promising tool to study oligodendroglial cells, widespread adoption is hindered by lengthy, complex protocols with poor reproducibility. To address these limitations, we used opti-ox™, a deterministic inducible overexpression technology, to deterministically program oligodendroglial cells (ioOligodendrocyte-like cells) from hiPSC. By day 1 post-thaw, the cells exhibit an OPC-like morphology and express the oligodendroglial marker O4. By day 8, cells have acquired an OL-like morphology and upregulate maturation markers, including MBP, seen by bulk RNA and scRNAseq. Equivalent transcriptomic profiles across three independent batches show high lot-to-lot consistency. MBP is a key OL marker, essential for myelin sheath formation, and enhancing its expression will improve the relevance of this model for myelination research. Interestingly, ioOligodendrocyte-like cells show increased MBP expression when in co-culture with hiPSC-derived ioGlutamatergic Neurons, enabling axon-glia interaction studies. As monocultures of OLCs show relative less MBP expression, we further optimised the media composition to significantly increase the proportion of MBP-positive cells. In multiple sclerosis (MS), endogenous OPCs fail to efficiently differentiate into OLs and remyelinate lesions. To model OPC biology in vitro and develop efficient remyelination-promoting therapies, we generated iPSC-derived OPC-like cells (ioOPC-like cells). These cells have the capacity to proliferate for up to 14 days and express OPC markers including PDGFRA. Upon switching to maturation medium, the cells downregulate PDGFRA and upregulate MBP and other mature markers, and acquire an oligodendrocyte-like cell morphology. Our OPC-like cells and oligodendrocyte-like cells provide human, relevant, consistent in vitro models to study the role of these cells in disease mechanisms, and support the development of novel therapeutics for neurodegenerative and demyelinating diseases.

Disclosures: **A. Veteleanu:** A. Employment/Salary (full or part-time): bit.bio. **M. Herrera Vaquero:** A. Employment/Salary (full or part-time): bit.bio. **T. Tatar Ozkan:** A. Employment/Salary (full or part-time): bit.bio. **S. Hussain:** A. Employment/Salary (full or part-time): bit.bio. **L. Foulser:** A. Employment/Salary (full or part-time): bit.bio. **S. Milde:** A. Employment/Salary (full or part-time): bit.bio. **C. Aruthan:** A. Employment/Salary (full or part-time): bit.bio. **J. Hundling:** A. Employment/Salary (full or part-time): bit.bio. **H. Garnett:** A. Employment/Salary (full or part-time): bit.bio. **N. James:** A. Employment/Salary (full or part-

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Poster

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NIH Grant R01 NS129632

Title: Fus depleted oligodendrocytes ameliorate AD pathology and reduce oxidative stress through Enhanced Energy Metabolism and Antioxidant Defense

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Abstract: Oligodendrocytes (OL) and myelin abnormalities have emerged as important contributors to the pathogenesis of Alzheimer's Disease (AD). OL maintain neuronal health through myelin protective function and by direct neurotrophic and bioenergetic support. To gain insight on how OL and myelin ameliorate neuronal deficits associated with AD, we have generated a new mouse model referred to as AD/cKO by crossing the App^{NL-G-F} mouse (referred to as AD) with the *Fus*^{OL}cKO, which has thicker myelin and greater cholesterol biosynthesis in white matter tracts. In previous studies, we showed that working memory is preserved, plaque burden is decreased, microglia density is reduced in the hippocampus of the AD/cKO vs. AD at late stages of disease (16 months). To elucidate mechanisms that lead to improved memory and reduced pathology, we have examined the changes in microglia states and synaptic integrity during disease progression. The percent of A β specific (TREM2⁺/Iba1⁺) microglia clustered around plaques of similar size in the CA1 subregion was higher in the ADcKO vs. AD (64.91 $\pm$ 3.70 % vs. 46.69 $\pm$ 3.11 %, p = 0.0003), consistent with a shift to a more phagocytic and neuroprotective state. Synaptophysin loss at plaques of similar size was reduced in the hippocampus of aged AD/cKO vs. AD (13.04 $\pm$ 2.33 % vs. 19.42 $\pm$ 3.84 %, p = 0.0295), indicating preservation of presynaptic structures. Since myelin density was similar in the AD/cKO vs. AD hippocampus, we reasoned that changes other than myelin contribute to Fus depleted OL beneficial effects. Single cell transcriptomics of AD/cKO hippocampal OL at early stages of amyloid accumulation showed greater expression of energy metabolism and antioxidant genes

compared to AD, suggesting greater bioenergetics support to neurons. Indeed, oxidative stress associated with AD assessed by measuring lipid peroxidation by 4-HNE in CA1 subregion was reduced in the aged AD/cKO hippocampus vs. AD ($18.73 \pm 2.24\%$ vs. $41.61 \pm 10.68\%$, $p = 0.0001$). In contrast, hypoxic damage measured by pyronin binding, an early event in AD pathology, was not statistically different in AD/cKO vs AD at early stages of disease ($0.00030 \pm 0.00013\%$ vs. $0.00063 \pm 0.00041\%$, $p = 0.17$). Studies are underway to characterize DNA oxidative damage, identify which cells (neurons, OL) have reduced oxidative damage and characterize the role of astrocyte activation. Collectively, the data suggest that enhancing OL energy metabolism leads to attenuation of AD progression likely through reduced oxidative damage, improved neuronal health and synaptic function and changes in microglia responses.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.15/C42

Topic: B.09. Glial Mechanisms

Support: 23K14690
JPMJMS2024

Title: Elucidating the Pathophysiology of Vascular Dementia with a Focus on Cerebral White Matter Vascular Fibrosis

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Abstract: [Background]

Subcortical ischemic vascular dementia (SIVD) is the most common subtype of vascular dementia and is defined by subcortical white matter lesions in humans. These lesions exhibit oligodendrocyte loss, demyelination, blood-brain barrier (BBB) disruption, and axonal damage. Fibroblasts are key components of fibrous connective tissues distributed throughout various organs and contribute to extracellular matrix (ECM) production during tissue development, maintenance, damage and repair. Recent studies have shown that pericytes and perivascular fibroblasts coordinate vascular remodeling and scar formation after ischemic stroke. However, the contribution of fibroblasts to the pathogenesis of SIVD remains unclear.

[Aim]

This study aimed to investigate the origin and role of fibroblasts in the pathogenesis of SIVD.

[Methods]

To induce chronic cerebral hypoperfusion, we used a mouse model of SIVD with bilateral common carotid artery stenosis (BCAS), which produces prolonged hypoperfusion and white matter ischemic lesions. Expression levels of oligodendrocyte and fibroblast markers were assessed in the corpus callosum following BCAS surgery. For *ex vivo* experiments, CD45^{int} CD11b^{high} microglia were isolated by fluorescence-activated cell sorting (FACS) after BCAS. These microglia were co-cultured with mouse pericytes. The expression of α SMA and fibronectin, myofibroblast markers, was evaluated in pericytes co-cultured with microglia from BCAS or sham mice, as well as in untreated control pericytes. Additionally, FACS-isolated microglia from BCAS mice were subjected to single-cell RNA sequencing (scRNA-seq, 10X Genomics).

[Results]

In *vivo* analyses revealed decreased levels of myelin basic protein and increased expression of laminin and α SMA in the corpus callosum of BCAS mice, as determined by western blotting. In *ex vivo* experiments, pericytes co-cultured with BCAS-derived microglia exhibited increased expression of α SMA and fibronectin compared to those co-cultured with microglia derived from the sham mice. Moreover, scRNA-seq analysis showed that BCAS-induced microglia exhibited an elevated ECM Regulatory Score.

[Conclusions]

These findings suggest that activated fibroblasts may play a crucial role in the pathogenesis of SIVD. In particular, activated microglia may drive pericyte-to-myofibroblast transition, providing a novel therapeutic target for white matter fibrosis in SIVD.

Disclosures: K. Yasuda: None. R. Takahashi: None. R. Matsumoto: None. T. Maki: None.

Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

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Program #/Poster #: PSTR263.16/C43

Topic: B.09. Glial Mechanisms

Support: National Multiple Sclerosis Society Grant RG1901-33209
Mayo Clinic Center for Regenerative Biotherapeutics

Title: Electrophysiological signatures of demyelination and remyelination in the cuprizone model

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Abstract: Background: Cuprizone (CPZ)-mediated demyelination is a well-established model of demyelinating disease that models the process of CNS demyelination and remyelination. To better understand impacts on impulse conduction, we applied electrophysiological methods to

assess conduction velocity (CV) in isolated whole-corpus callosum (wCC) preparations at stages of demyelination and early remyelination and evaluated any correlations with axon integrity and glial populations. Methods: The effects of CPZ on CVs in the corpus callosum were investigated after feeding 10-week-old male C57BL/6J mice a diet containing 0.3% CPZ for 4 weeks (demyelination stage, n=5), or 2 weeks after mice were returned to a standard diet (SD) to elicit early stages of remyelination (n=5). For both groups, age- and sex-matched cohorts of mice were fed a standard diet as controls (n=5). The wCC was dissected from 400 μ m coronal slices and incubated in carboxygenated artificial cerebrospinal fluid. Rostral wCC slices were associated with the lateral ventricles and caudal wCC was associated with para-hippocampal regions. Compound action potentials (CAPs) were recorded by suspending wCC slices between suction electrodes and stimulating to yield 95% maximal responses. The tissue was then immersed in 4% paraformaldehyde and embedded in paraffin. Oligodendrocyte subpopulations (i.e., PDGFR α , Olig2, CC1), MBP, NFH, Iba1, and GFAP were assessed with immunohistochemistry. CAP traces were analyzed by fitting a multimodal gaussian function, resulting in a composite CV metric. Results: At 4 weeks of CPZ-demyelination, composite fast CV was impaired in the wCC, with trending greater reductions caudally. A reduction in CC1+ and Olig2+ cells was seen, while PDGFR α + cells increased. Immunoreactivity for Iba1 and GFAP was increased, and that for NFH and MBP was unchanged. Increases in Iba1, GFAP and PDGFR α were accentuated in caudal sections. At the 4+2 week timepoint of early remyelination, the reductions in wCC CV and CC1+ cells persisted, and significant reductions in immunoreactivity for NFH and MBP were observed. Conclusion: Whole CC CAP recordings represent a sensitive metric for axon function in the CPZ model. Reductions in CV correlated with loss of mature oligodendrocytes and occurred prior to detectable loss of MBP. Taken with changes across the glial and axonal compartments, CPZ-mediated reductions in CV highlight greater impacts of CPZ within the caudal compared to rostral regions of the CC.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

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Program #/Poster #: PSTR263.17/C44

Topic: B.09. Glial Mechanisms

Support: R01 NS120877
R21 NS107946
RG1901-33209

Title: Targeting the Thrombin Receptor to Improve Neural Stem Cell-Mediated Myelin Regeneration

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Abstract: The thrombin receptor (Protease Activated Receptor 1 (PAR1)) is a G-protein coupled receptor (GPCR) activated by proteolytic cleavage in the N-terminus to elicit intracellular signaling. Previously, we established that PAR1 is expressed by adult subventricular zone (SVZ)-derived neural stem cells (NSCs) and demonstrated that switching off the receptor promotes the production and differentiation of NSCs towards the oligodendrocyte lineage. These results are exciting as they suggest that blocking PAR1 function may be a useful method to improve the therapeutic utility of NSC transplantation for myelin regeneration, which is the subject of several completed or ongoing clinical trials.

In this study, we tested the hypothesis that switching off PAR1 in NSCs through genetic targeting increases their capacity to promote myelin regeneration following engraftment into the cuprizone-demyelinated CNS of adult female C57BL6J mice. Quantification of GFP-labeled NSCs 4 wk after engraftment into the corpus callosum demonstrated higher numbers in the case of PAR1 knockout (PAR1^{-/-}) NSCs compared to wild types (PAR1^{+/+}) (P=0.01). Notably, NSCs lacking PAR1 also migrated further along the rostral-caudal extent of the demyelinated corpus callosum compared to wild types (P<0.05, Two Way ANOVA). In addition, 4 wk after engraftment a significantly higher proportion of PAR1^{-/-} NSCs had differentiated into oligodendrocyte progenitor cells (PDGFR α +GFP+, P=0.006), and into mature myelinating oligodendrocytes (ASPA+GFP+, P=0.001, Student's t-test), compared to PAR1^{+/+} NSCs. Complementary RNAseq of PAR1^{+/+} and PAR1^{-/-} NSCs at stages of proliferation highlight differential regulation of Creb, Netrin, Rac, Wnt/b-catenin, and myelinating signaling pathways. Myelination and Wnt/b-catenin signaling continued to be substantially enriched in PAR1^{-/-} NSCs after differentiation to oligodendrocytes, along with axonal guidance, neurovascular coupling, and ephrin signaling, in addition to glutathione-mediated detoxification. The molecular pathways identified here are being further investigated by Western blot, including Creb, Pka, Akt, Gsk3, and Wnt/b-catenin.

Altogether, these new findings suggest that genetic knockout of PAR1 in NSCs enhances their ability to survive, migrate and differentiate towards the myelinating lineage following engraftment into the demyelinated CNS. These studies point to the need for additional investigation into the therapeutic utility of targeting PAR1 to improve stem cell-based remyelinating therapies for CNS demyelinating conditions and other neurological disorders.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

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Program #/Poster #: PSTR263.18/C45

Topic: B.09. Glial Mechanisms

Support: RF1NS128607

Title: Reconsidering the role of oligodendrocyte lineage cells in Parkinson's disease pathogenesis

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Abstract: Parkinson's disease (PD) is the second most common neurodegenerative disorder, characterized by the progressive degeneration of dopaminergic neurons in the substantia nigra. Despite extensive research, the precise molecular mechanisms underlying PD pathogenesis remain poorly understood. Recent single-nucleus RNA sequencing (snRNA-seq) studies in PD patient substantia nigra have revealed transcriptomic alterations in oligodendrocytes and oligodendrocyte precursor cells (OPCs), suggesting these glial populations may play important roles beyond their traditional function in myelination. In this study, we utilized the 6-hydroxydopamine (6-OHDA) mouse model of PD to investigate dynamic changes in OPC proliferation and morphology during dopaminergic degeneration. Furthermore, through comparative snRNA-seq analyses of human PD samples and the mouse model, we identified shared and distinct transcriptomic signatures in oligodendrocytes and OPCs, highlighting their potential involvement in PD pathophysiology. Our findings provide novel insights into implication of oligodendrocyte lineage cells in PD beyond their canonical myelination function, paving the way for the exploration of new therapeutic targets.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.19/C46

Topic: B.09. Glial Mechanisms

Support: CSIR
DST-Inspire
IISER Kolkata

Title: Deciphering Murine- β -Coronavirus induced modulations of oligodendroglial survival signaling: Correlating its implications in neuroinflammatory demyelination

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Abstract: Oligodendrocytes (OLGs) are vital for myelination, provide trophic and metabolic support to axons, and crucial for neuronal health. Communication between OLGs-astrocytes mediated by gap junctions (GJs) like Connexin (Cx) 47-Cx43 GJ intercellular communications (GJICs) and leukemia inhibitory factor (LIF)-LIFR/gp130 signaling supports myelination and cell survival. Research indicates astrocytic LIF is triggered by neuronal ATP, through Cx43 hemichannels, prevents OLGs death and helps in myelination. Clinical studies showed disrupted GJICs and loss of OLGs GJs within Multiple Sclerosis (MS) lesions and immune mediated OLGs apoptosis leads to the demyelination. MHV-A59/RSA59, a dual hepato neurotropic virus induced experimental model mimics certain pathology of human demyelinating disease MS, helps to explore disease mechanism. Thus, we investigated the direct detrimental effects of RSA59 infection on OLGs as well as its impact on its survival pathways. For this we used primary neonatal OLGs precursor cells (OPCs), differentiated OLGs and *In vivo* studies. Results demonstrate RSA59 induces progressive demyelination in brain as confirmed by loss of PLP staining and MBP gene expression. RSA59 directly target OLGs both *in vitro* and *in vivo* and elicits apoptosis validated by tunnel assay. Alongside direct death, RSA59 also alters OLGs survival by impairing Cx47 phosphorylation, affecting its membrane stability and interfering with LIF signaling. Virus downregulates phosphorylation of Cx47 and simultaneously increases nonphosphorylated forms and mRNA expression. It also reduces Cx47 levels in primary OLGs. RSA59 infection triggers LIF expression in brain during acute phase (Day 5 p.i) as a host protective response, but post virus clearance the LIF expression gradually declines towards chronic phase (Day 30 p.i). In contrast, LIFR/gp130 mRNA levels and gp130 protein expression increases progressively towards chronic phase indicating LIF dependent modulation of LIFR/gp130 to restore homeostasis and help in remyelination. Further we investigated ERK1/2 and AKT pathways and observed region specific differential expression of ERK1/2 phosphorylation post RSA59 Infection. It significantly decreased in the cerebellum during acute phase but increased in the cerebrum during acute to chronic transition phase (Day 10 p.i) and also in primary OPCs. A marked decline of Phospho Akt levels was observed in the brain during acute phase of RSA59 infection. This suggests understanding of virus induced OLGs death and alteration of its survival signaling provides insight into unravelling the etiopathology of MS and also a rationale for cell protective therapies.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.20/C47

Topic: B.09. Glial Mechanisms

Support: 84312-NCA-24

Title: Role of disease-associated oligodendrocytes in CNS demyelination and normal aging

Authors: *M. ZHU¹, H. KIM², Y. WANG³, J. PARK⁵, H. CHEN⁶, F. GUO⁴;
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Abstract: Oligodendrocytes (OLs), the myelinating cells of the central nervous system (CNS), are critical for neural connectivity and are increasingly recognized as active contributors to CNS pathology. While their degeneration is a hallmark of multiple sclerosis (MS), OL dysfunction also occurs during normal aging, preceding cognitive decline and neurodegeneration. Recent single-cell transcriptomic studies have identified disease-associated oligodendrocytes (DOLs) with distinct pro-inflammatory and stress-response gene signatures, though their in vivo function remains poorly understood. Our study focuses on a specific DOL subtype expressing Serpina3n (SerpinaOLs). Using Serpina3n-reporter and OL-specific knockout mice, we show that OLs—not astrocytes—are the primary source of SERPINA3N during demyelination and aging. Loss of Serpina3n in OLs leads to reduced neuroinflammation, preserved myelin integrity, and improved pathological outcomes in both autoimmune (EAE) and chemically induced demyelination models. We further investigate the role of SerpinaOLs in age-associated neurodegenerative changes, including OL survival and microglial activation. These findings uncover a non-myelinating role of OLs in modulating CNS inflammation and degeneration and identify SERPINA3N as a promising therapeutic target for both MS and age-related neurological disorders.

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Poster

PSTR263: Oligodendrocyte Biology in Health and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR263.21/C48

Topic: A.07. Development of Neural Systems

Support: Ingrassia Research Award
Harrison Research Award

Title: Defining the role of tmem125b in oligodendrocyte myelination and the microglial-mediated resolution of ectopic myelin wrapping

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Abstract: Oligodendrocytes are essential glial cells in the central nervous system (CNS), responsible for producing and maintaining myelin to ensure efficient neural signal transmission. Single-nuclei RNA sequencing showed that tmem125b is highly expressed in myelinating

oligodendrocytes (mOLs), and we have determined that loss-of-function mutations in *tmem125b* lead oligodendrocytes to aberrantly wrap neuronal cell bodies with myelin. This phenotype emerges during early myelin development, but surprisingly the phenotype resolves by 30 days post-fertilization (dpf). Given our observations and the understudied nature of *tmem125b*, this project consists of two parts that aim to clarify its role in oligodendrocyte and myelin development, and aberrant myelin correction. RNAscope in situ hybridization confirms that *tmem125b* expression is co-localized with the mOL marker *mbp:egfp*. We hypothesize that the spatial localization of *tmem125b* may provide insight into its specific function. Currently, we are using immunocytochemistry, immunohistochemistry, and analysis of a *Tmem125b*-EGFP fusion protein to determine where *tmem125b* is specifically functioning in mOLs. Preliminary findings suggest that *tmem125b* is distributed along the myelin sheath and may be co-localized with a paranode marker, but further research is necessary to confirm whether it exhibits a consistent and specific localization pattern. In part two of this project, we aim to identify the mechanism responsible for phenotype resolution in juvenile *tmem125b* mutants, with emphasis on determining the potential regulatory role of microglia given their known ability to phagocytose excess myelin and undergo developmental maturation in waves. Specifically, we are imaging live embryos beginning at 15 dpf, when ectopic wrapping begins to decline, using fluorescently labeled myelin and microglia to assess their interactions. We hypothesize that a second wave of mature microglia clears ectopic myelin during this correction phase. Together, this study seeks to elucidate the functional role of *tmem125b* in oligodendrocyte behavior and to determine whether distinct microglial populations contribute to CNS myelin remodeling during development.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.01/C49

Topic: C.01. Brain Wellness and Aging

Support: NIH/NIA Grant R25AG076396
NIH/NIAAA Grant F31AA031440
NIH/NIDDK Grant R01DK099334
McKnight Brain Research Foundation

Title: Is statin use associated with brain-predicted age in the cognitively-intact oldest-old?

Authors: *J. JUHASZ¹, M. BRITTON¹, H. HOOGERWOERD¹, K. JOHNSON¹, P. STEWART³, S. NOLIN⁴, D. A. RAICHLEN⁵, G. E. ALEXANDER⁶, B. E. LEVIN⁷, T. RUNDEK⁸, V. A. DEL BENE⁹, K. M. VISSCHER¹⁰, R. A. COHEN¹, J. GULLETT¹, E. C. PORGES²;

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Angeles, CA; ⁶Dept. of Psychology, Univ. of Arizona, Tucson, AZ; ⁷Neurol., ⁸Univ. of Miami, Miami, FL; ¹⁰Neurobio., ⁹Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Introduction: Statins (HMG-CoA reductase inhibitors) are commonly used to treat hyperlipidemia and prevent cardiovascular disease. Preclinical rodent studies suggest statins benefit brain health in aging. However, human literature is mixed in older adults and limited in the oldest-old (85+). Brain-predicted age is a brain-based biomarker of biological aging that uses machine-learning analysis of neuroimaging data to predict age from brain structure. Brain-predicted age difference (brainPAD) is defined as the predicted age minus the chronological age. As brainPAD values become more positive (predicted age is higher than chronological age), hazard of dementia and mortality increase. This project evaluates whether statin use is associated with brainPAD in the cognitively-intact oldest-old, hypothesizing a relationship between statin use and more negative brainPAD values. Methods: T1-weighted structural MRI data and self-reported statin use were collected in 157 (54% female) cognitively-intact oldest-old human participants with a mean chronological age of 88.34 years (87.46 for statin users and 88.87 for nonusers). 48% of participants reported hyperlipidemia and 57% reported hypertension, with 156 and 150 individuals reporting, respectively. Sex was not associated with statin use. BrainageR software was used to generate brainPAD from T1-weighted structural images. Difference in brainPAD between statin users and nonusers was tested with a Mann-Whitney U test and with multiple regression adjusted for age, hyperlipidemia, and hypertension. Results: 38% of participants reported statin use. Among participants on statins, 81% had hyperlipidemia and 49% had hypertension. Statin use was not associated with brainPAD in bivariate models ($U=2750$, $p = 0.616$) or after adjustment for age, hyperlipidemia, and hypertension ($B = 0.172$, $p = 0.87$). Hyperlipidemia ($B=-0.621$, $p=0.538$) and hypertension ($B=0.064$, $p=0.942$) were not associated with brainPAD. Conclusion: Statin use, hyperlipidemia, and hypertension may not associate cross-sectionally with brainPAD in the cognitively-intact oldest-old. Cognitively-intact oldest-old individuals are heavily selected and likely healthier than their birth cohort peers, therefore survival bias may have attenuated associations commonly-found in younger age groups. Longitudinal studies may improve understanding of the relationship between statin use and brain aging by testing for an association with changes in brainPAD as individuals age and allowing for the collection of medication adherence data.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.02/C50

Topic: C.01. Brain Wellness and Aging

Support: AHA

Title: Chronic kidney disease in old age contributes to severe ischemic stroke and poor outcomes

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Abstract: Chronic kidney disease (CKD) has a graded and independent inverse impact on cerebrovascular health due to shared hemodynamic autoregulation, endothelial dysfunction, and inflammation. Stroke risk is high in the aged population with kidney dysfunction compared to adult population. CKD induced mechanisms and its significance in old age is unknown due to lack of preclinical models. We investigated in the current study how CKD progress in the aged mice and how it impact brain damage after stroke. Adult (3 months) and age (18 months) C57BL/6 mice were administered with daily injections of adenine (50mg/Kg, intraperitoneally) for up to 14 days. CKD progression assessed using a combination of hematological and non-invasive imaging methods including dual-energy X-ray absorptiometry, and ultrasound on days 7 and 14. Mice were subjected to 60 minutes (adults; on 14 day)/ 30 minutes (aged; on 7 day) of focal cerebral ischemia, followed by 3 days of reperfusion. Post-stroke brain damage and renal functions were evaluated noninvasively. Post-stroke dysfunction was assessed by the modified neurological severity score, beam walk test and rotarod test. The blood levels of creatinine and urea nitrogen were significantly elevated in both groups and the levels in aged cohorts on 7 days of CKD are equivalent to levels at 14 days in adults. This suggests age associated vulnerability to CKD. In addition, microbubble assisted ultrasound showed a significant reduction in renal perfusion, an indicator of glomerular filtration rate in both adult aged mice. Brain damage was assessed by T2-MRI and both mice showed accelerated brain damage compared to healthy mice that underwent stroke. Aged mice with CKD+stroke showed severe disability and high mortality when compared with the adults. We conclude that mild CKD in aged mice exacerbate post-stroke mortality when compared to the moderate CKD in adult mice.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.03/C51

Topic: C.01. Brain Wellness and Aging

Support: the Natural Science Foundation of China Grant 32171085

Title: Cognitive modeling of the episodic memory task as an early detection for mild cognitive impairment

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Abstract: Background: Mild cognitive impairment (MCI) represents a critical early stage of Alzheimer's disease (AD), where timely detection is essential for intervention. However, current biomarker-based assessments are invasive, costly, and often inaccessible in primary care settings, while conventional and commonly practiced neuropsychological tests suffer from subjectivity and poor standardization. There is a pressing need for objective, scalable, and biologically relevant cognitive screening tools. Method: This study employed an old/new recognition episodic memory modeled with Multinomial Processing Tree (MPT) to extract memory success probability (ρ) and guessing bias (γ). The performance of these MPT parameters was compared with the signal detection theory (SDT) parameters (d' for memory discriminability, c for response bias) in their capacity to predict hippocampal volume, AD-signature cortical thickness (ADSCt), and to distinguish MCI. This comparison was done with or without the reaction time (RT) as an additional predictive indicator. A total of 573 participants were involved. Results: MPT-derived parameters demonstrated superior performance over signal detection theory (SDT) parameters in classifying MCI (AUC = 0.70 vs. AUC = 0.66) and predicting hippocampal volume and ADSCt. Incorporating RT further enhanced classification accuracy for MPT (AUC increased to 0.76) and improved the prediction of neurodegenerative biomarkers. Conclusion: Decomposing cognitive processes within the episodic memory task into mechanistic behavioral parameters, combined with RT measurement, enhances the sensitivity and biological relevance of early AD screening tools. The study offers a novel, objective, and scalable approach for early MCI detection. This framework shows great potential for clinical translation and community-based cognitive screening programs.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

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Program #/Poster #: PSTR264.04/C52

Topic: C.01. Brain Wellness and Aging

Support: National Science and Technology Council, Taiwan: NSTC 110-2926-I-A49-001-MY4
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National Science and Technology Council, Taiwan: NSTC 111-2634-F-A49-014

National Science and Technology Council, Taiwan: NSTC 112-2634-F-A49-003

Title: Regional brain aging trajectories in major depressive disorder: A brain age prediction study

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Abstract: Background: Major depressive disorder (MDD) is increasingly associated with accelerated brain aging, measured by the brain age gap (BAG)—the difference between predicted brain age and chronological age. However, little is known about how illness duration affects regional brain aging patterns. **Methods:** Participants included 111 patients with MDD and 434 healthy controls (HCs). The MDD group had a mean age of 56.7 ± 16.3 years, with 59% females and an average of 11.6 ± 4.2 years of education. The HC group had a mean age of 44.5 ± 16.3 years, with 61% females and an average of 15.5 ± 4.0 years of education. Depression severity in the MDD group, assessed using the 21-item Hamilton Depression Rating Scale (HAM-D), yielded a mean score of 11.1 ± 7.2 . Structural (T1-weighted MRI) and functional (resting-state fMRI) data were processed to extract gray matter volumes and functional connectivity across 90 AAL-defined brain regions. A support vector regression model was trained on HC data to predict brain age. BAG was calculated and MDD patients were grouped into 5-year sliding illness-duration windows with 1-year steps. To minimize age-related bias, BAG values were corrected for chronological age using linear regression, and group-level comparisons were conducted on the residualized BAG scores. **Results:** The brain age model trained on healthy controls achieved a bias-corrected mean absolute error (MAE) of 5.39 years and a Pearson correlation of $r = 0.93$ in the test set. When applied to MDD patients, performance declined (MAE = 6.24; $r = 0.91$), indicating deviation from normative aging trajectories. The sliding window analysis revealed *spatiotemporal heterogeneity* in BAG trajectories across brain regions. Orbitofrontal and medial prefrontal areas showed early BAG elevation in the course of illness, whereas subcortical regions such as the caudate and insula exhibited more pronounced BAG differences in later stages. These results suggest that different brain regions diverge from normative aging at different stages of depression. **Conclusion:** This study introduces a temporally dynamic framework to examine brain aging in MDD across illness duration. By mapping region- and stage-specific BAG trajectories, it aims to reveal key brain areas vulnerable to progressive aging in depression. These insights could inform future diagnostic and therapeutic strategies tailored to illness stage.

Disclosures: Y. Chen: None. A. Yang: None.

Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.05/C53

Topic: C.01. Brain Wellness and Aging

Support: R01 AG 079957
RF1 AG 082201
USC Provost's Research Fellowship

Title: Genome-wide clustering of allelic variants identifies distinct regional patterns of brain aging

Authors: *N. KIM, A. MISHRA, N. CHOWDHURY, A. IRIMIA;
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Abstract: Local brain age gap (LAG)—the difference between deep learning-predicted and chronological age at the regional level—offers a promising biomarker for understanding heterogeneous brain aging processes. While prior genome-wide association studies (GWAS) have identified genetic loci associated with global brain age, the polygenic architecture underlying regional patterns of aging is poorly characterized. To address this gap, we studied whether single-nucleotide polymorphisms (SNPs) with shared cortical association patterns could be grouped into biologically meaningful clusters. Identifying these region-specific genetic signatures is critical for understanding the biological mechanisms that drive spatially uneven brain aging.

Using MRI data from 41,859 cognitively normal individuals in the UK Biobank (aged 45-83 years; 21,983 females), we performed a voxel-level deep learning analysis to predict local brain age and compute LAG. A GWAS was conducted to identify SNPs associated with LAG across 148 cortical regions. From these results, we selected 220 SNPs that were significantly associated with LAG in >7 regions. For these SNPs, we constructed cortical beta coefficient correlation maps and applied Uniform Manifold Approximation and density-based spatial clustering to identify SNP clusters based on shared spatial association patterns. For each cluster, cortical association maps were averaged across SNPs and gene set enrichment was performed using Stanford's Genomic Regions Enrichment of Annotations Tool.

This analysis yielded three distinct SNP groups. Group A (106 SNPs) exhibited widespread positive LAG associations in midline and parietal regions, including the precuneus and posterior cingulate. Group B (106 SNPs) exhibited widespread negative associations concentrated in the anterior cingulate, insula, and temporal cortex. Group C (6 SNPs) displayed focal positive effects in the frontoinsular and perisylvian cortices. Each group was enriched for distinct gene sets: Group A included KCNK2, PRDM6, and BMP6, and was linked to steroid hormone biosynthesis; Group B included NUA1 and LPAR1, with enrichment in cytoskeletal and signaling processes; Group C mapped to immune-related genes such as TRIM38, linked to interferon signaling. These spatially distinct genetic clusters suggest that different biological pathways contribute to aging in unique cortical regions, reinforcing the concept of mosaic brain aging. Understanding these pathways provides insight into why certain brain regions may be more vulnerable to age-related decline and points to potential targets for developing region-specific biomarkers and interventions.

Disclosures: N. Kim: None. A. Mishra: None. N. Chowdhury: None. A. Irimia: None.

Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.06/C54

Topic: C.01. Brain Wellness and Aging

Title: Associations between physical activity and neuroimaging biomarkers: findings from the LifeAfter90 (LA90) study

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Abstract: Leisure time physical activity (PA) is a modifiable lifestyle factor linked to brain health, yet its relationship with neuroimaging biomarkers remains understudied among the oldest-old. We tested associations between PA and markers of neurodegeneration and cerebrovascular disease in adults aged 90 and older. The LifeAfter90 Study (2018-2024) included a neuroimaging subsample of 240 participants aged 90+. Light PA was assessed using two items- light house/yard work and light exercise/sports, and vigorous PA using two items- vigorous house/yard work and vigorous exercise/sports. Items were rated on a 5-point scale: 0 (never), 1 (several times a year), 2 (several times a month), 3 (several times a week), and 4 (every day or almost every day). For each PA type, the two items were averaged to create continuous scores, with higher scores reflecting more frequent activity. We used PA data from the visit closest in time to each participant's MRI scan. Neuroimaging outcomes included hippocampal volumes (total, right and left) and log-white matter hyperintensities (WMH), measured by 3T MRI and adjusted for intracranial volume. Linear regression models tested associations between PA and brain imaging markers, adjusting for age and sex. Sex-stratified models were used to explore potential sex differences. Participant mean age was 93.2 (± 2.2) years, and 56.9% were women. About 24% and 11% of participants reported frequent vigorous house/yard work and sports/exercise, respectively, while 66% and 65% reported frequent light house/yard work and sports/exercise (several times a week to every day). Higher levels of light PA were associated with significantly lower WMH volume in both unadjusted ($\beta_{\log WMH} = -0.14$, 95% CI -0.26, -0.01) and adjusted ($\beta_{\log WMH} = -0.14$, 95% CI -0.27, -0.02) models. Vigorous PA was not associated with WMH (Adjusted: $\beta_{\log WMH} = -0.05$, 95% CI -0.18, 0.08). Frequency of neither light (Adjusted: $\beta_{total} = 0.05$, 95% CI -0.07, 0.18; $\beta_{right} = 0.10$, 95% CI -0.02, 0.23; $\beta_{left} = -0.01$, 95% CI -0.14, 0.12) nor vigorous PA (Adjusted: $\beta_{total} = -0.04$, 95% CI -0.18, 0.09; $\beta_{right} = -0.002$, 95% CI -0.13, 0.13; $\beta_{left} = -0.08$, 95% CI -0.21, 0.06) was associated with total, right, or left hippocampal volume. No significant associations or sex differences were observed in sex-stratified models. Finally, frequency of light PA was associated with lower WMH volume, while frequency of vigorous PA showed no significant associations with WMH or hippocampal

volumes. These findings underscore the potential cerebrovascular benefits of light household and recreational physical activities and highlight the importance of feasible lifestyle interventions for brain health in the oldest-old.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

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Program #/Poster #: PSTR264.07/C55

Topic: C.01. Brain Wellness and Aging

Support: 2023R1A2C2006894
RS- 2024-00409554

Title: Age-related mechanical softening of the brain alters astrocyte function and extracellular matrix organization

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Abstract: Aging is known to alter the mechanical properties of brain tissue, particularly by reducing stiffness, which is regulated by neuronal myelin and astrocyte-derived extracellular matrix (ECM). Although these biomechanical changes are likely to impact neuronal and glial cell function, the underlying mechanisms remain poorly understood. In this study, we investigated how age-associated reductions in brain stiffness affect astrocyte behavior. To model the mechanical microenvironment of aged and young brains, we cultured primary mouse astrocytes on polyacrylamide gels with defined stiffness: soft (1.2 kPa) to mimic aged brain tissue, and stiff (3.5 kPa) to represent young brain conditions. After three days of culture, astrocytes on soft gels displayed a rounded morphology, whereas those on stiff gels exhibited a stellate, more differentiated shape. qRT-PCR analysis revealed decreased expression of Lamin B1 and increased expression of senescence markers p21, p53, and IL-6 in astrocytes on soft gels, indicating a senescence-like phenotype. Moreover, expression of ECM-related genes, including Tnc, Fibronectin, Colla1, and Itgb1, was significantly upregulated under soft matrix conditions. Functionally, astrocytes cultured on soft substrates exhibited reduced uptake of glutamate and amyloid-beta—key molecules involved in brain homeostasis—compared to those on stiff substrates. Notably, when mild oxidative stress was applied, astrocytes on soft gels showed more pronounced changes in stress-responsive gene expression. These findings suggest that age-associated changes in brain stiffness can modulate astrocyte morphology, gene expression, and function. Our results highlight mechanotransduction as a critical regulator of astrocyte behavior and a potential contributor to aging-related neurobiological decline.

Disclosures: E. Chang: None. O. Kim: None. H. Lee: None.

Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.08/C56

Topic: C.01. Brain Wellness and Aging

Support: National Institute of Health [R01 AG053952 (PI: KE); R35 AG072307 (PI: KE)]

Title: Associations between physical activity, fitness and depressive symptoms in older adulthood: Examining the moderating role of sex, amyloid pathology, and APOE4 carriage

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Abstract: BACKGROUND: Physical activity (PA) can improve mood and reduce depressive symptoms in late adulthood. Evidence shows greater benefits of PA and fitness in females relative to males for depressive symptoms. However, little is known about the relationship of PA to subclinical depressive symptoms, which are prevalent and increase the risk for clinical depression in late adulthood, and whether these associations may differ by sex and risk factors for cognitive and functional decline (A β -positivity; Apolipoprotein E ϵ 4 (APOE4) carriage). In a large, well-characterized sample of older adults without clinical depression, we examined the role of sex, amyloid pathology, and APOE4 carriage in moderating the relationships between cardiorespiratory fitness (CRF) and PA with depressive symptoms. METHODS: CRF (VO₂peak) was measured with a graded exercise test on a motorized treadmill. Moderate-to-vigorous physical activity (MVPA) was measured using a wrist-worn accelerometer (ActiGraph GT9X Link) over seven days and processed using GGIR. The Hospital Anxiety and Depression Scale was used to measure symptoms of depression and anxiety. Plasma phosphorylated tau₂₁₇ (ptau₂₁₇) was collected and used to classify A β status. Separate linear regression models adjusting for age, years of education, anxiety symptoms, study site, and body mass index evaluated the associations between CRF and MVPA with depressive symptoms. Interaction terms examined moderation by sex, A β status, and APOE4 carriage. RESULTS: The sample included 648 older adults (Mage = 69.88; SD=3.75; females=71.1%). No significant differences were found between males and females in depressive symptoms (p=0.809). Participants had an average VO₂peak of 21.7(5) mL/kg/min and MVPA of 31.2(26) min/day. VO₂peak and MVPA

were not associated with depressive symptoms (all $p > .05$). However, sex moderated the association between VO₂peak and depressive symptoms ($\beta = -.31$; $p = .04$), such that higher CRF was related to fewer depressive symptoms in females ($\beta = -.19$; $p \leq .001$) but not in males ($\beta = -.03$; $p = .620$). For MVPA, there were no significant sex x MVPA interactions. However, sex-stratified analyses revealed that higher MVPA was associated with fewer depressive symptoms only in females ($\beta = .10$, $p = .02$). No significant moderation effects were observed for amyloid pathology or APOE4. **CONCLUSIONS:** Among community-dwelling older adults without clinical depression, we observed that sex moderated the association between CRF and subclinical depressive symptoms, such that higher fitness was associated with fewer depressive symptoms in females. Future work with broader depressive symptom variability is needed.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.09/C57

Topic: C.01. Brain Wellness and Aging

Support: Sammon's Foundation

Title: Network-based latent modeling of brain health: predicting longitudinal change from task-evoked hemodynamic responses.

Authors: *Q. COPPOLA¹, M. D'ESPOSITO³, S. B. CHAPMAN⁴, A. SEITZ²;
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Abstract: Precision brain health hinges on assessment strategies and biomarkers that accurately capture and predict the longitudinal interdependencies of brain change. Creating affordable and scalable multi-dimensional health assessment batteries for longitudinal monitoring requires validation with key neural biomarkers. The BrainHealth Index™ (BHI) is a multi-dimensional battery consisting of 20 behavioral measures assessing cognition, emotional well-being, and real-world functioning that can be administered through an online platform, providing an affordable and accessible testing paradigm for longitudinal monitoring. Recent work has shown that task-evoked hemodynamic response function (HRF) metrics predict change in BHI factor scores extracted from exploratory factor analysis, specifically in the domain of cognition. However, BHI may be better characterized via more sophisticated latent models that utilize networks that summarize latent-level dependencies or residual task-level interactions. Here, we develop two network-based latent models to characterize multidimensional brain health using existing BHI

measures and functional imaging data from 48 healthy adults (Age Range: 21-65) scanned twice, approximately 6 months apart, during a checkerboard task. For both sessions, we extracted HRF features (baseline canonicity via integrated squared difference from a canonical HRF; peak amplitude; full width at half-maximum) from the 2,500 most responsive voxels. We reorganized BHI manifest variables into three modified latent domains: Cognition, mood, and socio-personal factors. This structure was fit in both (i) a latent network model (LNM), which fits a regularized partial network between latent domains and (ii) a residual network model (RNM), which models a regularized partial network of residual conditional dependencies among manifest variables after accounting for latent factors. Machine learning techniques are employed to examine the predictive relationship between key HRF features and BHI change scores in both models. We investigate critical differences in predictive performance that may be dependent on the canonicity of baseline HRFs. By comparing predictive performance across more sophisticated, data-driven models, this study aims to refine BHI measurement through network-driven latent modeling validated against simple, task-evoked HRF metrics. Ultimately, this advances the validity of BHI as a tool for precision, longitudinal monitoring of key indices related to brain health.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.10/C58

Topic: C.01. Brain Wellness and Aging

Title: Two-channel sleep eeg-based brain age prediction model development

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Abstract: Neurophysiological age can be predicted using machine learning approaches applied to sleep or resting-state electroencephalography (EEG). However, current sleep EEG approaches rely solely on comprehensive polysomnography with six-channel EEG, which is highly cost-ineffective. To facilitate accessibility of such methods through home sleep studies or wearables, we aimed to develop and validate a brain age prediction model based on a two-channel frontal EEG. To train the brain age prediction model, we utilized data from 1,259 healthy sleepers (HS) provided by Samsung Medical Center. We analyzed EEG recordings from two frontal channels (F3, F4), which are commonly used in wearable settings. The EEG data were converted into scalograms (2,000 times $\times$ 16 frequencies) for training with a ConvNeXt model, which has been chosen for its robustness in learning hierarchical representations from time-frequency images,

and its computational efficiency in processing moderate-sized datasets compared to transformer-based architectures. A brain age index (BAI) was calculated by subtracting chronological age from predicted brain age, indicating the degree of brain aging acceleration relative to chronological age. To validate the two-channel-based BAI, we correlated it with the BAI predicted from a previously validated six-channel model. Additionally, to assess whether the two-channel BAI can capture increased brain aging due to sleep disorders, we evaluated BAI values in participants with obstructive sleep apnea (OSA) and insomnia. The proposed two-channel EEG-based brain age model achieved a mean absolute error (MAE) of 6.8 years. Furthermore, it significantly correlated with the six-channel BAI ($r=0.52$, $p < 0.0001$). In groups with sleep disorders, BAI was significantly elevated compared to healthy sleepers: OSA group by 2.3 ± 7.7 years and insomnia group by 3.1 ± 6.5 years (both $p < 0.05$). The proposed method sensitively detects brain aging acceleration in individuals with sleep disorders such as OSA and insomnia, highlighting its effectiveness in identifying disorder-related brain health deterioration. This model based on the minimum number of EEG channels enables home sleep studies or wearable solutions for daily monitoring of brain health outside clinical settings.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.11/C59

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grants 1UG3EB036465

Title: Using Deep Learning for the Prediction of the Amyloidogenic Propensity of Non-Classical neurodegenerative disease related Biomarkers

Authors: *Y. HAN¹, F. BOUKARI²;

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Abstract: Protein biomarkers have been a classical way to recognize neurodegenerative disorders including Alzheimer's, Parkinson's, and Huntington's diseases. Each of these disorders has its own distinct protein biomarkers that are unique to the disease, such as Amyloid-beta for Alzheimer's disease. Although there has been expansive research into well-known biomarkers such as Amyloid-beta, there exist less researched proteins like Neurogranin (NRGN), Progranulin (PGRN), Growth Associated Protein 43 (GAP-43), and Neuronal Pentraxin 2 (NPTX2) which have the potential to serve as biomarkers for neurodegenerative disorders. This work seeks to utilize Deep Learning models to evaluate/analyze the folding and misfolding of these under-researched proteins and understand their potential roles in neurodegenerative disease. Using AlphaFold and related protein folding software, we will first establish the structures of these proteins, then build a Deep Learning model which predicts the behavior of

these under-researched proteins to establish the relatedness of disease progression and behavior. By comparing the disease progression related to these lesser-studied proteins with that of well-characterized biomarkers, we hope to find new insights into the mechanisms of neurodegenerative disorders. This study of these proteins and any new emerging protein biomarkers could enhance diagnostic accuracy and provide new solutions for therapeutic intervention in neurodegenerative diseases.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR264.12/C60

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: The eye-tracking-based Stroop test offers novel features that can provide more accurate detection of frontal lobe impairment through machine learning

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Abstract: Background People with dementia often show attention deficits and frontal lobe impairment. The assessment of these cognitive functions is clinically essential for the early detection and differential diagnosis of dementia. The Stroop test has been used to assess attention and frontal lobe functions, and its validity and reliability are well established. We previously developed an eye-tracking-based Stroop test and demonstrated its validity and utility. **Objective** The eye-tracking-based Stroop test can provide scores for each item as independent features. This study aimed to determine whether the use of these features in machine learning could lead to improved accuracy in screening for frontal lobe impairment. **Design** The study cohort included 97 individuals with a mean age of 76.4 (SD 7.1) recruited from the memory clinic at Osaka University Hospital. All participants completed both a traditional pen-and-paper Stroop test and the eye-tracking-based Stroop test on the same day. The Mini-Mental State Exam (MMSE), Frontal Assessment Battery (FAB), and Trail Making Test - A/B were performed to assess participants' global cognitive and frontal lobe impairments. The eye-tracking-based Stroop test consisted of four distinct color and word tests, including both congruent and incongruent conditions, and took approximately 3 minutes to complete. It provided both a total score and scores for each individual question. **Results** The participants' mean MMSE score was 24.1 (SD 4.4). To detect individuals with frontal lobe impairment (defined as an FAB score <11), the total score from the eye-tracking-based Stroop test achieved an AUC-ROC of 0.716 (95% CI 0.60-

0.81). When the score from each question was used as an independent feature, random forest analyses yielded an AUC-ROC of 0.833 (95% CI 0.60-0.98). **Conclusion** The eye-tracking-based Stroop test may enhance the accuracy of detecting frontal lobe impairment by adding the scores from each question as explanatory variables for machine learning.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

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Program #/Poster #: PSTR264.13/C61

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ROI AG066683
P30 AG066519

Title: Tracking Memory: Eye Movements as Biomarkers for Age-Related Episodic Memory Decline

Authors: *H. SALIM¹, C. E. STARK¹, C. R. VANDERLIP²;

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Abstract: This study investigated whether eye movements during image viewing could serve as non-invasive biomarkers for age-related episodic memory decline. As the global population ages, early detection of cognitive changes is crucial for timely intervention, necessitating accessible methods to detect subtle memory deficits. We employed eye-tracking technology to measure visual exploration patterns while participants viewed repeated images in an Implicit Mnemonic Similarity Task, and assessed episodic memory using the Rey Auditory Verbal Learning Test. Participants included 23 younger and 7 older adults, with eye movement parameters (fixation duration, fixation count, saccade amplitude, and saccade velocity) analyzed and compared against memory performance. Results revealed significant age-related differences in visual exploration strategies, with older adults exhibiting increased fixation counts and younger adults demonstrating greater saccade amplitudes and velocities. However, contrary to our hypothesis, these eye movement parameters did not correlate with episodic memory performance. While our findings confirm age-related changes in both memory function and visual exploration patterns, they suggest the relationship between these domains is more complex than anticipated. This research highlights the need for more sophisticated approaches to understanding the interplay between visual processing and memory in aging populations, potentially incorporating more complex visual tasks that better mimic real-world scenarios. Future studies with larger sample sizes and longitudinal designs may further elucidate the potential of eye-tracking as a biomarker for cognitive decline.

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Poster

PSTR264: Aging: Machine Learning, Modeling, and Biomarkers

Location: SDCC Halls B-H

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Program #/Poster #: PSTR264.14/C62

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: U01AG082350-01A1
U01AG068057-04

Title: Evaluation of Deep Learning Algorithms to Predict Multiple Dementia-Related Neuropathologies from Brain MRI, Clinical and Genetic Data

Authors: T. CHATTOPADHYAY¹, S. C. JOHNSON⁴, E. C. MORMINO⁵, S. I. THOMOPOULOS², *P. M. THOMPSON³;

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Abstract: Most individuals with Alzheimer's disease and related dementias (ADRD) harbor multiple co-existing neuropathologies-such as amyloid- β (A β), tau, cerebral amyloid angiopathy (CAA), TDP-43, and Lewy body disease (DLB)--making accurate differential diagnosis essential for targeted therapies. We compared two deep learning frameworks (Fig. 1) to infer molecular pathologies directly from in vivo structural MRI, supplemented with demographic, clinical, and genetic covariates. Using autopsy-confirmed diagnostic labels from the National Alzheimer's Coordinating Center dataset (N=939; age: 76.0 ± 12.0 , 390F/549M), we trained and evaluated models across five binary classification tasks: A β , Lewy bodies, hippocampal sclerosis (HPSC), TDP-43, and CAA. As HPSC and TDP-43 labels were unevenly distributed across diagnostic groups (613 dementia, 201 cognitively normal, 125 MCI), we manually balanced training and validation sets and included time from imaging to death and death to autopsy as covariates. The AutoGluon model, based on 2D MRI slices, achieved balanced accuracy between 0.54-0.70, with strong performance for A β (F1: 0.91) but poor results for HPSC (0.17) and CAA (0.40). Performance was consistent across axial and coronal planes, indicating minimal sensitivity to slice orientation. By contrast, our hybrid CNN, which integrates 3D MRI with non-imaging features, achieved superior balanced accuracy across all tasks (A β : 0.74, DLB: 0.67, HPSC: 0.67, TDP-43: 0.72, CAA: 0.76) and improved F1 scores for DLB (0.68), TDP-43 (0.61), and CAA (0.71), with modest gains for A β and HPSC. These findings underscore the limitations of 2D models and the advantages of combining 3D spatial context with multimodal data. Our results suggest deep multi-modal fusion is a promising approach for neuropathological classification, even in the absence of PET or biofluid biomarkers.

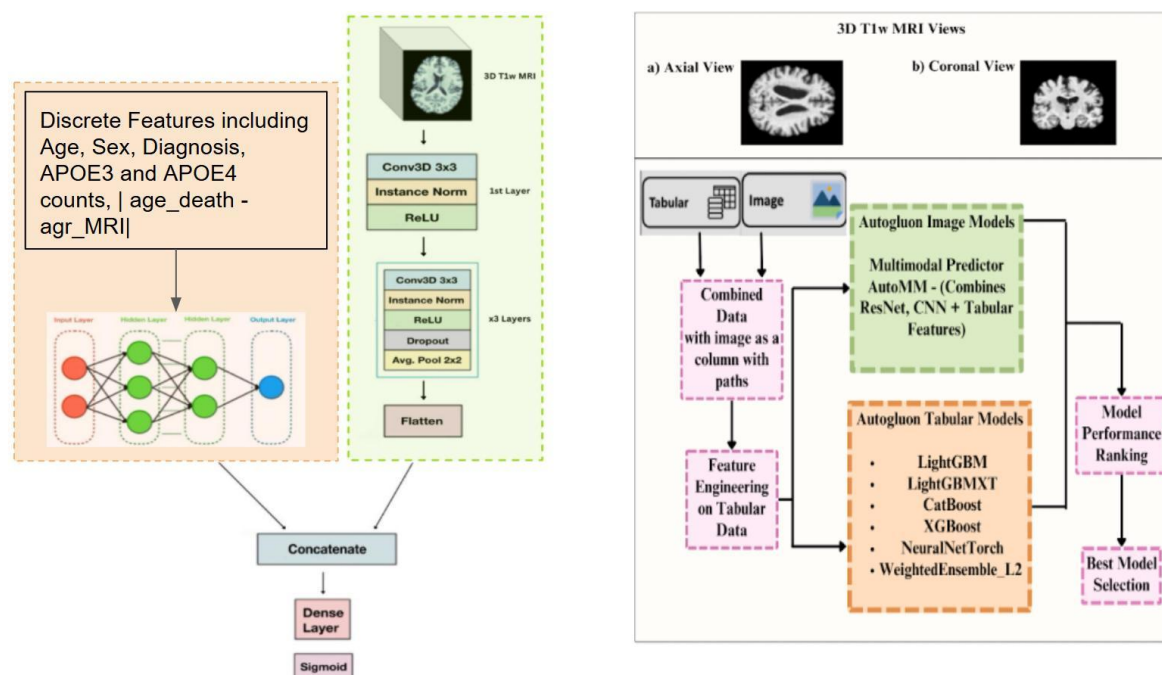


Fig. 1 (a) Deep Learning Algorithms to Predict Neuropathology from Brain MRI. The hybrid CNN architecture uses a 3D convolutional neural network (CNN) to process neuroimaging with an artificial neural network (ANN) for clinical and genetic data in a Y-shaped architecture. (b) For comparison, the AutoGluon ensemble model was trained with 2D axial or coronal slices (middle brain slices) and the same tabular features: sex, diagnosis, age at scan, APOE4 count, APOE3 count, and the absolute age difference between death and scan time. Models reproduced from Chattopadhyay, T., et al. (2025). Deep learning to predict future cognitive decline: A multimodal approach using brain MRI and clinical data. bioRxiv. <https://doi.org/10.1101/2025.02.21.639520>

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Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.01/C63

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA AG079838

Title: Endogenous γ -Secretase Is Linked to Phagocytic Activity in Microglial Cells

Authors: *E. WILLIAMS¹, M. HOUSER¹, S. TORRES¹, N. WIECKIEWICZ¹, M. SADEK², M. YOKOMIZO¹, M. MAESAKO^{1,2};

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Abstract: Gamma-secretase has been primarily studied in neurons, whereas increasing evidence highlights its importance in microglia. Previous research has shown that the pharmacological inhibition of gamma-secretase impairs microglial phagocytic activity. In this study, we used a genetically encoded Förster resonance energy transfer (FRET)-based biosensor to record gamma-

secretase activity, aiming to determine if naturally occurring cell-by-cell variations in endogenous gamma-secretase activity are associated with phagocytic activity. Using the Notch1 N100 Y-T biosensor, we found that the regulation of endogenous gamma-secretase activity varies among individual BV-2 microglial cells. Furthermore, our multiplexed time-lapse imaging revealed that the phagocytosis of E. coli bioparticles was impaired in cells with lower gamma-secretase activity compared to those with higher activity. Complementary biochemical analysis, utilizing Zymosan bioparticles and fluorescence-activated cell sorting (FACS), further demonstrated that cells with reduced phagocytic activity exhibited decreased endogenous gamma-secretase activity. Collectively, our confirmatory study supports previous findings that microglial phagocytic activity is closely linked to gamma-secretase and emphasizes the essential role of gamma-secretase in microglia.

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Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.02/C64

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: A versatile toolbox of human iPSC-derived microglia for disease modelling and multicellular in vitro models for neurodegeneration drug discovery

Authors: N. MATHAROO¹, P. BARTON², E. YATES¹, H. GARNETT¹, S. MILDE¹, L. KOWALSKI¹, R. HICKMAN¹, S. BARBARIA³, C. SCHMIDT³, M. GAMPERL⁴, S. SALIC-HAINZL³, *R. NORTHEAST², K. FIRTH¹, W. BERNARD²;

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Abstract: Microglia, the resident macrophages of the brain, are essential for neural homeostasis. They regulate neurogenesis, synaptic remodelling, and act as first responders to injury or infection. Dysregulated microglial function is implicated in neurodegenerative diseases such as Alzheimer's disease (AD). Despite their critical role in disease progression, existing in vitro models fail to replicate the complexity of microglia, limiting advances in drug discovery. Therefore, new tools are needed to model disease and track cellular responses in co-culture systems.

In this study, we used opti-ox™, a deterministic cell programming technology, to generate scalable human-induced pluripotent stem cell (hiPSC)-derived microglia from both male and female genetic backgrounds. These microglia express key markers, including CD45, P2RY12, CD11b, CD14, IBA1, and TREM2, and exhibit robust phagocytic activity, including uptake of pHrodo labelled BioParticles and secretion of pro-inflammatory cytokines, with distinct responses based on genetic background. Microglia from both backgrounds were benchmarked

against the HMC3 immortalised cell line, and exhibited superior phenotypic and functional characteristics, including more accurate gene expression profiles, enhanced phagocytosis, and higher expression of key microglial markers.

To support complex model development in multicellular and 3D culture system development, we engineered GFP-expressing microglia for live-cell imaging, antibody-free cell sorting, and real-time tracking within multicellular and 3D culture systems.

To advance AD modelling, we engineered hiPSC-derived microglia with AD-relevant mutations, such as TREM2 (R47H) and APOE (C112R), known to alter microglial function. These models exhibited phenotypic changes, including reduced phagocytosis in the APOE (C112R) lines as expected. Ongoing work includes assessing phagocytosis of fluorescently labelled Amyloid- β (1-42) and analysing bulk RNA sequencing data following LPS stimulation to further elucidate disease-relevant phenotypes.

In conclusion, our microglia models provide a physiologically relevant platform for investigating AD mechanisms, supporting critical advancements for drug discovery and neurodegeneration research.

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Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.03/D1

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH AG079517

Title: Microglia-to-neuron signaling increases lipid droplet metabolism, enhancing neuronal network activity

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⁴Scripps Res. Inst., La Jolla, CA

Abstract: Microglia are key players in neuronal circuit plasticity and disrupting their homeostatic function has detrimental effects on neuronal circuit health. Neuroinflammation contributes to the onset and progression of neurodegenerative diseases, including Alzheimer's disease (AD), with several microglial activation genes linked to increased risk for these conditions. Inflammatory microglia alter neuronal excitability, inducing metabolic strain. Interestingly, expression of *APOE4*, the strongest genetic risk factor for AD, affects both

microglial activation and neuronal excitability, highlighting the interplay between lipid metabolism, inflammation, and neuronal function. It remains unclear how microglial inflammatory state is conveyed to neurons to affect circuit function and whether *APOE4* expression alters this intercellular communication. Here, we use a reductionist model of human iPSC-derived microglial and neuronal monocultures to dissect how *APOE4* genotype in each cell-type independently contributes to microglial regulation of neuronal activity during inflammation. Conditioned media (CM) from LPS-stimulated microglia increases spontaneous neuronal network activity, assessed by calcium imaging, with *APOE4* microglial CM driving higher neuronal firing rates than *APOE3* CM. Both *APOE3* and *APOE4* neurons show similar increases in network activity in response to CM treatments, but *APOE4* neurons uniquely increase presynaptic puncta with *APOE4* microglial CM. CM-derived exosomes from LPS-stimulated *APOE3* microglia recapitulate increases in spontaneous network activity. Lastly, increased network activity is accompanied by increased lipid droplet (LD) metabolism, and blocking LD metabolism abolishes network activity. These findings illuminate how microglia-to-neuron communication drives inflammation-induced changes in neuronal circuit function, demonstrate a role for neuronal LDs in network activity, and support a potential mechanism through which *APOE4* increases neuronal excitability.

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Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.04/D2

Topic: B.09. Glial Mechanisms

Support: NIH Grant R01AG083943
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Title: Bioengineering 3D hybrid cortical microtissues to model human neuron-microglial interactions in Alzheimer's disease

Authors: *Y.-W. HUANG;
Mol. Biology, Cell Biol. and Biochem., Brown Univ., Providence, RI

Abstract: Background: Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by synaptic dysfunction and neuronal loss, with growing evidence that glial cells—particularly microglia—play central roles in disease progression. Existing rodent models and 2D in vitro systems often fail to replicate the complex, human-specific interactions among neurons and glia. Therefore, improved model systems are needed to investigate how an AD-mutant microenvironment influences neuron-glial interactions. **Method:** We developed three-dimensional (3D) primary cortical microtissues derived from wild-type or AD-mutant rat cortices, which recapitulate hallmark AD pathologies. Into these microtissues, we engrafted

human induced microglia (iMG) and neurons(iN) derived from control or AD-relevant iPSC lines. Through longitudinal live imaging of calcium activities, immunohistochemistry, and molecular analyses (e.g., ELISA, transcriptomics), we characterized the effects of the AD-mutant microenvironment on human iMG and iN activity, morphology, and gene expression. **Result:** Our AD-mutant cortical microtissues exhibited accelerated amyloid-beta accumulation, pro-inflammatory signatures, synaptic dysfunction and neuronal loss. Engrafted iMG demonstrated robust engraftment, responding to the AD microenvironment with increased markers of activation and phagocytosis. Co-engrafted iNs displayed synaptic deficits and altered calcium dynamics, implicating microglia-neuron interactions in disease-relevant phenotypes. These findings highlight the influence of an AD-mutant environment on human microglial and neuronal function within a physiologically relevant 3D context. **Conclusion:** By successfully establishing AD-mutant 3D microtissues and engrafting human iMG alongside iNs, we have created a powerful ex vivo model that captures key aspects of human neuron-glial interactions in AD. This suite of new model systems provides a valuable platform for dissecting the molecular and cellular mechanisms underlying AD pathogenesis and offers significant potential for therapeutic discovery.

Disclosures: **Y. Huang:** A. Employment/Salary (full or part-time): Brown University. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Acre Therapeutics LLC.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.05/D3

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Investigating the intersection of estrogen signaling, m6A RNA modification, and microglial function

Authors: *Y. L. PEÑA¹, Z. ZHANG³, S. KO², S. C. HOPP⁴, L. CHEN⁵;

²Cell Systems & Anatomy, UT health SA, ¹Univ. of Texas Hlth. Sci. Ctr. at San Antonio, San Antonio, TX; ³Mol. Med., ⁴Biggs Inst. for Alzheimer's and Neurodegenerative Dis., UT Hlth. San Antonio, San Antonio, TX; ⁵Barshop Inst., Univ. of Texas Hlth. Sci. Center, San Antonio, San Antonio, TX

Abstract: Alzheimer's disease (AD) is a neurodegenerative disease, and its progression has been linked to chronic neuroinflammation. This neuroinflammatory response in the disease has been widely studied and it is shown to be partially mediated by microglial responses. However, estrogens are known to shift microglial activation toward a neuroprotective state, allowing to attenuate inflammatory responses, such as those manifested in AD (Villa et al., 2016). According to recent research, the RNA modification N6-methyladenosine (m6A) has been seen to regulate transcript stability and, therefore, promote gene expression in inflammatory signaling. For

example, m6A methylation writer enzyme called N6-adenosine-methyltransferase 3 (METTL3) has been observed to help in stabilizing pro-inflammatory mRNAs in microglia, such as BATF (Wu et al., 2025). Furthermore, m6A-marked enhancer RNAs had been shown to facilitate the formation of transcriptional condensates, which in turn, enhance gene activation via phase separation (Lee et al., 2021). On the other hand, the estrogen receptor alpha (ER α) has been shown to form dynamic phase-separated condensates which also help in recruiting transcriptional coactivators and promoting estrogen-mediated gene expression (Hsia et al., 2019). In this work, we explore the intersection of estrogen signaling and m6A-regulated condensate formation in the human microglial HMC3 cell line. We have performed single-nucleus RNA-sequencing using 5xFAD mouse brains and our results show that ER α 's expression is enriched in microglia, supporting its relevance in AD pathology. Furthermore, our data indicates that using estradiol (E2) treatment promotes formation of ER α condensates in HCM3, which could incentivize distinct gene expression compared to non-treated microglia. Moreover, pharmacological inhibition of METTL3 through the catalytic inhibitor STM2457, has been observed to disrupt ER α condensate formation in our preliminary data. These findings suggest that estrogen can promote ER α condensate formation through m6A-dependent mechanisms which might be involved in microglial inflammatory modulation. Future work will focus on establishing how m6A modifications mediate estrogen-induced transcriptional and functional responses in microglia, especially focusing on ER α condensate dynamics and how these alter inflammatory gene expression. Our work underlines a novel axis linking m6A-mediated estrogen signaling and microglial function in hopes to establish a novel approach for sex-specific therapies in AD.

Disclosures: Y.L. Peña: None. Z. Zhang: None. S. Ko: None. S.C. Hopp: None. L. Chen: None.

Poster

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.06/D4

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Kine-pd-28, an ultra-short peptide, rescues cognitive and synaptic deficits through microglial immune activation and attenuation of amyloid pathology and neuroinflammation.

Authors: *H.-L. CHA;

Dept. of Neurolog. disease, Kine Sciences. Ltd, Seongnam-si / Gyeonggi-do, Korea, Republic of

Abstract: Alzheimer's disease (AD) is the most prevalent form of dementia, characterized by the pathological accumulation of amyloid-beta (A β) and hyperphosphorylated tau proteins. Despite decades of research, effective disease-modifying therapies (DMTs) remain limited due to the heterogenetic etiology and complex interplay between molecular pathology and immune dysregulation. We present KINE-PD-28, a novel nonameric peptide that modulates microglial immunity, as a promising therapeutic candidate for AD. KINE-PD-28 suppresses disease-

associated microglia (DAM, or M1-like) that are causing notorious neuroinflammation in AD and promotes anti-inflammatory (M2-like) microglial phenotypes. In the meantime, KINE-PD-28 reduced pathological A β accumulation through enhancing the phagocytic function of microglia. Consequently, we found that KINE-PD-28 improved the synaptic dysfunction in the hippocampus and mitigated cognitive impairment in 5xFAD mice at 6 months of age. Collectively, our findings endorse KINE-PD-28 as a first-in-class peptide-based DMT candidate that modulates microglial multifunctional immunity in AD, thereby ameliorating cognitive impairment. KINE-PD-28 is poised to fulfill critical unmet needs in current AD therapy.

Disclosures: H. Cha: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.07/D5

Topic: C.02. Alzheimer's Disease and Other Dementias

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JST Moonshot R&D JPMJMS2024

Title: Inpp5d deficiency enhances microglial neuroprotective function in Alzheimer disease by inhibiting Amyloid- β plaque maturation and toxicity

Authors: *A. AKAHORI¹, S. TAKATORI¹, Y.-N. CHU¹, J. SASAKI², T. SASAKI², T. TOMITA¹;

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Abstract: In Alzheimer disease (AD) pathogenesis, microglial clustering around amyloid- β (A β) deposition demonstrates neuroprotective functions against surrounding dystrophic neurites. The microglial receptor TREM2 is essential for this response, but the downstream signaling pathways remain incompletely defined. Our previous work identified INPP5D, an AD risk gene, as a negative regulator of TREM2-downstream PIP₃ signaling (Iguchi et al., *iScience* 2023). In the current study, we investigate the role of INPP5D in microglial neuroprotective functions using male microglia-specific *Inpp5d* knockout mice (*App*^{NL-G-F/NL-G-F}; *Inpp5d*^{fllox/fllox}; *Cx3cr1*^{Cre}). Comprehensive immunohistochemical analyses revealed that *Inpp5d* deficiency significantly enhanced microglial clustering around A β plaques at 4 months and markedly reduced dystrophic

neurites at 9 months. While total A β burden remained unchanged, we observed significant reductions in thioflavin-positive plaques and N3pE-A β accumulation (a donanemab target). This suggests that INPP5D negatively regulates neuroprotective functions by promoting plaque maturation and toxicity rather than affecting overall A β burden. To investigate underlying mechanisms, we further analyzed the microglial gene expression and microglial-A β interactions in 9-month-old mice. Immunohistochemistry revealed attenuated expression of disease-associated microglia (DAM) markers, such as CD11c and Galectin-3, in the plaque-associated microglia, indicating altered microglial phenotypes. Additionally, three-dimensional electron microscopy demonstrated altered physical interactions between A β , microglia, and neurons in *Inpp5d*-deficient mice, with reduced dystrophic neurites at the ultrastructural level. In conclusion, INPP5D regulates the microglial-A β interaction patterns and microglial phenotypes, attenuating neuroprotective functions through promoting A β plaque maturation. These findings suggest that INPP5D inhibitors represent promising candidates for novel microglia-targeted AD therapeutics that could alter disease progression by enhancing neuroprotective functions of microglia.

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Poster

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Title: Microglia morphology and phagocytic activation in visual brain regions of the 5xfad mouse

Authors: *S. MCCOOL, J. C. SMITH, M. J. VAN HOOK;
Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: Vision changes are some of the first reported symptoms in Alzheimer's disease (AD), a neurodegenerative disorder characterized in part by the formation of amyloid beta (A β) plaques leading to cognitive deficits, brain atrophy, and decreased quality of life. Microglia, responsible for maintaining homeostasis and responding to disease pathology, are no longer able to keep up with the A β plaque load found in AD patients. Due to the increasing number of individuals

diagnosed with AD and its early impact on vision, we aim to provide an in-depth analysis of microglia in several visual regions of the brain. The regions of interest include the dorsal lateral geniculate nucleus (dLGN), a retinorecipient thalamic relay, the primary visual cortex (V1) which receives input from dLGN, and two additional retinorecipient regions: superior colliculus (SC), important for sensory-motor integration, and suprachiasmatic nucleus (SCN), which regulates circadian rhythms. To study A β pathology and microglial responses in these regions, we used the 5xFAD mouse model which develops A β plaques throughout the brain beginning around 2 months (mo). We used male and female 5xFAD mice at 6, 9, and 12 mo with age-matched C57 mice as controls. Tissue sections containing dLGN, SC, SCN, or V1 were stained for A β plaques using thioflavin s. We found a high density of plaques in 5xFAD dLGN, yet did not find plaques in 5xFAD SCN or visual layer of the SC. We found a significant density of plaques in 5xFAD V1, but significantly lower than in dLGN. We stained for Iba1, a marker of microglia morphology, to perform a skeleton analysis on 6, 9, and 12 mo dLGN as well as 9 mo dLGN, SCN, SC, and V1. This analysis showed decreased branch length and endpoints per microglia in 5xFAD dLGN at all ages indicative of microglia reactivity. We saw decreased branch length but not endpoints in 9 mo V1 indicating that V1 is trending toward amoeboid morphology with no change detected in SC or SCN. We co-stained with CD68, a lysosomal marker, which allowed us to correlate microglia morphology with its function. Calculating percent of CD68 colocalized with Iba1 allowed us to determine changes in microglial phagocytic activity which revealed a significant colocalization of Iba1 and CD68 in 6, 9, and 12 mo 5xFAD dLGN as well as significant colocalization in 9 mo V1 but not SC or SCN. In regions with plaques, we found a significant change in microglia morphology toward an amoeboid state as well as significant colocalization of Iba1 and CD68 indicating phagocytic activity in plaque-infested areas. These results suggest amyloid pathology drives microglia activation and phagocytic activity in only a subset of visual areas of the 5xFAD brain.

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Poster

PSTR265: Microglia in Alzheimer's Disease

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Title: Microglia States are Susceptible to Senescence and Cholesterol Dysregulation in Alzheimer's Disease

Authors: *B. LI¹, S. WANG², B. E. KERMAN³, C. HUGO⁴, H. N. YASSINE⁴;

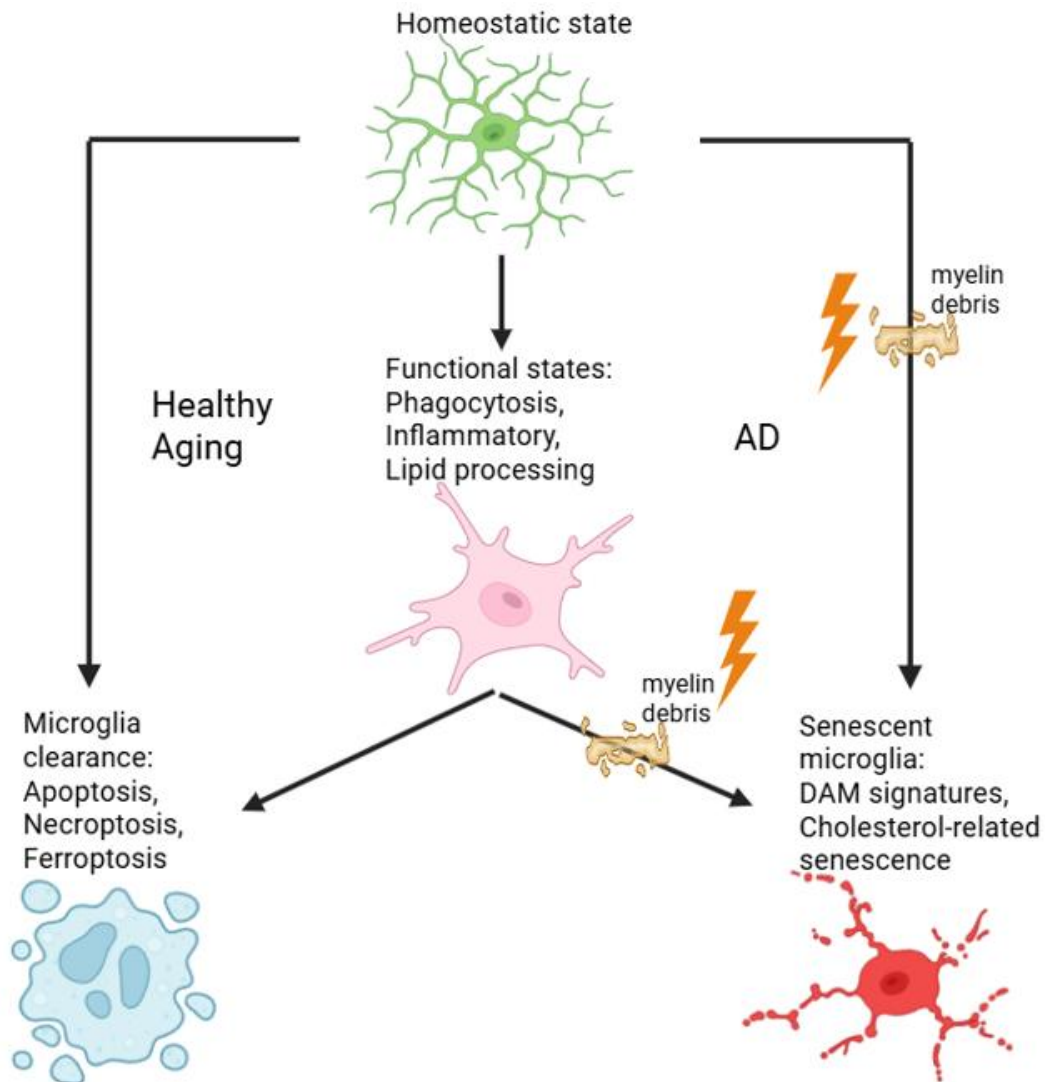
¹Keck Sch. of Med. of USC, Los Angeles, CA; ²Keck Sch. of Med., ³REMER, ⁴USC, Los Angeles, CA

Abstract: Cellular senescence is a key contributor to aging-related neurodegenerative diseases, including Alzheimer's disease (AD). However, the specific brain cell types and molecular pathways driving senescence remain poorly defined. Here, we hypothesize that dysregulated cholesterol metabolism drives microglial senescence in AD. To test this, we leveraged single-nucleus RNA sequencing (snRNA-seq) data from both the ROSMAP and Sea-AD studies to dissect cell-type-specific senescence mechanisms in the AD human brain.

Our analysis of ROSMAP data (982,384 nuclei from postmortem prefrontal cortex) revealed that microglia are primary contributors to AD-associated senescence among non-neuronal cell types. We identified multiple microglial states—including homeostatic, inflammatory, phagocytic, lipid-processing, and neuronal surveillance phenotypes—that are vulnerable to senescence in both ROSMAP (152,459 microglial nuclei across six brain regions) and Sea-AD (82,486 microglial nuclei). Integrative pathway analysis showed that senescent microglia exhibit profound alterations in cholesterol-related biological processes.

We further identified three cholesterol-associated gene co-expression modules that define a senescence-linked transcriptional program in AD brains. To validate these findings functionally, we assessed these transcriptomic signatures in iPSC-derived microglia (iMGs) exposed to myelin. Myelin exposure recapitulated the postmortem brain cholesterol-senescence signatures in iMGs, and treatment with myelin debris or oxidized cholesterol induced a disease-associated microglia (DAM)-like state marked by cholesterol-driven senescence.

Together, these results provide the first human evidence linking cholesterol dysregulation to microglial senescence in AD. Our findings highlight cholesterol metabolism in microglia as a potential therapeutic target to mitigate cellular senescence and slow AD progression.



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Poster

PSTR265: Microglia in Alzheimer's Disease

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.10/D8

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Investigating the role of APOE3 Christchurch (APOE3ch) mutation in modulating microglial function in Alzheimer's disease

Authors: *A. SINGH^{1,2}, Y. ZHANG⁴, S. H. YUAN^{5,3};

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⁴Minnesota Supercomputing Inst., ⁵Neurol., Univ. of Minnesota, Minneapolis, MN

Abstract: The APOE3 Christchurch (APOE3ch) mutation has drawn recent attention for its potential protective effect against Alzheimer's disease (AD), yet its impact on microglial function remains unclear. To explore this, we generated microglia from isogenic patient-derived induced pluripotent stem cells (iPSCs) with APOE3ch mutation by CRISPR/Cas9 editing. Microglial identity was confirmed by qPCR and immunocytochemistry for IBA1. Functional assays assessing inflammation and lipid droplet accumulation were performed. Transcriptomic profiling was conducted using bulk RNA sequencing. All edited microglial lines expressed IBA1. IL6 expression was elevated across all groups from baseline among APOE3ch, PSEN1, and PSEN1-corrected microglia. Notably, APOE3ch and PSEN1-corrected microglia exhibited significantly reduced lipid accumulation compared to PSEN1 mutant microglia. Transcriptomic analysis revealed variation in expression of phagocytosis and DAM -related genes in APOE3ch microglia. These findings suggest that APOE3ch may modulate microglial lipid metabolism and AD risk gene expression, contributing to its proposed neuroprotective effects. Further studies are warranted to validate these findings across additional cell lines and investigate APOE3ch-targeted therapeutic strategies.

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Poster

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Title: Transcriptional regulatory mechanisms underlying IL-33-stimulated microglial state transition in Alzheimer's disease

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Abstract: Microglia play a crucial role in maintaining brain homeostasis by responding to damage-associated molecular patterns such as the pathological hallmark of Alzheimer's disease (AD), amyloid-beta (A β). In response to A β , microglia first transit to chemotactic state, which allows them to migrate toward A β depositions, and then further transit to a phagocytic state for effective clearance of A β . However, as pathological conditions of AD progress, the chemotactic and phagocytic abilities of microglia are compromised. This impairment leads to the accumulation of A β aggregates and thus exacerbates disease progression. We previously demonstrated that interleukin-33 (IL-33) treatment in transgenic mouse models of AD ameliorates AD-related pathologies, including decrease of A β plaque deposition and neuroinflammation. Single-cell transcriptome profiling revealed that microglia undergo a stepwise state transition upon IL-33 treatment. However, the underlying regulatory mechanisms remain largely unclear. Here, we conducted transcription and proteomic analyses in an IL-33-treated murine microglial cell line and identified NF- κ B transcription factor family members and other transcription factors as potential key regulators of the microglial state transition. Notably, NF- κ B siRNA knockdown in mouse microglial BV2 cells significantly downregulated chemotactic microglial markers induced by IL-33, indicating that NF- κ B is essential for regulating microglial transcriptomic reprogramming during the cellular state transition of microglia. Thus, understanding the transcriptional regulations of microglial subpopulations and the precise control over these regulatory mechanisms holds valuable insights into the pathological basis of AD.

Disclosures: W. Wu: None. Y. Qiao: None. S. Lau: None. X. wang: None. Y. Bao: None. Y. Duan: None. A. Wong: None. X. Yang: None. W. Fu: None. A. Fu: None. N.Y. Ip: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.12/D10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: the Research Grants Council of Hong Kong (the Collaborative Research Fund [C6027-19GF], the Theme-Based Research Scheme [T13-605/18W] and the General Research Fund [HKUST16103122 and HKUST16102824])
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the Areas of Excellence Scheme of the University Grants Committee (AoE/M-604/16)

Title: Regulatory Roles of Soluble ST2 in Microglial State Transition and Functions in Alzheimer's Disease

Authors: *Y. QIAO^{1,2}, W. WU^{1,2}, X. WANG^{1,2}, A. WONG^{1,2}, Y. DUAN^{1,2}, Y. BAO^{1,2}, X. YANG^{1,2}, S. GILL^{1,2}, L. SHI³, W.-Y. FU^{1,2}, A.-Y. FU^{1,2,4}, N. Y. IP^{1,2,4};
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Abstract: The microglial immune response plays a key role in the pathogenesis of Alzheimer's disease (AD). In response to damage-associated molecular patterns, such as the pathological hallmark, beta-amyloid (A β), microglia undergo reprogramming into various molecular and functional cell states that enable them to restrict the pathology development. However, as AD progresses, the accumulation of pathological features impedes the beneficial transition of microglia to clear A β . We previously demonstrated that interleukin-33 (IL-33)/ST2 signaling is important for A β clearance in AD. This signaling is dysregulated in patients with AD due to increased levels of soluble ST2 (sST2), the decoy receptor of IL-33. To investigate whether and how sST2 regulates the microglial state transition and function in AD, we examined the molecular changes of microglia in an amyloidosis transgenic mouse model, 5xFAD mice, upon deletion of sST2. Single-cell RNA sequencing revealed that deletion of sST2 enhanced the microglial state transition from a homeostatic state to a chemotactic state in 5xFAD mice, indicating the microglia are more responsive to A β . Furthermore, transcriptome analysis of chemotactic microglia in these transgenic mice showed an increased expression of genes associated with cytoplasmic translation, providing protein synthesis machinery for microglial state transition and beneficial functions. Moreover, IL-33-treated amyloidosis transgenic mouse models also showed similar transcriptomic changes in chemotactic microglial subpopulation, suggesting that the endogenous IL-33/ST2 signaling contributes to the cell state transition of microglia. These findings highlight the important role of endogenous IL-33/ST2 signaling in the regulation of cellular state transition of microglia and their beneficial functions in AD.

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Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.13/D11

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01AG075897
BrightFocus Foundation Award A2021036S

Title: Mighty Morphin" Microglia: Neonatal Estradiol Exposure Induces Sex-Specific Changes to Microglia in 5xFAD Mice

Authors: *P. R. KELLER¹, B. T. CASALI², E. G. REED²;
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Abstract: Microglia, the brain's resident immune cells, participate in neurodevelopmental processes such as growth cone guidance and synaptic pruning. These processes occur in a sex-specific manner, as microglia are sensitive to early gonadal hormone pulses, and serve as key mediators of brain masculinization. Male microglia in early development favor a short-lived pro-inflammatory phenotype, contrasting with female microglia, which favor an anti-inflammatory phenotype until adolescence. Once matured, female microglia exhibit persistent pro-inflammatory gene expression throughout adulthood and late life, which may play a role in the female bias of neurodegenerative diseases, such as Alzheimer's Disease (AD). The current study utilized a masculinization model driven by neonatal estradiol administration (during the critical period, postnatal days 2-8) to assess the impact of masculinization on disease state outcomes in the 5xFAD model of AD. We observed modest transcriptomic and cytoarchitectural changes in response to E2 treatment in juvenile 5xFAD mice, assessed via qPCR and immunohistochemistry, with notable changes to the expression of Xist, a marker of X chromosome inactivation. These changes indicate that the paradigm was sufficient to drive masculinization and that the effects of E2 masculinization are global in nature. We will further assess E2-driven changes to pathological outcomes in aged mice, assessed via immunohistochemistry, qPCR, and ProteinSimple Jess automated western blotting.

Disclosures: P.R. Keller: None. B.T. Casali: None. E.G. Reed: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.14/D12

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01AG056287
T32 AI007290

Title: Spatial Proteomics Reveals Alzheimer's Disease-Specific Human Microglial States

Authors: *B. CANNON;
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Abstract: Microglia contribute to aging, neurodegeneration, and Alzheimer's disease (AD), but their diversity and interactions within the human brain remain poorly characterized due to limitations of low-plex protein imaging and differences from rodent models. Using Multiplexed Ion Beam Imaging (MIBI), we spatially mapped cellular states and microenvironments in cognitively normal (CN) human brains, identifying a spectrum of proteomic microglial profiles across ten individuals. Single-nucleus assay for transposase-accessible chromatin sequencing (snATAC-seq) was used in parallel to validate molecular functions associated with these microglial states. Microglial immune activation states varied across brain regions and microenvironments, revealing proteomic trends among CN individuals. In AD tissues, microglia exhibited significant shifts at the immunologically active end of the proteomic spectrum, including increased expression of CD33 and CD44 and decreased expression of HLA-DR, P2RY12, and ApoE. These findings establish an in situ, single-cell spatial proteomic framework that defines Alzheimer's disease-specific microglial states and regulatory alterations.

Disclosures: B. Cannon: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.15/D13

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: National Research Foundation of Korea(NRF) grant funded by the Korea government (MSIT) RS-2024-00335208
National Research Foundation of Korea(NRF) grant funded by the Korea government (MSIT) RS-2024-00509145

Title: Muscle-derived exosomes maintain microglial homeostasis and attenuate Alzheimer's pathology

Authors: *S. KIM¹, K.-F. LEE², J. LEE¹;

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Abstract: In Alzheimer's disease (AD), microglia exhibit dysfunction in clearing pathological proteins, due to their chronically activated inflammatory responses, which exacerbates neuroinflammation and accelerates disease progression. Skeletal muscle has been recognized as a tissue which is capable of modulating brain inflammation through the secretion of anti-inflammatory cytokines. Recent meta-analysis revealed a significant association between sarcopenia—the age-related loss of skeletal muscle mass—and AD. However, the underlying mechanism linking sarcopenia and AD remains poorly understood. In this study, we observed an increase in disease-associated microglia (DAM), a key pathological hallmark of AD, in a mouse model of sarcopenia. Sarcopenia may be induced by impaired differentiation of myogenic progenitor cells into myocytes. During this differentiation process, myocytes secrete exosomes to mediate intercellular communication. We found that myocyte-derived exosomes are taken up by microglial cells via endocytosis and suppress inflammatory responses. Currently, we are investigating the molecular mechanisms by which these ectosomes regulate the anti-inflammatory response in microglia. Furthermore, to investigate their physiological impact in a human-related system, we established a muscle-spinal cord-brain assembloid. This assembloid enables the assessment of microglial anti-inflammatory responses induced by myocyte-derived ectosomes. Collectively, our data suggest that sarcopenia leads to impaired microglial anti-inflammatory regulation, thereby promoting the progression of Alzheimer's disease. These findings highlight the skeletal muscle as a novel therapeutic target for modulating neuroinflammation in AD and provide new insight into peripheral-to-central signaling pathways in neurodegeneration.

Disclosures: S. kim: None. K. Lee: None. J. Lee: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.16/D14

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Investigating the Structural and Functional Implications of TREM2 and IL-34 Interactions in Alzheimer's Disease

Authors: *J. WYDRA¹, J. GREVEN¹, T. BRETT², J. ALEXANDER-BRETT³;

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Abstract: TREM2, an extracellular receptor prominently expressed on microglia, plays a critical role in modulating neuroinflammation, a hallmark of Alzheimer's disease (AD) and other neurodegenerative disorders. Microglia, the brain's resident immune cells, contribute

significantly to neuronal homeostasis and inflammatory responses. Dysregulated microglial activation is strongly associated with AD pathogenesis. Recent evidence has identified IL-34, a cytokine involved in microglial proliferation, differentiation, and survival, as a novel and biologically relevant ligand for TREM2. Using comprehensive biolayer interferometry (BLI), we characterized binding interactions of over 20 different TREM2 protein variants and newly synthesized IL-34 variants, identifying a critical binding interface centered on residue R76 of TREM2. Mutations at this residue substantially diminished affinity for IL-34, which allude to its critical role in the receptor-ligand interaction. Additionally, mass photometry analyses demonstrated that the TREM2/IL-34 complex forms a stable 2:2 complex (MW = 161 kDa), suitable for structural determination by cryo-electron microscopy (cryo-EM). Furthermore, to better investigate functional consequences of TREM2/IL-34 interactions and their relevance to neurodegenerative disease pathology, we employed BV2 microglial cells. These assays use BV2 cells to assess the impact of nine distinct anti-TREM2 monoclonal antibodies, revealing differential modulation of microglial activation and inflammatory signaling. Collectively, our findings demonstrate that specific structural interactions between TREM2 and IL-34 significantly influence microglial inflammatory pathways implicated in Alzheimer's pathology. This structural and functional characterization of TREM2/IL-34 interaction offers valuable insights for the targeted design of novel therapeutics aimed at modulating microglial activity, potentially attenuating neuroinflammation, and slowing progression in Alzheimer's disease and related neurodegenerative disorders.

Disclosures: J. Wydra: None. J. Greven: None. T. Brett: None. J. Alexander-Brett: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.17/D15

Topic: B.09. Glial Mechanisms

Support: NIH/NIA Grant T32AG000222
JPB Foundation
Cure Alzheimer's Fund

Title: Inhibition of Acyl-CoA:Cholesterol Acyltransferase 1 (ACAT1) promotes shedding of soluble TREM2 and LRP1-dependent phagocytosis of amyloid b in microglia

Authors: *M. HOVDE¹, A. MAASER-HECKER^{1,2}, R. E. TANZI³;
²Neurol., ¹Massachusetts Gen. Hospital/Harvard Med. Sch., Boston, MA; ³Massachusetts Gen Hosp, Harvard Med. Sch., Charlestown, MA

Abstract: Alzheimer's disease (AD) is a neurodegenerative disease with hallmarks that include accumulation of plaques containing mostly amyloid-beta (A β) peptides, neurofibrillary tangles composed of hyperphosphorylated Tau, and chronic neuroinflammation. In addition to

commonly known hallmarks of AD, increased lipid droplets in glial cells were originally noted by Alois Alzheimer while more recent studies have shown elevated levels of cholesteryl esters (CE), a major component of lipid droplets, in vulnerable brain regions of AD patients. Formation of CE occurs in the ER by Acyl-CoA: Cholesterol Acyltransferase 1 (ACAT1) which esterifies cholesterol for lipid droplet storage. Previous work from our lab shows ACAT1 inhibition in neurons reduces APP processing, A β production, and plaque load in AD mouse models. Moreover, others have shown ACAT1 inhibition in microglia increases the capacity for internalization and phagocytosis of A β and Tau. While these studies underscore the therapeutic potential of ACAT1 inhibitors in mitigating AD pathology, the mechanisms underlying these beneficial effects remain unclear. Here we show Triggering Receptor in Myeloid Cells 2 (TREM2) and Low-Density Lipoprotein-like Receptor 1 (LRP1) play a central role in the mechanism by which ACAT1 inhibition increases A β uptake by microglial cells. Specifically, ACAT1 inhibition upregulates LRP1 levels and promotes the release of soluble TREM2 (sTREM2) via enhanced cleavage of TREM2 by ADAM10/17. Knocking out TREM2 or preventing TREM2 cleavage abolishes enhanced phagocytosis of A β induced by ACAT1 inhibition. This could be rescued by the addition of recombinant sTREM2, but only in the presence of LRP1, strongly suggesting LRP1 is a receptor for the sTREM2-A β complex. Collectively, these findings reveal a previously unrecognized mechanism by which ACAT1 inhibition enhances microglial A β clearance suggesting that small molecule ACAT1 inhibitors may offer a promising therapeutic avenue for AD and as TREM2 modulators.

Disclosures: M. Hovde: None. A. Maaser-Hecker: None. R.E. Tanzi: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.18/D16

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH U54 AG065181
NIH U54 AG065187

Title: Generation and Characterization of iPSC Derived Microglia for Investigation of Alzheimer's Disease Therapeutic Molecules

Authors: *J. A. KULAS;
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Abstract: In this study, we differentiated iPSC derived microglia-like cells (iMG) using a newly generated iPSC cell line termed IBRI 104.G. We performed extensive characterization of both the parent iPSC line and iMG. We find that iMG express a number of core microglial marker genes including P2RY12, TREM2 and HEXB. iMG are morphologically plastic and retain a ramified morphology under control conditions and rapidly adopt an ameboid morphology during

stimulation with inflammatory stimuli. iMG responses to physiologic stimuli including myelin resulted in highly specific transcriptional signatures and metabolic changes without amoeboid cell morphologies. iMG responded to cytokine stimulation by upregulating TREM2 signaling molecules and broadly shifting transcription of IRFs. Finally, we tested a number of tool molecules including TREM2 agonists and amyloid antibodies for their effects on iMG and observed both transcriptional and morphology changes. We conclude IBRI 104.G iMG are a useful new model system for the interrogation of Alzheimer's disease therapeutic molecules seeking to modulate myeloid cell phenotypes.

Disclosures: J.A. Kulas: None.

Poster

PSTR265: Microglia in Alzheimer's Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR265.19/D17

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: National Institutes of Health AG062734
Hope Center for Neurological Disorders
Chan Zuckerberg Initiative
Thome Memorial Foundation
UL1tR002345
NIH P30AG066444
P01AG03991

Title: Ms4a4a deficiency ameliorates plaque pathology in a mouse model of amyloid accumulation

Authors: *A. S. DÍAZ-PACHECO¹, A. VERBECK², J. R. CIRRITO³;

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Abstract: Genome-wide association studies have identified over 70 Alzheimer's disease (AD) risk loci, many of which are enriched in microglia. Common variants in the microglia-enriched gene MS4A4A influence AD risk, MS4A4A expression, TREM2 signaling, and a distinct microglial transcriptional state, yet the precise role of MS4A4A in AD pathogenesis remains unclear. To assess the functional impact of MS4A4A loss, we generated Ms4a4a-deficient 5xFAD mice (5xFAD Ms4a4a WT n = 6; 5xFAD Ms4a4a KO n = 7) using CRISPR/Cas9. Utilizing in vivo microdialysis, we observed that Ms4a4a deficiency significantly reduced basal amyloid-beta (A β) concentration and increased A β half-life in brain interstitial fluid (ISF) prior to plaque accumulation. Plaque burden and compaction were assessed using HJ3.4 and X-34 immunostaining. Aged 6-month-old 5xFAD Ms4a4a KO mice exhibited more compact plaques and an approximate 55% reduction in overall plaque burden in the hippocampus at 6 months.

While Ms4a4a loss did not affect overall astrogliosis or microgliosis, immunoblotting in aged mice revealed that in the absence of Ms4a4a, microglia-enriched A β -degrading enzyme, MMP-9, was significantly elevated. Supporting these results, an analysis of a cohort of 3,506 individuals with genetic and cerebrospinal fluid (CSF) proteomic data revealed that carriers of an AD-protective variant near MS4A4A (rs1582763) have exhibited significantly elevated CSF MMP-9 levels, whereas carriers of an AD-risk variant (rs6591562) have exhibited reduced CSF MMP-9 levels. This work highlights the therapeutic potential of targeting MS4A4A and modulating MMP-9 expression to promote plaque clearance and mitigate both amyloid deposition and the broader development of AD pathologies.

Disclosures: A.S. Díaz-Pacheco: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.01/D18

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Health and Medical Research Fund (Hong Kong)
CUHK direct grant scheme
United College endowment fund

Title: Roles of Amyloid precursor protein interactors in regulating autophagy in Alzheimer's disease

Authors: *K.-F. LAU;
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Abstract: Autophagy, a conserved catabolic process, removes dysfunctional cellular components and is implicated in neurodegenerative diseases like Alzheimer's disease (AD). Dysregulation of autophagy contributes to AD pathology, such as amyloid precursor protein (APP) metabolism, making its molecular mechanisms critical for therapeutic development. Emerging evidence suggests that APP interactors, including the adaptor proteins GULP1 and FE65, have roles in autophagy. We identified that GULP1 interacts with autophagy-related 14 (ATG14), a regulator of autophagosome formation, enhancing class III phosphatidylinositol 3-kinase complex 1 (PI3KC3-C1) activity. This interaction facilitates ATG14 targeting to the endoplasmic reticulum, promoting autophagy and APP processing. A GULP1 mutation disrupting the GULP1-ATG14 interaction attenuates these effects, while APP expression enhances PI3KC3-C1 activity via GULP1. Similarly, FE65 interacts with Beclin 1, a key PI3KC3-C1 component, to positively regulate autophagy. Overexpression of FE65 enhances Beclin 1-mediated autophagy, whereas FE65 knockout or a C-terminus deletion mutant diminishes this effect. Both GULP1 and FE65 modulate PI3KC3-C1 kinase activity, influencing autophagosome biogenesis and APP processing. Notably, GULP1 increases ATG14 and APP

levels in autophagic vacuoles, enhancing APP processing, while FE65 modulates Beclin 1 to similar ends. These findings highlight GULP1 and FE65 as novel regulators of autophagy through distinct interactions with ATG14 and Beclin 1, respectively, offering insights into their roles in AD pathology and potential therapeutic targets for modulating autophagy and APP processing.

Disclosures: K. Lau: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.02/D19

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: iPSC-derived Alzheimer's disease models show increased secretion of pathogenic amyloid beta peptides in glutamatergic neurons and reduced inflammatory responses in microglia

Authors: A. VETLEANU¹, P. BARTON¹, S. MILDE¹, M. GAMPERL², S. SALIC-HAINZL², G. BELLIVALLETTA¹, G. SHIPLEY¹, H. GARNETT¹, C. ARUTHAN¹, T. BUERCKSTUEMMER², O. DOVEY¹, K. FIRTH¹, *A. TURNER¹, **B. HAUGEN**¹, T. OOSTERVEEN¹, W. BERNARD¹;

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Abstract: Alzheimer's disease (AD), a complex, multifactorial neurodegenerative disease, is challenging to study in vitro due to a lack of physiologically relevant models. Using deterministic cell programming technology, opti-ox, we generated iPSC-derived excitatory neurons (ioGlutamatergic Neurons) and microglia (ioMicroglia) and a panel of genetically matched disease models with AD-related mutations in APP or PSEN1 in neurons, and the APOE 4/4 genotype in microglia, and investigated the effects of the mutations on the amyloid beta (A-beta) production and inflammatory responses respectively.

CRISPR/Cas9 was used to engineer heterozygous and homozygous PSEN1 M146L, APP KM670/671NL or APP V717I mutations in the parental iPSC line of the ioGlutamatergic Neurons, which were subsequently programmed using opti-ox technology to generate the disease model cells. The disease models were cultured for 30 days alongside their genetically matched wild type control. Supernatant was collected on days 10, 20 and 30, and concentrations of A-beta38, A-beta40 and A-beta42 were determined using the V-PLEX A-beta Peptide Panel ELISA kit.

ioGlutamatergic Neurons carrying the PSEN1 M146L and APP V717I mutations secreted significantly more A-beta42 compared to their wild type control, showing higher A-beta42:40 ratios at days 20 and 30. Importantly, a clear correlation between genotype and A-beta42:40 ratios was observed, as wild type, heterozygous and homozygous mutants showed a stepwise increase in A-beta42 production relative to A-beta40. ioGlutamatergic Neurons APP KM670/671NL secreted significantly more A-beta38, A-beta40, and A-beta42 than their wild

type control but the A-beta42:40 ratio did not increase, as expected. ioGlutamatergic Neurons with mutations in PSEN1 or APP recapitulate the increase in A-beta42 secretion observed in Alzheimer's patients. This demonstrates their validity as an in vitro model to study AD and for the discovery of drugs targeting the pathogenic A-beta pathway.

CRISPR/Cas9 was also used to generate ioMicroglia with the APOE 4/4 genotype, which significantly increases the risk of developing AD. These cells show a reduction in phagocytosis and reduced secretion of pro-inflammatory cytokines on activation compared to their genetically matched parental wild type cells.

Finally, we developed a co-culture protocol to combine wild type and disease model ioGlutamatergic Neurons and ioMicroglia, to build more physiological in vitro models to mimic and disentangle the complex cellular interactions during AD.

Disclosures: **A. Veteleanu:** A. Employment/Salary (full or part-time): bit.bio. **P. Barton:** A. Employment/Salary (full or part-time): bit.bio. **S. Milde:** A. Employment/Salary (full or part-time): bit.bio. **M. Gamperl:** A. Employment/Salary (full or part-time): bit.bio Discovery. **S. Salic-Hainzl:** A. Employment/Salary (full or part-time): bit.bio Discovery. **G. Belli Valletta:** A. Employment/Salary (full or part-time): bit.bio. **G. Shipley:** A. Employment/Salary (full or part-time): bit.bio. **H. Garnett:** A. Employment/Salary (full or part-time): bit.bio. **C. Aruthan:** A. Employment/Salary (full or part-time): bit.bio. **T. Buerckstuemmer:** A. Employment/Salary (full or part-time): bit.bio Discovery. **O. Dovey:** A. Employment/Salary (full or part-time): bit.bio. **K. Firth:** A. Employment/Salary (full or part-time): bit.bio. **A. Turner:** A. Employment/Salary (full or part-time): bit.bio. **B. Haugen:** A. Employment/Salary (full or part-time): bit.bio. **T. Oosterveen:** A. Employment/Salary (full or part-time): bit.bio. **W. Bernard:** A. Employment/Salary (full or part-time): bit.bio.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.03/D20

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CIHR Catalyst Grant SVB-158618
CCNA Team 2 and WSGD funds

Title: Gonadal hormones contribute to sex differences in behavior, pathology, and epigenetic modifications in the 3xTg-AD mouse model of Alzheimer's disease

Authors: W. SONG¹, S. D. CREIGHTON², B. MICHALSKI¹, J. S. MOJGANI³, B. SAKIC¹, I. B. ZOVKIC², ***M. FAHNESTOCK**¹;

¹Psychiatry & Behavioural Neurosciences, McMaster Univ., Hamilton, ON, Canada;

²Psychology, Univ. of Toronto Mississauga, Mississauga, ON, Canada; ³Queen's Univ., Kingston, ON, Canada

Abstract: Sex-dependent differences in prevalence and severity are characteristics of Alzheimer's disease (AD). Using the 3xTg-AD mouse model, we previously reported that adult males show early behavioral dysfunction, altered epigenetic factors and lack of plaque/tangle pathology. Conversely, adult females retain more severe AD-like pathology and behavior. The present study examines whether gonadal hormones play a role in these differences in current cohorts of 3xTg-AD mice. 3xTg-AD and wild-type mice were gonadectomized or sham-operated at 3 months of age. After behavioral phenotyping at 6 months of age, the animals were assessed for molecular markers of AD pathology and expression of genes and histone variants associated with neurodegeneration. In female transgenic (AD) mice, gonadectomy resulted in poorer spatial learning performance. In contrast, in transgenic male animals, gonadectomy improved spatial memory. Compared to sham-operated AD females, gonadectomized AD females exhibited enhanced expression of mouse (*m*) *Mapt* and *App* genes, consistent with upregulated *MacroH2a1* and reduced binding activity of the repressive histone variant macroH2A1 at *mMapt*. In contrast, gonadectomized AD males showed significantly reduced cortical soluble A β 42 levels, reduced expression of the *App* and *MacroH2a1* genes, and increased macroH2A1 binding at the *mPsen1* promoter, compared to sham-operated AD males. In sum, female sex hormones enhance spatial learning, whereas male sex hormones reduce spatial memory at this age. Furthermore, female sex hormones increase expression of the *mMapt* and *mApp* genes but have no effect on human transgene expression or A β 42 levels. Conversely, male sex hormones increase *App* expression and A β 42 levels but have no effect on tau expression. Our work suggests that adult gonadal hormones contribute to sex differences in AD pathology and cognitive function, and that sex-specific differences in AD pathology are partially due to the action of histone variants associated with neurodegeneration such as macroH2A1.

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Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.04/D21

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Tspo regulates amyloid plaques formation and app processing in alzheimer's disease models in vivo and vitro

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Abstract: The 18-kDa translocator protein (TSPO), traditionally recognized as a marker of neuroinflammation and localized to the outer mitochondrial membrane, has recently been implicated in mitochondrial function and inter-organelle communication. However, its direct role in Alzheimer's disease (AD) pathogenesis remains unclear. In this study, we investigated the regulatory function of TSPO in amyloid plaque formation and amyloid precursor protein (APP) processing using both in vivo and in vitro models of AD. APP23 transgenic mice were crossed with TSPO knockout mice to assess the effects of TSPO deletion on amyloid pathology. Immunohistochemical analysis revealed a significant increase in plaque burden and altered morphology in the brains of TSPO^{-/-} x APP23 transgenic mice compared to TSPO^{+/+} x APP23 transgenic controls. To complement these findings, we established TSPO-knockout HEK293T cells and initiated lentiviral transduction with APP695 constructs to evaluate TSPO's effect on APP expression and downstream processing. Ongoing in vitro assays will quantify A β production and assess APP metabolism. These preliminary results suggest that TSPO influences both extracellular plaque formation and intracellular APP dynamics, supporting its potential role as a regulatory factor in AD progression.

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Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

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Program #/Poster #: PSTR266.05/D22

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA R03AG087436
NIH/NINDS T32NS096050

Title: Increasing SCN8A activity exacerbates multiple phenotypes in an amyloid beta model of Alzheimer's disease

Authors: *K. E. JAMES¹, J. C. WONG²;

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Abstract: *SCN8A* encodes the voltage-gated sodium channel Nav1.6, which regulates neuronal excitability and action potential initiation. *SCN8A* is differentially expressed in brain tissue from patients with Alzheimer's disease (AD), a progressive neurodegenerative disease characterized by cognitive impairment and extracellular amyloid- β (A β) plaques and intracellular neurofibrillary tau tangle (NFT) pathologies. Furthermore, up to 42% of AD patients show subclinical epileptiform activity, often with no previous history of seizures, and this activity has been linked to worsened cognitive performance in AD patients. Interestingly, a recent genome wide association study identified an AD risk locus on chromosome 12 containing *SCN8A*, which suggests that *SCN8A* may contribute to AD. Both amyloid precursor protein (APP) and A β have

been shown to increase expression of Nav1.6, which might contribute to neuronal hyperexcitability in AD. Given these observations, we hypothesize that *SCN8A* may be a genetic modifier for multiple AD phenotypes, such that increasing *SCN8A* activity in AD will exacerbate disease progression and severity. To test this, we crossed mice expressing the human *SCN8A* R1620L mutation (RL/+) to two well-established AD mouse models: 5xFAD and P301S mice that develop A β and NFT pathology, respectively. Mice that harbor both the 5xFAD/+ and RL/+ mutations (5xFAD/RL) exhibit learning and memory deficits, increased anxiety-like behavior, spontaneous seizures, and premature mortality, with most females dying by 4 months of age. We found that 5xFAD/RL mutants spent significantly less time in the center of the open field apparatus, suggesting increased anxiety-like behavior by 2 months of age in females ($N=4-7$ /genotype) and 4 months of age in males ($N=12-15$). Furthermore, we observed deficits in spatial memory in 4-month-old male 5xFAD/RL mutants compared to 5xFAD/+ mice ($N=8-10$). In addition to behavioral deficits, we also detected greater AD neuropathology (A β and GFAP) in the dentate gyrus of male 5xFAD/RL mutants at 4 months of age ($N=6$). Ongoing studies include evaluating behavior and AD neuropathology at other time points. Mice that harbor both the P301S and RL/+ mutations (P301S/RL) are currently undergoing assessments for behavioral deficits, seizure susceptibility, and neuropathological burden. These results support our hypothesis that *SCN8A* might be a genetic modifier for AD, such that increasing *SCN8A* activity in AD will exacerbate multiple phenotypes. The outcomes from our study provide the foundation for exploring *SCN8A* as a potential therapeutic target for AD and a blueprint for treatment development in other neurological disorders.

Disclosures: K.E. James: None. J.C. Wong: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

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Program #/Poster #: PSTR266.06/D23

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Behaviour and Biomarker Changes in ARTE10 mice reveal Translational Potential for Alzheimer's Disease and Mood-Related Comorbidities

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Abstract: Background: Alzheimer's disease (AD) is characterized by progressive cognitive decline and hallmark neuropathological changes including amyloid β (A β) plaque deposition. Patients often suffer from prodromal and co-morbid mood symptoms including anxiety and

agitation which evade current therapies. The ARTE10 transgenic mouse model, which co-expresses human APP and PS1 mutations linked to familial AD, develops early and robust A β pathology. Previous studies have focused on cognitive changes and pathology in advanced stages of AD. Here we aimed to comprehensively characterize the ARTE10 model for prodromal and disease state behavioural phenotypes and biomarkers. **Methods:** 5-month (N=16 WT, N=13 Homozygous (HM) ARTE10) and 10-month (N=16 WT, N=16 HM ARTE10) male mice were tested. Many behavioural domains were assessed with specific assays: mood: nest building, anxiety: canopy test, hyperactivity/agitation: running wheel, sensory-motor gating: pre-pulse inhibition, cognition: Y maze and Morris Water maze (MWM), motor function: rotarod. Plasma, CSF and brain samples were collected to evaluate the levels of NfL and amyloid using the MesoScale Discovery platform and then correlated to behavioural results. **Results:** Canopy test revealed increased anxiety-like behaviours in HM ARTE10's via reduced distance in the open zone ($p=0.0086$), and decreased looking over the edge ($p=0.0452$) at 5 months. By 10 months, both endpoints progressed and remained significant ($p<0.0001$; $p<0.0001$). Nest building consistently demonstrated mood disturbances (5M: $p=0.0006$; 10M: $p<0.0001$), and running wheel evidenced consistent hyperactivity (5M: $p=0.0233$; 10M: $p=0.0201$) in ARTE10 animals. Spatial working memory deficits were only found at 10 months using Y maze ($p=0.0039$) and MWM ($p=0.0429$). Biomarker analysis revealed significantly increased A β 42/40 ratio in the hippocampus ($p=0.015$) and cortex ($p<0.0001$) of 5 month ARTE10 mice. By 10 months amyloid load was further increased ($p<0.0001$). NfL was increased in the cortex ($p=0.0016$) and trending in CSF ($p=0.06$) at 5 months, these increased at 10M (cortex: $p=0.0004$, CSF: $p=0.0037$). Behaviour and biomarkers showed correlations between amyloid load and some behaviours. **Conclusions:** The ARTE10 mouse model exhibits prodromal behavioural phenotypes related to AD, pronounced amyloid pathology and early detectable biomarkers. Several phenotypes and biomarkers progress with age. These features support the strong translational potential of this model to advance therapeutic developments for AD, including those related to core pathology and comorbid symptoms that are not currently managed by approved treatments.

Disclosures: **A.M. Bernardo:** A. Employment/Salary (full or part-time): Transpharmation Ltd. Canada. **M. Baryla:** A. Employment/Salary (full or part-time): Transpharmation Ltd. Poland. **E. Sokolowska:** A. Employment/Salary (full or part-time): Transpharmation Ltd. Poland. **L. Griffin:** A. Employment/Salary (full or part-time): Taconic Biosciences Inc. **M. Jacobson:** A. Employment/Salary (full or part-time): Taconic Biosciences Inc. **J.A. Araujo:** A. Employment/Salary (full or part-time): Transpharmation Ltd. Canada. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Transpharmation Ltd. **G.A. Higgins:** A. Employment/Salary (full or part-time): Transpharmation Ltd. Canada.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

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Program #/Poster #: PSTR266.07/D24

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant R01 AG070214

Title: Loss of CELF2 Alters Critical Neurodevelopmental Splicing Programs and Attenuates Pathology in an Alzheimer's Disease Model

Authors: *I. SYED¹, X. LI⁴, Z. ZHANG⁵, Z. LIU², L. CHEN³;

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Abstract: CELF2 is an RNA-binding protein essential for mRNA splicing and stability in the nervous system, but its roles in brain development and neurodegeneration remain incompletely understood. Here, we show that CELF2 orchestrates transcriptomic programs critical for early neuronal maturation and modulates RNA processing of key Alzheimer's disease (AD)-related genes. Using a constitutive global CELF2 knockout mouse line, we observed perinatal lethality in homozygous knockouts, indicating a vital role for CELF2 in brain development. RNA sequencing and immunostaining of postnatal day 0 brains revealed widespread dysregulation of gene expression and alternative splicing, especially in pathways governing neuronal differentiation and synaptogenesis. Integration with published CLIP-seq datasets identified direct CELF2 targets among the affected transcripts, including *Dclk2* and *Camk2a*, which are essential for neuronal polarity and synaptic plasticity. We are currently testing whether overexpression of *Dclk2* or *Camk2a* can rescue synaptic deficits in primary neuronal cultures derived from CELF2 knockout pups, using dendritic spine density and PSD95 immunostaining as readouts. Previous work from our lab demonstrated that CELF2 promotes the production of the amyloidogenic APP751 isoform and contributes to tau mis-splicing, leading to an imbalance in 3R:4R tau isoforms. Based on these findings, we hypothesized that reducing CELF2 levels may protect against neurodegeneration. To test this, we crossed CELF2 heterozygous mice with the 5xFAD AD model. Remarkably, CELF2 haploinsufficiency reduced hallmark AD pathologies, including amyloid plaque load, AT8+ phospho-tau accumulation, and microglial activation. These data suggest that CELF2 drives neurodegeneration by altering the expression and splicing of tau- and amyloid-related transcripts. Thus, reducing CELF2 activity may attenuate disease progression by limiting toxic protein buildup and dampening neuroinflammatory responses, identifying CELF2 as a promising therapeutic target in AD.

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Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

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Program #/Poster #: PSTR266.08/D25

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA R00 AG065441

Title: Valosin-containing protein is cargo in amyloid precursor protein extracellular vesicles

Authors: *Y. LU;
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Abstract: The Amyloid Precursor Protein (APP), a genetic cause of Alzheimer's disease, is a type-I transmembrane protein that is metabolized by proteolysis in the endolysosomal system. APP is processed via two pathways: the non-amyloidogenic pathway, where metabolites of α -CTF and p3 are generated; and the amyloidogenic pathway, where metabolites including β -CTF and A β peptides are generated. Both full-length APP and its metabolites are secreted by cells in extracellular vesicles (EVs), but the content and function of APP-containing EVs remain unknown. We used genetically humanized rats to isolate App-EVs from primary neuronal conditioned media, followed by electron microscopy to confirm their vesicular identity. Further proteomic analysis identified the Valosin-containing protein (Vcp) as molecular cargo. Vcp helps maintain proteostasis by unfolding ubiquitinated proteins, and it facilitates protein degradation at endolysosomal compartments. Therefore, we probed for Vcp's functional relevance to App-EV. Pharmacological inhibition of Vcp activity was found to alter App metabolism by decreasing cellular α -CTF levels, caused by increased global EV secretion. While Vcp activity modulates the trafficking of App, App mutations independently influence the sorting of its metabolites. The Swedish mutation of App, which promotes amyloidogenic processing, was found to reduce App-EV abundance. While humanized control App-EVs were enriched in full-length App and α -CTF, App β -CTF was undetectable in EVs despite its high cellular levels in the Swedish mutant, suggesting selective sorting mechanism based on epitopes generated by nonamyloidogenic processing. Indeed, exogenous p3, which contains the α -cleaved neoepitope, induced a dose-dependent increase in small EVs, indicating that the α -cleaved neoepitope promotes EV biogenesis. Our findings highlight the importance of Vcp in the regulation of App-EV sorting and secretion, and link EV-mediated protein clearance to differential processing of App.

Disclosures: Y. Lu: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH grants NS085171
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Belfer Neurodegeneration Consortium at MD Anderson

Title: Ablation of Bach1 improves memory and reduces pathology in human amyloid precursor protein transgenic mice

Authors: *P.-Y. CHUANG, C.-H. FU, W. YU, J. CHIN;
Neurosci., Baylor Col. of Med., Houston, TX

Abstract: Bach1 is a transcriptional repressor that negatively regulates genes involved in antioxidant defense and has been implicated in several neurological disorders, including Alzheimer's disease (AD), Parkinson's disease, and Down's Syndrome. Transcriptomic analysis of transgenic mice expressing mutant human amyloid precursor protein (APP), a model of AD neuropathology, identified upregulation of Bach1 expression in APP mice that are more susceptible to cognitive impairment. Furthermore, pharmacologically induced seizures also elevated Bach1 expression, suggesting a role for this factor in mediating oxidative stress responses linked to disease vulnerability. To evaluate whether Bach1 plays a causal role in modulating neurological outcomes, we generated Bach1 knockout (Bach1-KO) mice and performed a comprehensive behavioral assessment of the mice. Across multiple assays, Bach1-KO mice showed normal performance, indicating that ablation of Bach1 does not cause overt behavioral deficits. In response to chemoconvulsant-induced seizures, Bach1-KO mice exhibited enhanced antioxidant responses and reduced expression of markers of astrocytic activation, suggesting increased resilience to neuronal stress. To investigate whether Bach1 contributes to AD-related neuropathology, we crossed Bach1-KO mice with APP mice. Compared to APP mice, APP/Bach1-KO mice displayed improved cognitive performance and reduced amyloid plaque burden. These results suggest that Bach1 influences both cognitive decline and neuropathological progression in APP mice. Together, our findings indicate that Bach1 deletion enhances endogenous neuroprotection without detrimental behavioral effects and point to Bach1 as a potential therapeutic target in neurodegenerative and seizure-related conditions.

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Poster

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the Guangdong-Hong Kong Joint Laboratory for Psychiatric Disorders
(2023B1212120004)

Title: Investigating the pathogenic mechanisms of familial Alzheimer's disease using iPSC-derived neuron models

Authors: *Y. FU^{1,2}, Y. DUAN^{1,2}, H. CAO^{1,2}, H. TSANG^{1,2}, A.-Y. FU^{1,2,3}, N. Y. IP^{1,2,3};
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Abstract: Familial Alzheimer's disease (FAD), which accounts for approximately 1-5% of all Alzheimer's disease cases, is characterized by early-onset cognitive decline, accelerated brain atrophy, and rapid disease progression. FAD is primarily due to single pathogenic mutations in *APP* (amyloid precursor protein), *PSEN1* (presenilin 1), or *PSEN2* (presenilin 2). As the *APP-V717I* mutation is the most common *APP* mutation in FAD, it is important to understand how this mutation regulates FAD pathogenesis. We selectively deleted the mutant allele in induced pluripotent stem cells (iPSCs) derived from a patient with FAD carrying the *APP-V717I* pathogenic mutation. We then differentiated these edited cells into cortical neurons. These edited neurons showed an inactivation of the mutant *APP* gene, leading to the elimination of aberrant APP processing and decreased amyloid-beta peptide production. In addition, the edited neurons exhibited decreased expression of genes associated with cell stress-related pathways. Thus, these findings elucidate the pathogenic effects of the *APP-V717I* mutation in dysregulated APP processing and exacerbated cellular stress. Importantly, the inactivation of the mutant allele not only rescues molecular and functional deficits but also provides insights into the pathological mechanisms of AD gene mutations, which may inform future therapeutic strategies for FAD.

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Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

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Program #/Poster #: PSTR266.11/D28

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: App gene dose drives transcriptional alterations in a model of Alzheimer's disease in Down syndrome

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Abstract: The basal forebrain provides major cholinergic, glutamatergic and GABAergic innervation locally and to the cortex and hippocampus, where neurotrophic support is received from target fields. People with Down syndrome are at extreme risk of developing Alzheimer's disease due to an extra allele of amyloid precursor protein (APP) on chromosome 21, and the basal forebrain is vulnerable to neurodegeneration. In mice, Alzheimer's disease in Down syndrome (DS-AD) has been modeled in the Dp16 line by duplication of a region of mouse chromosome 16 with substantial homology to human chromosome 21, resulting in 113 trisomic genes. To understand how trisomy manifests with aging in specific cellular subpopulations, we utilized single-nucleus RNAseq to investigate transcriptional alterations in basal forebrain of young (3-5 months) and aged (14-16 months) DS-AD mice and euploid 2N controls. These analyses identified significant downregulation of mRNAs associated with energy metabolism and the electron transport chain, synaptic function, translation, and proteolysis in GABAergic neurons, and robust activation of transposable elements, including LINE1, with aging. We also noted decreased expression of neurotrophin signaling-associated factors Akt1, Erk, and Ras in aged Dp16 GABAergic neurons, and an increase in TrkC mRNA in astrocytes. We hypothesized that among the trisomic genes, App is a major contributor to these alterations. To test this, we compared young and old 2N and Dp16 mice to a strain of modified Dp16 mice in which App gene dose has been corrected to two copies (Dp16^{App+/-}). By correcting App against an otherwise-Dp16 genetic background, we found a remarkable rescue of transcription of energy metabolism, synaptic function, translation, and proteolysis-related factors, and transposable element activation in GABAergic neurons. We also observed a correction of Akt1, Erk and Ras downregulation in neurons, a rescue of alterations in markers of microglial activation, and a reversal of TrkC transcriptional activation in astrocytes. Taken together, these findings indicate that among the 113 trisomic genes in Dp16, App is a key contributor to aberrations in diverse and critical cellular functions, transposable element activation, and alterations in downstream components of neurotrophin signaling pathways.

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Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH-NS07357
T32 AG00096-41

Title: Novel approaches to track neurodegeneration in 5xFAD mice reveal novel aspects of extracellular aggregate deposition

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Abstract: 5xFAD, a commonly-used mouse model of Alzheimer's disease, expresses 5 familial AD (FAD) mutations - three in human Amyloid Precursor Protein (hAPP) and two in human Presenilin 1 (hPSEN1). These mice exhibit age-dependent neurodegeneration of select populations of neurons, including those in layer V of the cerebral cortex and subiculum, along with other hallmarks of AD. However, there have been limited studies on features of neurodegeneration in particular neuronal populations. To selectively label a population of neurons that degenerate, we created a double transgenic mouse line by crossing 5xFAD mice with Rosa^{tdTomato(tdT)} reporter mice in which expression of Cre-recombinase permanently activates tdT expression (5xFAD/Rosa^{tdT}). Male and female 2-3-month-old 5xFAD/Rosa^{tdT}, and control Rosa^{tdT} mice, received AAV-retrograde/Cre (AAV-rg/Cre) into the spinal cord to transduce corticospinal neurons (CSNs) in layer V. Mice were perfused at 2-3, 5-6, 8 and 12-15 months of age. 5xFAD transgene expression was assessed using an antibody that recognizes only hAPP and not mouse APP. IHC for hAPP revealed age-dependent accumulation of hAPP aggregates in areas containing hAPP expressing neuronal cell bodies and in deep cortical layers containing axons from hAPP expressing neurons; these co-localized with Aβ plaques. TdT labeling enabled detailed tracking of degenerative changes including appearance of large axonal swellings and fragmentation, dendritic dystrophy, and provided a means to quantify loss of tdT-positive CSN cell bodies as mice age. In control Rosa^{tdT} mice without 5xFAD, retrogradely transduced layer V neurons expressing tdT exhibited normal morphology across age. Surprisingly, extracellular tdT aggregates appeared in areas with degenerating cell bodies and co-localized with extracellular aggregates of hAPP. Extracellular aggregates of hAPP and tdT were also present in areas containing axons and synaptic terminals from tdT-expressing neurons where none of the resident neurons expressed hAPP, including, in the pons where axons of corticospinal neurons form collaterals to synapse on neurons in the pontine nuclei. IHC for hAPP revealed that neurons in the red nucleus also expressed the 5xFAD transgenes and exhibited previously unreported age-related degeneration. Our results indicate that both hAPP and the otherwise soluble protein tdT are deposited in extracellular clusters as neurons degenerate, including areas where both hAPP and tdT presumably originate from the axons and synaptic terminals but where none of the resident neurons express the 5xFAD transgenes.

Disclosures: D. Gallardo: None. O. Steward: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); OS is Co-founder and has economic interests in the company Axonis Inc. which is seeking to develop new therapies for spinal cord injury and other neurological disorders..

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

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Program #/Poster #: PSTR266.13/D30

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: APP and ECE-1 conditional knockout mice reveal neuronal subtype-specific differences in A β production and regulation

Authors: D. CLAUSEN^{1,2}, G. BUONO², H. PENG², J. PACHECO-QUINTO², *E. A. ECKMAN²;

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Abstract: Amyloid- β (A β) levels in brain are governed by the balance between production, via proteolytic processing of the amyloid precursor protein (APP), and degradation or clearance through multiple pathways. The A β -degrading enzyme endothelin-converting enzyme (ECE)-1 is a membrane-bound protease that resides in A β -producing compartments, controlling the net production of A β by cells that co-express APP. Studies indicate that A β produced by multiple cell types, including excitatory neurons, inhibitory neurons, and oligodendrocytes, contributes to amyloid deposition in AD models. However, the individual contributions of these different cell types to normal physiological brain A β levels, in the absence of amyloid pathology, remain incompletely characterized. This study aims to address this gap in knowledge, beginning with analysis of excitatory vs. inhibitory neurons. In parallel experiments, we generated 4 conditional knockout (cKO) models by crossing *App* and *Ece1* floxed mice with *Camk2a-cre* and *Gad2-cre* recombinase-expressing mice. Littermates lacking cre served as controls. A β 40 levels were measured by sandwich ELISA in forebrain synaptosomes and soluble extracellular-enriched extracts from adult mice of either sex, n= 5-11 per group. APP levels were quantified by western blot. Data were analyzed by unpaired t-test. In mice with ablation of APP in *Camk2a*(+) excitatory neurons, APP protein in forebrain extracts was reduced by 39% (P=0.001). Synaptosomal A β was correspondingly reduced by 45% (P<0.0001) and extracellular A β by 37% (P=0.0007). Since APP and ECE-1 are co-expressed in neurons, we hypothesized that ablation of ECE-1 activity would increase net A β production. Synaptosomal A β was indeed doubled (P<0.0001) in *Camk2a* ECE-1 cKO mice, and extracellular A β was increased by 26% (P=0.0166). In mice with ablation of APP in *Gad2*(+) inhibitory neurons, changes in APP levels were not detectable in forebrain by western blot. Extracellular A β was reduced by 25% (P=0.0256) but synaptosomal A β was unchanged. In *Gad2* ECE-1 cKO mice, extracellular A β was increased by 33% (P=0.0277) but synaptosomal A β was also unchanged. Our results indicate that *Camk2a-cre* expressing excitatory neurons contribute ~40% of total A β in forebrain, and this pool of A β is majorly controlled by ECE-1 activity in the same neurons, such that loss of ECE-1 expression causes significant A β accumulation, especially in synapses. *Gad2-cre* expressing neurons contribute less to ECE-1 regulated A β production in forebrain and affect only extracellular A β . In ongoing studies we are analyzing other brain regions that contain a larger proportion of cre-expressing inhibitory neurons.

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Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

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Program #/Poster #: PSTR266.14/D31

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R21 AG079292
UtGP FIU Fellowship

Title: PSEN1 and 5xFAD Alzheimer's disease pig models show ambulatory differences compared to wild-type pigs during delays in a T-maze alternation task

Authors: *V. ROLDAN¹, D. MURPHY¹, L. M. ALLEN¹, K. LEE², T. J. JAROME³, T. A. ALLEN¹;

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Abstract: Alzheimer's Disease (AD) is the most common form of dementia, and it is projected to exceed seven million cases in the United States (US) this year (Rajan et al., 2021). Approved treatments have not reversed the disease or its signature cognitive decline (Cummings, 2023). Common observations in patients with AD include disturbed gait and other ambulatory alterations. Furthermore, mild cognitive impairment patients with gait and motor dysfunctions are at higher risk of developing AD (Ahn et al., 2023). Importantly, AD diagnoses are limited since these can only be done post-mortem. Thus, it has become crucial to understand the effects of the disease and its progression through other models. Pigs have emerged as helpful AD translational models due to their vast anatomical, physiological, and genetic similarities to humans. Together with our colleagues at the National Swine Resource and Research Center, we have developed the first pig presenilin-1 (PSEN1) (Uh et al., 2024) and 5xFAD familial AD models in the United States. We trained wild-type (WT) pigs (n = 7), PSEN1 pigs (n = 6), and 5xFAD pigs (n = 2) on an automated T-maze delayed-alternation task. Pigs were held in the start area before each trial for 5, 60, 120, and 240-second delays, and rewarded after alternating sides at the T-junction. We have shown that PSEN1 pigs take longer to learn the task and exhibit diminished spatial working memory compared to WT pigs (Allen et al., 2023). Here, we utilized DeepLabCut to track the ambulatory behaviors of wild-type (WT), PSEN1, and 5xFAD pigs in the T-maze task. We found that PSEN1 (PSEN1 vs. WT: $t(11) = -2.362$, $p = 0.038$) and 5xFAD pigs (5xFAD vs. WT: $t(7) = -2.790$, $p = 0.027$) had more walking bouts than WT pigs during delays, and their average walking speeds were lower than WT pigs (PSEN1 vs. WT: $t(11) = 2.587$, $p = 0.025$). Average speed comparisons between WT and 5xFAD pigs showed a similar trend but did not reach significance, likely due to our small 5xFAD sample size. These ambulatory behaviors in our AD pigs could be considered more frantic-like pacing behaviors during delay periods, although this is speculative. Importantly, these findings are generally consistent with the ambulatory activity observed in AD patients, who exhibit shorter step lengths and lower gait speeds (Visser, 1983). Ongoing analyses will assess behavioral indications of the

next choice in the start area on our AD pigs. Additionally, we will determine ambulatory activity and other behavioral indicators of memory in the decision area across WT and AD pigs. Overall, our results show pacing-like ambulatory disturbances in PSEN1 and 5xFAD pigs, providing additional validating evidence of the AD-like states in these models.

Disclosures: V. Roldan: None. D. Murphy: None. L.M. Allen: None. K. Lee: None. T.J. Jarome: None. T.A. Allen: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.15/D32

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: 800015822

Title: Cognitive, neurovascular and molecular features of the PSEN1 and 5xFAD pigAD models

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Abstract: We genetically engineered two novel pig models of Alzheimer's disease (pigAD) to bridge the translational gap between rodent models and human disease. The first model was generated by manipulating the *presenilin-1* gene (*PSEN1* on chromosome 14), given its pivotal role in early-onset familial Alzheimer's disease (AD). *PSEN1* encodes the catalytic subunit of the gamma-secretase enzyme, which breaks down amyloid beta precursor protein (APP) into smaller subunits, including beta-amyloid (A β) peptides. It is also crucial in the development of endothelial cell migration and barrier formation. Next, we developed a second pigAD model based on the widely used 5xFAD mouse model. 5xFAD models have three human *APP* gene mutations and two *PSEN1* mutations. 5xFAD models are considered significantly more aggressive, as they exhibit early and extensive amyloid plaques and tauopathies. Both models were made using CRISPR/Cas9 gene editing and were confirmed via genotyping. Here we wanted to validate these pigAD models at the cognitive, physiological, and molecular levels of analysis. To assess cognitive function, we tested PSEN1 pigs in a fully automated T-maze, which tests spatial working memory, a hallmark of AD. We observed significant differences in memory during the delay sessions between *PSEN1* ($n=6$, D5 ($t_{(181)} = 2.344$, $p = 0.0011$) and D120 ($t_{(181)} = 2.632$, $p = 0.0092$)), and D60 was significantly trending ($t_{(181)} = 1.724$, $p=0.08$) and D240 was not significant ($t_{(181)} = 0.398$, $p = 0.69$) compared to control pigs ($n=7$), indicative of spatial working memory deficits similar to those observed in mild to moderate AD patients. Next, we examined cerebral blood flow using pulsed-wave Doppler, targeting the carotid artery and retinal vasculature. We found that the PSEN1 pigs had significantly increased blood flow velocity

compared to controls ($p=0.0179$, $n=2$, $t_{(5)} = 3.470$). In contrast, 5xFAD pigs did not exhibit a significant change in velocity ($p=0.2822$, $n=2$, $t_{(4)} = 1.242$). Lastly, we investigated the molecular characteristics using immunohistochemistry and found little evidence of amyloid plaque burden in *PSEN1* pigs. Overall, the initial results indicate that pigAD models replicate key cognitive and vascular features of AD. In particular, the PSEN1 model yields promising results at both cognitive and physiological levels of analysis, and low-amyloid burden at the molecular level, suggesting that these pigs are well-positioned to isolate non-amyloid disease pathways in AD. The 5xFAD pig results remain inconclusive due to a small sample size; experiments are ongoing.

Disclosures: D. Murphy: None. T.A. Allen: None. J. Vargas: None. K. Monarch: None. K. Lee: None. T.J. Jarome: None. J. Yoon: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.16/Web Only

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Research grants to C.F.I. from Peking University, Chinese Institute for Brain Research, Beijing

Title: Conditional impairment of p75^{NTR} function in forebrain excitatory neurons affords significant neuroprotection in the 5xFAD mouse model of Alzheimer's Disease even during symptomatic disease stages

Authors: *X. LI¹, C. F. IBANEZ^{2,3,4,5}, M. XIE^{1,6,4},

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Abstract: p75 neurotrophin receptor (p75^{NTR}) contributes to the development of Alzheimer's Disease (AD) pathology by enhancing amyloid precursor protein (APP) cleavage and subsequent amyloid plaque formation. Introduction of p75^{NTR} variants lacking the death domain (Δ DD) or carrying a point mutation at Cys²⁵⁹ in the transmembrane domain (C259A) into 5xFAD mice confers neuroprotection against AD pathology. However, the cell type-specificity and timing during disease progression of p75^{NTR} action remain unclear. Here, we report that conditional knock-in of Δ DD and C259A p75^{NTR} specifically in forebrain excitatory neurons of 5xFAD mice significantly attenuated multiple AD pathological features, including amyloid plaque accumulation, neurite dystrophy and gliosis. Notably, the hippocampal amyloid plaque burden was reduced to a comparable extent as observed in whole body knock-in mice. Moreover, learning and memory were markedly improved in 5xFAD mice carrying Δ DD and C259A

p75^{NTR} variants in forebrain neurons. Strikingly, disruption of p75^{NTR} activity in advanced disease stage still have similar beneficial effect on AD progression. Introducing Δ DD and C259A p75^{NTR} variants after cognitive impairment rescued behavior performance in 5xFAD mice as well. These findings suggest that blunting p75^{NTR} function in AD patients can have beneficial effects even during symptomatic stages of the disease, offering a complementary approach to A β vaccination.

Disclosures: X. Li: None. C.F. Ibanez: None. M. Xie: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.17/D33

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Beijing PostDoc Scientific Research Funding 2021-ZZ-039

Title: Palmitoylation of p75^{NTR} contributes to Alzheimer's disease progression by regulating APP protein trafficking and degradation

Authors: *Y. MA¹, C. F. IBANEZ²;

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Abstract: Internalization and trafficking of amyloid precursor protein (APP) to endocytic compartments has been recognized as a key event in the generation of A β species and amyloid plaques in Alzheimer's disease (AD). The p75 neurotrophin receptor (p75^{NTR}) has been shown to interact with APP and regulate its internalization and intracellular trafficking. Here, we report that 5xFAD mice, an animal model of AD, expressing a palmitoylation-deficient mutant of this receptor (p75^{C281A}) display significantly attenuated AD neuropathology and cognitive deficits. p75^{C281A} showed enhanced internalization, trafficking to Rab5/Rab7 endosomes and lysosomal-mediated degradation. In mutant p75^{C281A} neurons, APP displayed accelerated co-internalization with p75^{NTR}, increased trafficking to late endosomes and lysosome, and enhanced degradation, thereby limiting neuronal A β production. Interestingly, the brain of 5xFAD mice shows increased levels of p75^{NTR} palmitoylation. These results indicate that palmitoylation of p75^{NTR} enhances its stability and, indirectly, that of APP by reducing their trafficking to the lysosome, resulting in increased A β accumulation and neuropathology in the AD brain. Selective inhibitors of p75^{NTR} palmitoylation may find applications in the treatment of AD.

Disclosures: Y. Ma: None. C.F. Ibanez: None.

Poster

PSTR266: APP/Abeta Pathway: Cellular and Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR266.18/D34

Topic: D.02. Ischemia

Support: RF1NS140137
RF1NS129022

Title: Ab-induced ros disrupts kir2.1-mediated neurovascular coupling via pip₂depletion in a mouse model of alzheimer's disease.

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Abstract: Functional hyperemia, the increase in cerebral blood flow in response to neuronal activity, is impaired in Alzheimer's disease (AD). This process is regulated by neurovascular coupling (NVC), in which capillary endothelial cells (cECs) sense elevated interstitial K⁺ through the inward rectifier K⁺ channel Kir2.1, triggering upstream vasodilation. While functional hyperemia and Kir2.1 activity in isolated cECs are known to be impaired at later stages of disease in the 5xFAD mouse model and can be rescued by phosphatidylinositol 4,5-bisphosphate (PIP₂) (Mughal et al., 2021), a necessary cofactor of Kir2.1, it remains unknown whether Kir2.1 in cECs can mediate the NVC response in AD, when this impairment begins, and what mechanisms underlie this dysfunction. Using 6-month-old male and female 5xFAD and littermate control mice, we assessed NVC with the *ex vivo* capillary-parenchymal arteriole (CaPA) preparation by stimulating capillaries with 10 mM K⁺ to mimic neuronal activity and measuring upstream arteriolar dilation. Arterial dilation in response to K⁺ stimulation was completely abolished in 5xFAD mice (0.286% dilation, SEM 0.9%), indicating a disruption in the capillary-to-arteriole signaling pathway. To investigate potential mechanisms, we applied Aβ oligomers (200 nM) or H₂O₂ (100 μM) to CaPA preparations from control mice and observed similarly impaired NVC responses (-0.34% ± 0.02% and 5.28% ± 2.4% respectively), suggesting a role for Aβ-induced oxidative stress. Co-application of ROS scavengers (superoxide dismutase, 120 U/mL + catalase, 1000 U/mL) preserved NVC during Aβ treatment, with K⁺ stimulation inducing a 59% (± 3.9%) dilation response. Kir2.1 expression assessed via immunohistochemistry (n=3 for both groups, 3 ROIs per mouse, ns, p = 0.3527 per Mann Whitney test) and RT-qPCR (control n = 6, 5xFAD n = 7, p= 0.7243 per unpaired t-test) showed no significant changes in 5xFAD compared to control mice, suggesting functional rather than transcriptional impairment. Calcium imaging in Cdh5-GCaMP8 mice revealed that Aβ increased endothelial Ca²⁺ signaling in a phospholipase C-dependent manner (n=3, p<0.01 per paired t-test) suggesting a PIP₂-depleting mechanism. Bath application or systemic intraperitoneal injection of PIP₂ rescued the NVC response in both 5xFAD and Aβ-treated control mice. These findings suggest that Aβ impairs Kir2.1-mediated NVC via ROS-dependent PIP₂ depletion and identify PIP₂ restoration as a potential therapeutic strategy for improving functional hyperemia in AD.

Disclosures: M.A. Bueno Guerrero: None. D. Jeffrey: None. F. Dabertrand: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.01/D35

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: DH Chen Foundation R-86U55A
NIH Grant R01AG061151
NIH Grant R01AG055523
NIH Grant R01AG070154
the Cure Alzheimer's Fund UCSD 2019-2544

Title: Tau pathology differs by sex in sarkosyl-insoluble fractions in Alzheimer's disease in Down syndrome

Authors: *X.-Q. CHEN, X. ZUO, W. C. MOBLEY;
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Abstract: Alzheimer's disease (AD), the leading cause of dementia, is more prevalent in females. Tau pathology, a hallmark of AD, has been reported to progress differently between sexes, but there are inconsistencies across studies. Down syndrome (DS), the most common genetic cause of AD (DS-AD), also exhibits tau pathology, yet sex differences are little studied. Here, we analyzed phosphorylated tau (PHF1), total tau, and sarkosyl-insoluble and soluble tau in DS-AD, DS without AD, and a rare partial trisomy 21 (PT) case. PHF1 levels were significantly elevated in DS-AD, while DS individuals without AD showed minimal tau pathology. Interestingly, total tau levels in DS-AD males were substantially lower than in female DS-AD. PHF1 levels normalized to total tau showed no sex difference, suggesting that differences in total tau underlie observed differences in PHF1 tau. Sarkosyl-insoluble tau, a marker of pathological tau aggregation, was also enriched in female versus male DS-AD brains, reinforcing sex-linked differences in tau pathology. These findings suggest sex-specific tau dynamics in DS-AD that parallel trends in sporadic AD. Understanding the source of these disparities may provide insights into AD pathogenesis and inform gender-specific therapeutic approaches for DS-AD.

Disclosures: X. Chen: None. X. Zuo: None. W.C. Mobley: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.02/Web Only

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ENEDIS (ERDF)
FRM DEQ20150331725
ANR-10-IAIHU-06

Title: Using eye-tracking and MRI to refine the digital phenotype of apathy and visual exploration in bvFTD under ecological conditions

Authors: *C. E. BERNER¹, E. KARPINSKI¹, J. BEHENSKA¹, V. ALTMAYER¹, E. VOLLE¹, R. A. LEVY¹, K. N'DIAYE¹, M. TOBA^{2,1}, B. BATRANCOURT¹;

¹Sorbonne Université, Inst. du Cerveau - Paris Brain Inst. – ICM, Inserm, CNRS AP-HP, Hôpital, Paris, France; ²Lab. of Functional Neurosciences (UR UPJV 4559), Univ. Hosp. of Amiens and Univ. of Picardie Jules Verne, Amiens, France, Amiens, France

Abstract: Apathy, a core feature of behavioral variant frontotemporal dementia (bvFTD), is characterized by reduced motivation and goal-directed activity. The ECOCAPTURE@LAB study (NCT03272230), part of the ECOCAPTURE program at the ICM, aims to define a digital phenotype of apathy within a naturalistic setting—a replicated waiting room. Prior work (Batrancourt et al., 2019) showed reduced exploration in apathetic bvFTD patients. Given that visual exploration reflects interaction with the environment and is sensitive to goal-directed behavior, we integrated eye-tracking and MRI to refine apathy's digital phenotype, hypothesizing reduced visual efficiency, engagement, and altered attentional network function. Participants included 18 bvFTD patients (mean age = 65.2; 11M/7F) diagnosed per clinical criteria, and 15 healthy controls (mean age = 62.3; 7M/8F) without neurological, psychiatric, or substance use history. Eye-tracking results showed that bvFTD participants had a lower saccade frequency than controls during free exploration, reflecting reduced visual exploration. In bvFTD, saccade frequency correlated with higher apathy severity (SAS). To refine this exploration deficit, we segmented behavior into sustained (e.g., reading) and unsustained (e.g., wandering) activity and analyzed heatmaps. Participants with higher apathy levels (mean SAS = 17.33), chose to engage in unsustained activity, and manifested greater gaze dispersion suggesting less focused engagement. In contrast, those with lower apathy levels (mean SAS = 14.8) chose to engage in sustained activity but explored less space than controls, indicating constrained scanning of the scene relative to the activity. These findings suggest that apathy impairs both the initiation and efficiency of visual exploration. To investigate the neural mechanisms underlying visual exploration, we conducted MRI analyses focusing on attentional networks that support goal-directed attention including the frontal eye fields (FEF) (Corbetta, 2002), responsible for voluntary saccades. We examined whether structural changes in these networks, particularly in the FEF, relate to impaired visual exploration and apathy severity. Results suggest that FEF alterations may be linked to apathy in bvFTD. These MRI and eye-tracking findings suggest that apathy in bvFTD is associated with deficits in visual exploration and altered attentional control networks. Future work will extend to real-world environments (ECOCAPTURE@HOME) to validate an at-home digital phenotype of apathy and guide therapeutical interventions.

Disclosures: C.E. Berner: A. Employment/Salary (full or part-time): IUIS: Institut Universitaire Ingénierie de la Santé, Sorbonne Université. E. Karpinski: None. J. Behenska: A. Employment/Salary (full or part-time): Sorbonne Université, Institut du Cerveau - Paris Brain Institute – ICM, Inserm, CNRS AP-HP, Hôpital de la Pitié Salpêtrière, Paris, France.. V. Altmayer: None. E. Volle: A. Employment/Salary (full or part-time): Sorbonne Université, Institut du Cerveau - Paris Brain Institute – ICM, Inserm, CNRS AP-HP, Hôpital de la Pitié

Salpêtrière, Paris, France. **R.A. Levy:** A. Employment/Salary (full or part-time): Hôpital de la Pitié Salpêtrière, Paris, France. **K. N'diaye:** A. Employment/Salary (full or part-time): Institut du Cerveau (Paris Brain Institute), CNRS. **M. Toba:** A. Employment/Salary (full or part-time): Laboratory of Functional Neurosciences (UR UPJV 4559), University Hospital of Amiens and University of Picardie Jules Verne, Amiens, France. **B. Batrancourt:** A. Employment/Salary (full or part-time): Sorbonne Université, Institut du Cerveau - Paris Brain Institute – ICM, Inserm, CNRS AP-HP, Hôpital de la Pitié Salpêtrière, Paris, France..

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.03/D36

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Developing Biomarkers for LATE Diagnosis and Differentiation from Alzheimer's Disease Using TDP-43 and Tau Seed Amplification Assays

Authors: ***M. GERASIMENKO**, T. L. GILLILAND, Z. WANG;
Case Western Reserve Univ., Cleveland, OH

Abstract: Limbic-predominant age-related TDP-43 encephalopathy (LATE) is a recently characterized neurodegenerative disorder primarily affecting individuals aged eighty and older, characterized by pathological accumulation of hyperphosphorylated TDP-43 protein in limbic structures. Clinically, LATE closely mimics Alzheimer's disease (AD), presenting predominantly as an amnesic dementia syndrome. Misdiagnosis is common due to overlapping clinical features and frequent co-existence of LATE and AD neuropathology. Currently, robust ante-mortem fluid biomarkers capable of accurately diagnosing LATE and distinguishing it from AD are lacking. In this study, we developed a novel seed amplification assay (SAA) targeting pathological TDP-43 and tau proteins. Our data from cerebrospinal fluid (CSF) and postmortem brain tissue demonstrate that our assay clearly distinguishes LATE samples from neurologically normal controls (NC) by TDP-43 SAA, and LATE-AD showed different seeding aggregation patterns with pure LATE. Furthermore, co-existing LATE-AD cases exhibited higher Thioflavin T (ThT) fluorescence than AD cases by tau SAA, and pure LATE showed no difference in tau seeding compared to NC, suggesting differential seeding behaviors between these groups. We also identified several novel site-specific phospho-tau (p-tau) epitopes capable of differentiating LATE pathology from AD. Further analyses will characterize the interaction between TDP-43 and tau pathologies through cross-seeding assays and immunohistochemical assessments across various neuropathological stages. This study developed a combination of tau and TDP-43 biomarkers for LATE, enabling effective ante-mortem diagnosis and accurate differentiation from AD. Successful development of these assays will significantly enhance clinical management by facilitating precise diagnoses, informed decision-making, and targeted therapeutic approaches for patients experiencing cognitive decline.

Disclosures: M. Gerasimenko: None. T.L. Gilliland: None. Z. Wang: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.04/D37

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant: R01 NS079172
Toulmin Pilot Award: HAND and Oligodendrocytes

Title: White Matter Pathology in HIV-Associated Neurocognitive Disorder: The Role of gp120-Induced Myelin Damage

Authors: *A. GAMBOA, M. KAJIKAWA, C. AGBEY, I. MOCCHETTI;
Neurosci., Georgetown Univ. Sch. of Med., Washington, DC

Abstract: Neuropathology in HIV-Associated Neurocognitive Disorders (HAND) is multifaceted, involving damage to both gray matter, which can impact neuronal integrity, and white matter, which is crucial for neural connectivity. Moreover, recent studies have shown a correlation between HIV, white matter damage and cognitive decline, despite the use of antiretroviral therapy (ART). Yet, little is known about how HIV infection of the brain causes damage to white matter. The observed white matter pathology could be the result of persistent viral reservoirs, ART, the associated comorbidities, or viral proteins. One potent neurotoxic viral protein is the envelope protein gp120, which is present in the brain of virally controlled study persons treated with ART. Thus, we used a gp120 transgenic (gp120tg) mouse model, which exhibits synaptic reduction in the hippocampus, as well as cognitive deficits to determine the role of gp120 in white matter damage. White matter damage often involves oligodendrocyte (OL) dysfunction, OL precursor cell (OPC) alterations, and Myelin Basic Protein (MBP) loss. Therefore, to investigate white matter pathology, we analyzed MBP intensity and altered OPC populations in coronal sections from the hippocampus of 15-month old female gp120tg mice. Aged-matched wild-type (WT) mice were used as controls. Immunohistochemical analysis revealed reduced MBP intensity, and altered OPC populations, in gp120tg when compared to WT. Moreover, analyses of these sections by electron microscopy uncovered white matter pathology denoted by areas of aberrant myelin structure, including hypermyelination. Future studies will establish the mechanisms utilized by gp120 to induce this pathology. These findings could identify novel therapeutic targets to mitigate white matter pathology and preserve cognitive function in people living with HIV.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.05/D38

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01NS128031
T32 Grant 1T32GM135028
Basic Science Pilot Grant from AFTD

Title: Development of a non-secreted progranulin mouse model

Authors: *M. LOVE¹, A. K. KAPLELACH², A. E. ARRANT³;

¹Univ. of Alabama at Birmingham, Birmingham, AL; ²Neurol., The Univ. of Alabama at Birmingham, Birmingham, AL; ³Neurol., Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: Loss-of-function mutations in progranulin (*GRN*) cause neurodegenerative disease. Heterozygous mutations, usually resulting in progranulin haploinsufficiency, cause frontotemporal dementia (FTD). Homozygous mutations, resulting in complete progranulin deficiency, cause an early-onset lysosomal storage disorder, neuronal ceroid lipofuscinosis. A common *GRN* polymorphism that reduces progranulin levels by ~20% is also associated with risk for FTD and AD. The association of low progranulin levels and neurodegenerative disease may be due to the loss of progranulin's effects. Progranulin is necessary for maintaining normal lysosomal function and plays a role in regulating lysosomal protease activity and lipid metabolism. Progranulin also exerts anti-inflammatory and neurotrophic effects. However, progranulin's site of action in mediating these effects remains unclear. Progranulin is a pro-protein that localizes to lysosomes. Before trafficking to lysosomes, progranulin is secreted and can engage in extracellular signaling through several receptors. To investigate progranulin's site of action, we previously developed lentiviral vectors expressing lysosome-targeted progranulin (L-PGRN), comprising human progranulin fused to the transmembrane domain and cytosolic tail of LAMP-1. This enables the delivery of progranulin to lysosomes with minimal secretion. Here, we generated an L-PGRN knock-in mouse line (*Grn^L*), in which endogenous progranulin is replaced with L-PGRN. We validated the *Grn^L* mouse line by confirming the loss of mouse progranulin and expression of human progranulin. We also confirmed that the human progranulin was not secreted, and therefore cannot engage in extracellular signaling. This *Grn^L* mouse line enables us to investigate the effects of the loss of progranulin secretion *in vivo*.

Disclosures: M. Love: None. A.K. Kaplelach: None. A.E. Arrant: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.06/D39

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01NS128031
AFTD Pilot Grant

Title: Progranulin acts in astrocytic lysosomes to promote neuronal growth

Authors: *A. K. KAPLELACH;
Neurol., The Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Loss-of-function progranulin (*GRN*) mutations are a major autosomal dominant cause of frontotemporal dementia (FTD). Progranulin is a secreted pro-protein that is necessary for maintaining lysosomal function and exerts neurotrophic and anti-inflammatory effects in the brain. Progranulin is constitutively secreted and may interact with signaling receptors before being taken up and trafficked to lysosomes. It is therefore not clear if progranulin's neurotrophic and anti-inflammatory effects are mediated through extracellular signaling or through lysosomes. To investigate progranulin's site of action, we developed lentiviral vectors expressing human progranulin (PGRN) fused to the transmembrane domain and cytosolic tail of LAMP-1 (L-PGRN). We used these vectors in primary hippocampal cultures from Sprague-Dawley rats to test the hypothesis that progranulin's neurotrophic effects are mediated in lysosomes. Consistent with prior reports, transduction with a PGRN lentiviral vector enhanced dendritic outgrowth in primary hippocampal cultures. Transduction with an L-PGRN lentiviral vector mimicked this effect. Further experiments using neuron- or astrocyte-specific lentiviral vectors revealed that L-PGRN only enhanced dendritic outgrowth when expressed in astrocytes. L-PGRN-transduced astrocytes were also able to promote dendritic growth in untreated neurons when co-cultured using transwell inserts, suggesting that changes to astrocyte secreted factors may enhance dendritic outgrowth. Bulk RNA sequencing of primary hippocampal astrocytes suggested that L-PGRN may reduce reactive phenotypes, and analysis of astrocyte conditioned media indicated that progranulin may suppress secretion of some proteins secreted by astrocytes. Consistent with these results, reducing astrocyte numbers with Ara-C increased dendritic outgrowth and occluded the effects of L-PGRN. Thus, progranulin may enhance dendritic outgrowth in primary hippocampal cultures reducing secretion of astrocytic factors that reduce dendritic growth in neurons. These data may inform mechanisms of astrocyte dysfunction and neurodegeneration in FTD due to *GRN* mutations.

Disclosures: A.K. Kaplelach: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.07/D40

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 1R21AG075798-01A1
Alzheimer's Association Grant AARG-22-974437
NIA Grant R21AG082193

Title: Blood-brain barrier breakdown contributes to neurovascular unit dysfunction in a mouse model of progranulin-deficient Frontotemporal dementia

Authors: *S. CHAKRABORTY¹, S. FRANCIOSA², O. BRACKO³;
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Abstract: Frontotemporal dementia (FTD) is the second leading cause of early-onset dementia, affecting an estimated 60,000-90,000 individuals in the USA, with no FDA-approved treatment currently available. Hereditary forms account for 30-40% of cases, with progranulin (PGRN) gene haploinsufficiency being a primary genetic cause. While PGRN is known to regulate diverse cellular processes, including lysosomal homeostasis and immune function, previous research on PGRN-mutated FTD patients and mouse models has predominantly focused on neuronal pathology. Here, we investigate neurovascular unit (NVU) dynamics in PGRN deficiency, with a specific emphasis on non-neuronal and vascular contributions to FTD pathogenesis. Utilizing the established PGRN-deficient mouse model, we employed a multi-modal approach, combining immunohistochemistry, Western blotting, qPCR, and *in vivo* multiphoton imaging. We observed significant alterations in the NVU cells of PGRN-deficient mice: microglial cell body area and branch length were decreased, while astrocytes exhibited altered branching patterns with reduced branch length and number. Astrocytic end-feet coverage on blood vessels was increased, whereas pericyte coverage was decreased. Furthermore, we found reduced cerebral blood vessel density and impaired blood flow, characterized by increased red blood cell capillary stalling. Given these widespread vascular and glial changes indicative of neuroinflammation, we assessed blood-brain barrier (BBB) integrity. Our analysis revealed decreased levels of the tight junction proteins Claudin-5 and Occludin, along with a reduction in their cerebral vessel coverage. In summary, our study corroborates previous reports of altered microglial and astrocytic morphology and BBB dysfunction in PGRN deficiency. Crucially, we unveil widespread NVU pathology, including altered pericyte-vessel interactions, reduced vascular density, and impaired cerebral blood flow. These findings underscore the critical role of non-neuronal and vascular dysfunction in PGRN-related FTD and could lead to the identification of novel targets for therapeutic intervention.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.08/D41

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Intramural Research Program 1ZIAAG000547

Title: Using ribosome profiling and proximity labeling to study mRNA translation and localization in FTD/ALS

Authors: *E. CRAIG¹, S. MEYDAN², V. RYAN¹;

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Abstract: Amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD) are co-occurring neurodegenerative disorders causing progressive motor and cognitive dysfunction. Both are associated with mutations in RNA-binding proteins (RBPs), which regulate translation and RNA metabolism. However, the mechanisms by which these mutations impact the spatial regulation of RNA and translation in neurons remain poorly defined. To investigate this, we use human induced pluripotent stem cell (iPSC)-derived neurons from wild-type and FTD/ALS-associated RBP-mutant lines. Our approach combines ribosome profiling (Ribo-seq), RNA-seq, pulsed SILAC (pSILAC), and proximity labeling (APEX) to study the localization and translation of RNA across subcellular compartments and organelles. We first use Ribo-seq to study active translation in whole cell and neurite fractions from wildtype neurons. We validate that both whole cell and neurite samples show active translation and find thousands of transcripts enriched in each compartment, which correspond to several neurodegenerative disease and neuron function pathways. Moreover, integration of Ribo-seq, RNA-seq, and pSILAC datasets reveals strong concordance among the three assays within each compartment, while distinct sets of genes are detected when comparing neurite versus whole cell fractions. Future experiments will apply the same techniques to RBP-mutant neurons to determine how disease-linked mutations alter local translation. In parallel, we are implementing APEX-based proximity labeling coupled with ribosome sedimentation to identify mRNAs localized to and translated on organelles of interest in neurons. Together, this study aims to define how local RNA regulation is altered in FTD/ALS and uncover new insights into the molecular basis of neurodegeneration.

Disclosures: E. Craig: None. S. Meydan: None. V. Ryan: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.09/D42

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Magnetic resonance imaging reveals novel neuroanatomical and microstructural phenotypes in the *grn*^{-/-} mouse model of frontotemporal dementia

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Abstract: Frontotemporal Dementia (FTD) is a group of related conditions resulting from the progressive degeneration of the temporal and frontal lobes of the brain. Patients display a spectrum of clinical syndromes that include progressive behavioral, language and motor dysfunction. One common form of FTD, termed FTD-GRN, is caused by haploinsufficiency of progranulin (PGRN coded by *GRN*). Homozygous loss of function of *GRN* causes the lysosomal storage disease Neuronal Ceroid Lipofuscinosis (CLN, type 11). Mice genetically deficient in *GRN* recapitulate many features of FTD-GRN and CLN-11 including lysosomal dysfunction, neuroinflammation, and behavioral deficits. However, despite these significant phenotypes, GRN knockout mice (GRN^{-/-}) do not display any neuronal death by 18 months of age. Having a fuller characterization of this model would help us better understand how PGRN insufficiency causes disease. An open question in the field is whether there are any neuroanatomical differences in this model, and if so, whether they correlate with or contribute to our interpretation of other findings in these mice.

Magnetic resonance imaging (MRI) is a non-invasive, translational tool used to measure neuroanatomical and microstructural phenotypes in mice and humans [Nieman *et al.*, 2007; Zatorre *et al.*, 2012]. Here, for the first time, we have used high-field 3D *ex vivo* MRI to examine the GRN^{-/-} mouse model of FTD, comparing regional volumes and diffusion tensor imaging (DTI) metrics. Specimens were also characterized by histology.

Comparing relative volumes in 18-month-old internally generated GRN^{-/-} mice (designed by Yajun Tand) versus WT littermates revealed 63 of 182 bilateral atlas regions with genotype effects (significant at 5% FDR). Regions in the olfactory bulb, cerebellum and thalamus were significantly smaller in GRN^{-/-} animals vs WT, while regions in the hippocampus were significantly larger in GRN^{-/-} animals vs WT. DTI MRI findings were dominated by genotype differences in mean radial diffusivity, which increased by up to 45% in 21 regions at 5% FDR. This change impacted both white and gray matter structures in the cerebellum and several white matter tracts, including the corpus callosum. Similar assessments were conducted in GRN^{-/-} mice versus their WT littermates at 3 months, before other phenotypes appear, and no significant differences were observed between genotypes at this age. To our knowledge, this is the largest MRI study of aged GRN^{-/-} mice containing measurements of neuroanatomy and DTI. This novel characterization provides further understanding of this widely-used model and offers additional readouts that can be used in therapeutic development.

Disclosures: **L. Spencer Noakes:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **J. Treinish:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **M. Preda:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **M. Karakatsani:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **M. Marzelli:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals. **M. Germino:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals. E.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.10/D43

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG068581
RF1AG076122
R21AG085314
R01AG077771

Title: The nuclear-transport protein Importin-13 can effectively counter TDP-43 proteinopathy.

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Abstract: TDP-43 aggregation is a hallmark of both Amyotrophic Lateral Sclerosis (ALS) and Frontotemporal dementia (FTD) and a common co-pathology in other neurodegenerative diseases. In both cases, TDP-43 is mislocalised from the nucleus to the cytoplasm, where it forms protein inclusions that are posttranslationally modified via phosphorylation, ubiquitination and proteasomal cleavage, among other changes. In addition, members of the Nuclear Pore

Complex (NPC) are sequestered into TDP-43 aggregates, triggering nuclear-cytoplasmic transport defects. Previous experiments in our lab found that coexpression of Importin-13 (IPO13) can reduce TDP-43 aggregation. Here, we characterize the effect of IPO13 on different TDP43 fragments and identify the minimal IPO13 fragment necessary and required for clearance of TDP43 aggregates. To assess IPO13 disaggregation activity, we transiently transfect HEK293 cells with plasmids encoding for different TDP-43 constructs and IPO13 fragments tagged with mEGFP or mCherry, respectively. We evaluate TDP-43 aggregation using immunofluorescence and RIPA-Urea biochemical fractionation. In addition, we used AAV-transduced murine brain slice cultures (BSCs) to investigate IPO13 disaggregation activity in the context of intact CNS tissue. We found that coexpression of full length IPO13 and specific fragments can effectively reduce TDP-43 aggregation in HEK293 cells and in BSCs. In addition, coexpression of full length IPO13 can effectively reduce the amount of RIPA-insoluble TDP-43, as well as the pathologically phosphorylated pS409/410 TDP-43. In conclusion, in this work we identified IPO13 as a potential suppressor of pathological TDP-43 aggregation. Furthermore, we found that shorter fragments of IPO13 are sufficient to reduce the burden of TDP-43 aggregates. This minimal, active fragment is a promising candidate to be further developed and rationally optimized into a novel therapeutic for ALS, FTLTDP, LATE, and TDP-43 co-pathology in AD and related neurodegenerative multiproteinopathies.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

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Program #/Poster #: PSTR267.11/D44

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH- R01EY023299
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All May See Foundation Postdoctoral Grant Awards

Title: A novel progranulin-mTOR axis regulates mitochondrial metabolism and microglial activation in neurodegenerative diseases

Authors: *S. PANDA, A. LAKKARAJU;
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Abstract: Progranulin (PGRN) has been shown to play critical roles in regulating neuronal survival and inflammation. In the brain, loss of PRGN is associated with microglial activation

and neurodegeneration in diseases such as Frontotemporal dementia (FTD), Alzheimer's disease (AD) and Parkinson's disease (PD). Recent noninvasive imaging studies of the retina, the most accessible part of the CNS, show that retinal deficits precede measurable cognitive changes in FTD and AD. Here, we investigated the molecular mechanisms by which PRGN loss compromises retinal function and how this temporally relates to changes in the brain. We used super-resolution high-speed live imaging of wildtype and *Grn*^{-/-} mice retinas, transcriptomics, and biochemical approaches to identify potential pathways regulated by PRGN in the retinal pigment epithelium (RPE), the tissue that nourishes and supports the light-sensing photoreceptors. Microglial activation and photoreceptor degeneration were monitored by immunostaining histological analyses and electroretinography (ERGs). These deficits occurred between 1-4 months of age compared to brain pathology that is apparent only after 12 months in *Grn*^{-/-} mice. Our data revealed a striking hypoactivation of the mTOR pathway in 4-month-old *Grn*^{-/-} mouse RPE. The resulting decrease in 4E-BP1-mediated translation of Mitochondrial Fission Process 1 (MTFP1) leading to hyperfused mitochondria with severe bioenergetic deficits. 4E-BP1 inhibition upregulated cathepsin L expression, leading to aberrant complement C3aR activation, recruitment of microglia to the subretinal space, and subsequent photoreceptor degeneration. Transcriptomic analyses of mouse RPE confirmed these findings. A small molecule C3aR inhibitor prevented microglial activation and restored photoreceptor function in *Grn*^{-/-} mice. Our results identify a novel mechanism by which PRGN loss leads to mTOR inhibition and early metabolic deficits in the RPE, subsequently promoting neurodegeneration in the retina. The early manifestation of retinal pathology compared to the brain strongly suggest that the retina is uniquely sensitive to PRGN loss. Our study underscores the significance of retinal changes as early indicators of neurodegenerative processes associated with PRGN deficiency.

Disclosures: **S. Panda:** A. Employment/Salary (full or part-time): National Institutes of Health (NIH), University of California San Francisco. **A. Lakkaraju:** A. Employment/Salary (full or part-time): National Institutes of Health (NIH), University of California San Francisco.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.12/D45

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: RNIH Grant: R01AG063857
HMRI development project support

Title: Altered Prefrontal Hemodynamics with Interference Challenging in Cognitively Healthy Individuals with High Plasma P-tau217

Authors: *A. AL-EZZI¹, X. WU², N. ASTRAEA², M. KLEINMAN⁷, A. FONTEH³, R. KLONER⁴, A. M. SUCHY-DICEY⁵, X. ARAKAKI⁶;

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Abstract: Introduction: Plasma p-tau217 has been considered as a promising biomarker for early detection of Alzheimer's disease. However, its relationship with brain hemodynamic responses remains unclear during resting or during cognitive tasks. We investigated blood oxygenated hemoglobin (HbO) concentration changes in cognitively healthy (CH) individuals with low and high plasma pTau-217 levels. **Methods:** Forty-two CH participants were classified into tertiles based on plasma pTau-217 levels (1.9- 18.9 pg/mL): including low (pTau-217=1.9- 3.2 pg/mL, n = 14) and high tertiles (pTau-217 = 6.7-18.9 pg/mL, n = 14) pTau-217. Participants completed Stroop task, where colored words were shown one at a time and participants were instructed to respond to the ink color, when the ink color either matched (congruent) or did not match (incongruent) the word shown. Using Functional Near-Infrared Spectroscopy (FNIRS) (Biopac), we examined comparisons of HbO at the lateral left, medial left, lateral right, and medial right prefrontal cortex (PFC) regions between the low and high tertiles during resting state (eyes open (EO), eyes close (EC)) and during Stroop interference task. We also assessed the association between HbO, and heart rate (HR) recorded after the Stroop task. **Results:** During EO, high pTau-217 tertile had a lower HbO in the lateral right PFC compared to low pTau-217 tertile ($p = .028$, mean difference = -0.006, 95%CI = -0.01 to -0.001) (**Figure 1A**). High pTau-217 tertile had significantly lower change in HbO from EC to EO conditions, at the lateral right PFC region ($p = .021$, mean difference = -0.007, 95%CI = -0.01 to -0.001) (**Figure 1B**). In addition, high pTau-217 tertile had significantly higher difference in HbO from EO to congruent trials in the lateral right region ($p = .042$, mean difference = 0.01, 95%CI = 0.001 to 0.022) (**Figure 1C**). Comparisons for other regions were not significant. Additionally, high pTau-217 tertile showed a positive correlation between lateral right PFC HbO and post-task HR during congruent trials ($r = .85$, $p = .001$), while Low pTau-217 tertile showed a negative correlation ($r = -.55$, $p = .043$) (**Figure 2**). **Conclusions:** Among CH participants, high pTau-217 tertile at lateral right PFC had lower HbO during EO, less increase of HbO from EC to EO conditions, and significant increase of HbO from EO to congruent trials. Lateral right HbO were positively correlated with HR in high pTau-217 participants, but a negative correlation in low pTau-217 individuals. An increase in HbO from the resting to the congruent trials may reflect compensatory neural-cardiovascular activity and suggest early cardiocerebrovascular changes in those with increased pTau-217 levels.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.13/D46

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Loss of function of *Sordd2* extends lifespan and alleviates neurodegeneration in a *Drosophila* model of C9ORF72 FTD/ALS

Authors: *V. NIKOLAKI¹, C. CHEN², S. LEE¹, T. YU³, Y. JUN¹, M. PONS⁴, Z. WENG³, P. EMERY², F.-B. GAO¹;

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Abstract: Frontotemporal dementia (FTD) is the most common form of dementia under the age of 60. The hexanucleotide GGGGCC repeat expansion in the first intron of *C9ORF72* is the most prevalent genetic cause of FTD and ALS. This expansion leads to the production of repeat RNAs that are bidirectionally translated into toxic dipeptide repeat (DPR) proteins, including poly(GR). Poly(GR) expression pattern in post mortem patient brain tissues strongly correlates with areas of neurodegeneration but the underlying pathogenic mechanisms remain largely unknown. We developed a (GR)₈₀ *Drosophila* transgenic model to study its toxicity *in vivo*. Using bulk RNA-seq we found that the most downregulated gene in the brain of poly(GR)-expressing male flies is *Sordd2*, which encodes a novel ubiquitin E3 ligase. Here we find that partial loss of function of *Sordd2* alleviates poly(GR)-induced rough eye phenotype. In contrast, *Sordd2* overexpression exacerbates disease phenotypes, such as climbing deficits and rough eye phenotype. Interestingly, complete loss of function of this gene in wildtype flies extends lifespan under both normal and malnutrition conditions and improves locomotor activity in an age-dependent manner. Genetic deletion of *Sordd2* also decreases DNA damage in aged flies, while *Sordd2* overexpression increases DNA damage, a hallmark of aging. Moreover, in contrast to old wildtype flies that exhibit reduced daytime and early night sleep and increased sleep fragmentation, aged male *Sordd2* loss of function mutants maintain youthful sleep patterns and quality, while aged female mutants show reduced daytime sleep but preserved early night sleep. These findings demonstrate a novel function of *Sordd2* in regulating sex-dimorphic aging-dependent sleep regulation in wildtype flies, as well as alleviation of neurodegeneration in C9ORF72-FTD/ALS. Further understanding the molecular functions of *Sordd2* will have broader implications in aging and neurodegeneration.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.14/D47

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant RF1NS091299 to DCZ
Penn State COM, Startup funds to DCZ

Title: A novel *Drosophila* model of dementia based on *C9orf72*-associated G4C2 hexanucleotide repeat expression in mushroom body neurons identifies Dally-like protein as a disease modifier

Authors: *B. S. CHAUHAN¹, M. A. BRENNAN¹, P. FORSTMEIER¹, R. GODFREY², D. C. ZARNESCU¹;

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Abstract: GGGGCC (G4C2) hexanucleotide repeat expansions (HREs) within the first intron of *C9orf72*, are the most common cause of inherited Amyotrophic lateral sclerosis (ALS) and Frontotemporal dementia (FTD). Previous studies suggest that disease pathomechanisms include both RNA toxicity and repeat-associated non-AUG (RAN) translation that generates dipeptide repeats (DPRs). Here we describe a novel *Drosophila* model of dementia based on the expression of G4C2 hexanucleotide repeats (HRs) that generate both RNA and DPRs in a subset of mushroom body neurons (MBNs). Overexpression of 44 G4C2 HRs ((G4C2)_{44X}) causes age dependent axonal morphology defects while, polyGR DPRs synthesized via RAN translation, as evidenced by expression of a GFP reporter (polyGR-GFP), are detected in axonal puncta that seem to increase in size during aging, concomitant with progressive axonal thinning, when compared to 12 G4C2 HRs ((G4C2)_{12X}) or w¹¹¹⁸ genetic background controls. Additionally, we observed nuclear to cytoplasmic mislocalization of endogenous *Drosophila* TDP-43 (TBPH) in Kenyon cells expressing G4C2 HREs, as well as increased cell death marked by cleaved Dcp-1. Lifespan analysis revealed a significant reduction in survival in both (G4C2)_{12X} and (G4C2)_{44X} flies, with males exhibiting more pronounced effects than females. Behaviorally, (G4C2)_{44X} flies show altered daytime and nighttime sleep patterns including sleep bout fragmentation. Y-maze assays indicate impairments in spatial working memory in (G4C2)_{44X} flies compared to controls. Importantly, we identified Dally-like protein (Dlp), a cell-surface glypican involved in axonal growth and signaling, as a potential target of *C9orf72* toxicity. Indeed, (G4C2)_{44X} expression reduces Dlp levels, while restoring Dlp expression in (G4C2)_{44X} improves aspects of spatial working memory. Taken together, these findings show that G4C2 HRs cause age-related axonal and behavioral deficits in flies, modeling key features of ALS/FTD, and suggest that Dlp, a glypican linked to Wnt signaling and synaptic plasticity may provide a therapeutic avenue to mitigate *C9orf72* toxicity.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG060731
MSU- Department of Translational Neuroscience
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Discretionary Funding Initiative (Caryl Sortwell and Scott Counts)

Title: Amyloid plaque co-pathology amplifies phosphorylated alpha-synuclein burden in the entorhinal cortex in a novel rat model of Dementia with Lewy Bodies

Authors: *M. HORE¹, C. J. KEMP¹, J. PATTERSON¹, J. R. HOWE¹, M. KUBIK¹, M. GIFANI², J. BECK¹, S. E. COUNTS³, C. E. SORTWELL⁴;
¹Michigan State Univ., Grand Rapids, MI; ²Michigan State Univ., Grand Rapids, MI; ³Transl Sci. and Mol Med, Family Med., ⁴Translational Sci. and Mol. Med., Michigan State Univ., Grand Rapids, MI

Abstract: Dementia with Lewy Bodies (DLB) is the second most common neurodegenerative cause of dementia. Pathologically, DLB is characterized by: 1) nigrostriatal degeneration, 2) amyloid-beta (A β) peptides in the form of plaques, and 3) Lewy bodies (LBs) consisting primarily of phosphorylated alpha-synuclein (pSyn), with or without 4) hyperphosphorylated tau protein as neurofibrillary tangles (NFTs). LB-like and amyloid plaque co-pathologies in DLB have been replicated in rodent models, but none have included nigrostriatal degeneration. In the present study, we used the strategy of α -synuclein (α -syn) preformed fibril (PFF) injections into Tg344-19 AD transgenic rats. PFF injections targeted to the nigrostriatal system result in accumulation of pathological LB-like inclusions in cortical, limbic and nigrostriatal regions, and nigrostriatal degeneration. The Tg344-19 AD rat expresses mutant human amyloid precursor protein and presenilin 1 genes resulting in age-dependent accumulation of plaques and NFTs, cortical and hippocampal neurodegeneration, and cognitive disturbances. In the present study, male and female AD Tg and wildtype (WT) F344 rats received bilateral intranigral injections of α -syn PFF or α -syn monomer control at 6 months of age, followed by additional bilateral intrastriatal injections of α -syn PFF or monomer at 10 months of age. Rats were then euthanized at 12 months of age. Treatment groups are designated as follows: Control = monomer injections into WT F344 rats, PD = PFF injections into WT F344 rats, AD = monomer injections into Tg344-19 AD rats, and DLB = PFF injections into Tg344-19 AD rats. pSyn aggregate and A β plaque co-pathology was prominent in DLB rats, particularly within the cingulate cortex, basolateral amygdala, and entorhinal cortex. A β plaque load was not impacted by the presence of pSyn co-pathology in any region examined. pSyn load was not impacted by the presence of A β plaque co-pathology in the cingulate cortex or basolateral amygdala. However, in the entorhinal cortex A β plaque co-pathology was associated with a significant, three-fold increase in pSyn load ($p < 0.0001$). These findings suggest that A β plaque co-pathology may potentiate pSyn aggregation, specifically in the entorhinal cortex, a region known to be affected in DLB that is critical for executive function and spatial memory. The present results provide a platform to support development of disease-modifying therapies for DLB as well as a model to investigate the mechanisms driving synergistic interactions between α -syn inclusion and A β plaque co-pathologies.

Disclosures: M. Hore: A. Employment/Salary (full or part-time): Michigan State University. C.J. Kemp: A. Employment/Salary (full or part-time): Michigan State University. J. Patterson:

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Employment/Salary (full or part-time): Michigan State University. **J. Beck:** A.
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Employment/Salary (full or part-time): Michigan State University. **C.E. Sortwell:** A.
Employment/Salary (full or part-time): Michigan State University.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.16/D49

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant RF1AG082478
NIH Grant R21NS125845

Title: The role of CYLD variants in frontotemporal dementia pathogenesis

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Abstract: Frontotemporal dementia (FTD) is the second most common cause of dementia after Alzheimer's Disease and the leading form of dementia in people under the age of 60. More than 50% of FTD cases present with the behavioral variant, characterized by personality and behavior changes. Up to 40% of FTD cases are inherited, with more than 15 genes linked to FTD, most of which are involved in autophagy. CYLD is a recently discovered FTD gene, with a gain-of-function mutation M719V linked to FTD. CYLD is a k63-specific deubiquitinase and a tumor suppressor involved in the inhibition of NF-κB signaling activation in the immune system and an activator of autophagy in neurons. Several variants of CYLD have been associated with FTD. Among them, CYLD-M719V, a missense mutant reported to increase deubiquitinase activity, has been reported to be potentially pathogenic and autosomal dominant. The underlying pathogenic mechanism remains unknown. We aim to uncover how M719V dysregulates neuronal autophagy, impairs intrinsic and synaptic properties, and mediates FTD pathogenesis. Toward this goal, we generated somatic transgenic mice expressing M719V or EGFP or wild-type CYLD (as controls) via intracerebroventricular (ICV) AAV injection at postnatal day 0 (p0). Behavioral profiling indicated that M719V mice showed age-dependent FTD-related behavioral impairments, including hypoactivity, risk-taking behavior, reduced social interaction, and loss of empathy, compared to CYLD wild-type and control mice. Moreover, slice electrophysiological analysis revealed profound loss of neuronal excitability, characterized by reduced spiking, increased rheobase, and decreased input resistance in mutant neurons

expressing M719V compared to WT and EGFP controls. M719V neurons showed reduced frequency of miniature excitatory synaptic currents (mEPSCs) compared to the control group. However, there were no significant changes in mEPSC amplitude. These results establish AAV-based somatic M719V transgenic mice as a viable mouse model of CYLD-FTD that can be used to elucidate the pathogenesis mechanisms by which CYLD mutations, and autophagy dysregulations in general, induce FTD.

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Poster

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Title: TGF β activation promotes astroglial TDP-43 proteinopathy in an organoid model of FTL D

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Abstract: Frontotemporal lobar degeneration (FTLD), a common disease in older adults, involves neurodegeneration in the cortex and thalamus. One leading genetic cause is the R493X nonsense mutation in the *Progranulin* (*GRN*) gene, leading to haploinsufficiency of the progranulin protein. Resulting pathologies include neuroinflammation, synaptic loss, and mislocalization of RNA-binding protein TDP-43 into the cytoplasm of neurons and glia. Recent studies have found highly conserved astrocyte transcriptomic signatures in the cortex and thalamus of FTLD-*GRN* patients, suggesting a role of astrocytes in the neuropathology of FTLD, but the specific mechanism remains unclear. In this study, we derived *GRN*^{+/+} and *GRN*^{-/-} cortical organoids from human induced pluripotent stem cells (iPSC) and conducted single-cell RNA sequencing (scRNA-seq) at ages 5 weeks (n = 5), 16 weeks (n = 5), and 25 weeks (n = 6). The scRNA-seq analysis revealed precocious astrogliogenesis and significant immune activation in *GRN*^{-/-} astrocytes compared *GRN*^{+/+} astrocytes. MultiNicheNet analysis suggested that the upregulation of canonical and noncanonical TGF- β signaling pathways might be the primary driver of this immune activation phenotype. To test this, we treated the *GRN*^{+/+} and *GRN*^{-/-}

organoids with TGF- β inhibitors (LDN193189 or SB431542) for five weeks. Indeed, we found significantly less immune activation in treated compared to non-treated *GRN*^{-/-} organoids as characterized by NF- κ B mean fluorescent intensity (n = 3, 97.7 $\pm$ 1.7 vs 95.9 $\pm$ 1.5 vs 116.8 $\pm$ 5.3, respectively) and reduced TDP-43 fibrils in astrocytes as characterized by percent of pTDP-43⁺ SOX9⁺ cells out of total SOX9⁺ cells (n = 3, 14.9 $\pm$ 2.6% vs 13.2 $\pm$ 3.8% vs 38.1 $\pm$ 1.2%, respectively). The treated *GRN*^{-/-} organoids were very phenotypically similar to the *GRN*^{+/+} organoids. These results support the hypothesis that upregulated TGF- β signaling contributes to the immune activation and TDP-43 fibril phenotypes in *GRN*^{-/-} astrocytes. Finally, more broadly, this study demonstrates the cell autonomous role of astrocytes in the pathogenesis of FTLD and offers a novel platform to test therapeutic targets that mitigate astroglial toxicity and TDP-43 proteinopathy.

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Poster

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Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Hidden players in MCI: how Lewy body pathology hastens Alzheimer's disease progression

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Abstract: Comorbid Lewy body (LB) pathology occurs in up to half of Alzheimer's disease (AD) cases and may exacerbate clinical outcomes by accelerating neurodegeneration. However, its role at the mild cognitive impairment (MCI) stage—when interventions could be most impactful—remains poorly understood. Using data from the ADNI database, we examined whether LB co-pathology influences subclinical decline and conversion to dementia in participants with biomarker-confirmed AD (AD+: CSF p-tau₁₈₁/A β ₄₂ ratio > 0.021). LB status was determined via CSF α -synuclein seed amplification assays. MCI was further stratified into stable MCI (sMCI; remained MCI $\geq$ 4 years) and progressive MCI (pMCI; converted to dementia $\leq$ 4 years). Among AD+ sMCI participants, individuals who were LB+ showed steeper decline in memory and language composite scores compared to LB- counterparts, despite not formally progressing to dementia (**Figure 1A**). In the AD+ pMCI, Kaplan-Meier analyses revealed that LB+ participants converted to dementia more rapidly than LB- participants (49% vs. 32% by ~18 months; $p < 0.05$), corresponding to a 47% increased hazard of progression after adjusting for age, sex, education, and APOE ϵ 4 (**Figure 1B**). Taken together, these findings show that LB co-

pathology not only accelerates cognitive decline within the MCI stage but also hastens the transition to dementia. By capturing LB pathology during MCI, our results highlight a critical juncture for potential therapeutic interventions, especially as anti-amyloid treatments gain traction. This work underscores the importance of identifying multiple proteinopathies early, refining prognoses, and optimizing therapeutic trials.

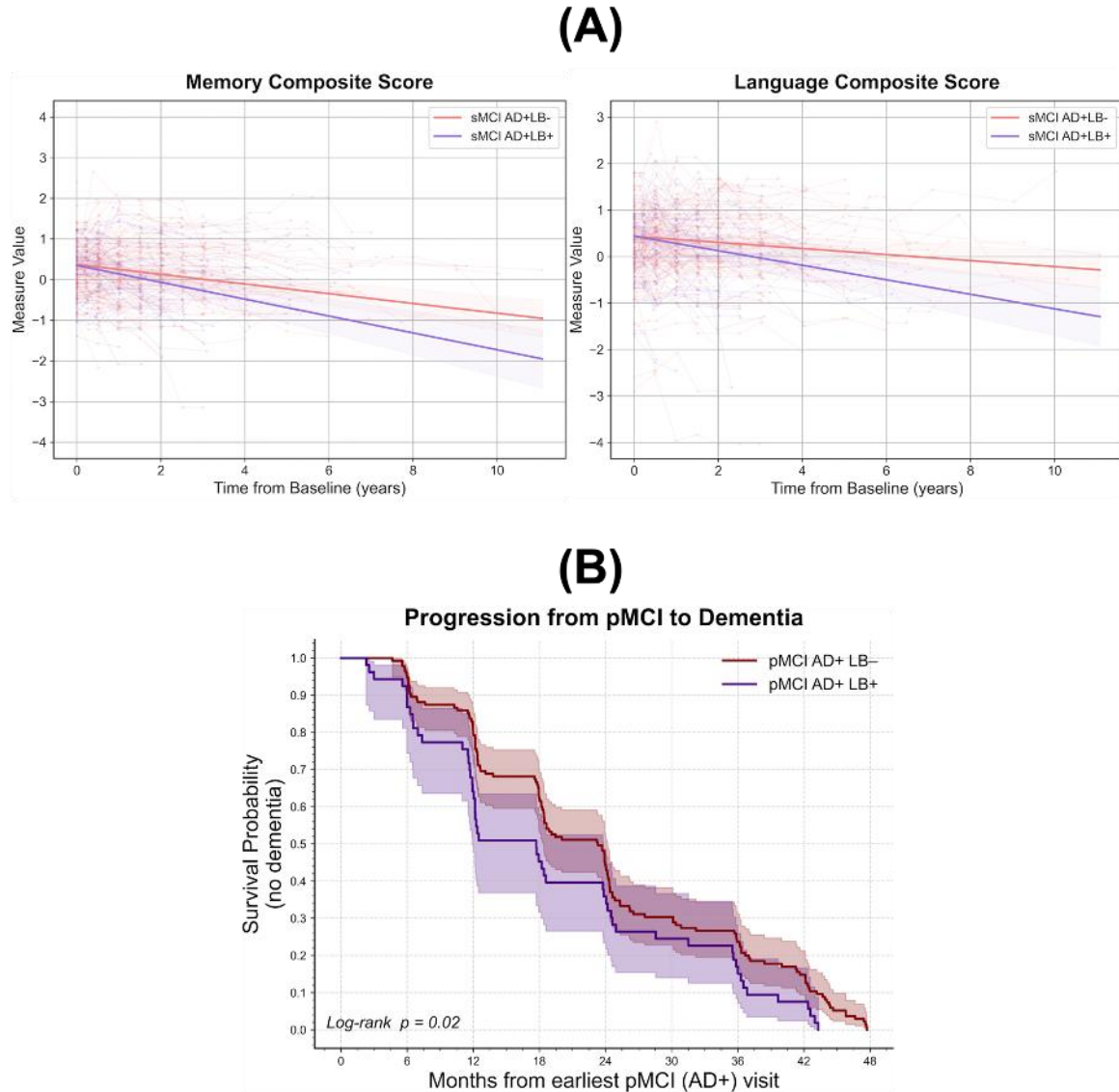


Figure 1. (A) Longitudinal memory and language composite scores for the sMCI subgroups. **(B)** Kaplan–Meier curves showing the time-to-conversion to dementia for the pMCI subgroups.

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Poster

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Program #/Poster #: PSTR267.19/D52

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 5P20GM139759
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Title: Importance of lysosomal stress responses to the pathogenesis and potential treatment of HIV-1 associated neurocognitive disorder

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Abstract: Relatively little has been published about lysosomal stress responses (LSR). LSR has been shown by others and us to be caused by a wide variety of insults including HIV-1 proteins, HIV-1 antiretroviral therapeutic drugs (ART), drugs of abuse, and weak base drugs. Approximately 75% of marketed drugs are weak base drugs that enter endolysosomes, de-acidify endolysosomes, and cause the release of divalent cations including iron from endolysosomes. Disruption in readily releasable pools of iron in endolysosomes are important because endolysosomes are “master regulators of iron homeostasis”, and neurodegeneration is linked to ferrous iron (Fe^{2+})-induced ROS generation via Fenton-like chemical reactions. Using established neural cell lines and primary cultures of neurons we conducted extensive studies of HIV-1 protein-, ART-, and weak base drug-induced morphological and functional changes in the greater lysosomal system. Pharmacologically and therapeutically relevant concentrations of these treatments induced LSR including endolysosome de-acidification, morphological changes to endolysosomes, release of endolysosome Fe^{2+} in sufficient levels to increase cytosolic and mitochondria levels of Fe^{2+} and ROS, and cause neural cell death. The HIV-1 proteins Tat and gp120, anti-retroviral therapeutic drugs including lamivudine, tenofovir disoproxil fumarate, efavirenz, nevirapine, ritonavir, nelfinavir, darunavir, and dolutegravir as well as the weak base drugs atropine (anticholinergic), azithromycin (antibiotic), fluoxetine (antidepressant), metoprolol (beta-adrenergic), and tamoxifen (anti-estrogen) caused morphological and functional characteristics of LSR including (1) changes in endolysosome morphology, numbers and distributions patterns in cells, (2) de-acidified endolysosomes, (3) decreased Fe^{2+} levels in endolysosomes, (4) increased Fe^{2+} and ROS levels in cytosol and mitochondria, (5) induced mitochondrial membrane potential depolarization, and (6) increased cell death; effects all prevented by deferoxamine. LSR causes significant changes in the morphology and function of neural cells. LSR appears to play an important role in the pathogenesis of HAND. And, weak base pharmaceuticals induce LSR that may affect their safety profiles. A better understanding of LSR and the effects of weak-base drugs on LSR as well as inter-organellar signaling may improve pharmacotherapeutics.

Disclosures: J.D. Geiger: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.20/D53

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Nanoparticle-based therapeutics for targeting neuroinflammation and amyloid pathology following traumatic brain injury in Alzheimer's disease models

Authors: *G. KARA¹, H. FLINN², M. BURKE³, S. VILLAPOL¹;

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Abstract: Alzheimer's disease (AD) is a neurodegenerative disorder characterized by the accumulation of amyloid-beta (A β) plaques, tau neurofibrillary tangles, and chronic neuroinflammation. Traumatic brain injury (TBI) is a major environmental risk factor for AD, accelerating pathological features such as A β accumulation and chronic neuroinflammation. However, effective therapeutic strategies targeting these post-injury mechanisms remain limited. In this study, we developed and tested two nanoparticle (NP)-based interventions to mitigate TBI-induced AD pathology in transgenic mouse models. Moderate TBI was induced via controlled cortical impact (CCI) injury in *APP/PS1* (27-32 weeks old) and *5xFAD* (34-38 weeks old) mice to simulate TBI. We investigated two targeted nanoparticle platforms: (1) liposomes loaded with dexamethasone (Lipo-Dex) to suppress neuroinflammation, and (2) iron oxide nanoparticles (IONPs) loaded with siRNA against β -site amyloid precursor protein cleaving enzyme 1 (BACE1), which is BACE1 siRNA-IONPs to reduce amyloidogenic processing. Dynamic Light Scattering (DLS) analysis revealed that Lipo-Dex and siRNA-IONPs had hydrodynamic sizes of 115.9 ± 4.8 nm and 174.6 ± 7.2 nm, respectively, with both formulations exhibiting a uniform size distribution. *Ex vivo* biodistribution analysis using IVIS confirmed that the NPs effectively localized to the injured brain regions following retro-orbital injection, supporting their potential for targeted post-TBI therapeutic delivery. Flow cytometry analysis revealed a significant reduction in activated B cells in the Lipo-Dex-treated group compared to Lipo-treated mice, indicating that Lipo-Dex effectively attenuates B cell-mediated immune activation following TBI. Functionally, Lipo-Dex treatment improved motor performance in female *APP/PS1* mice and enhanced recognition memory in males, compared to Lipo-treated controls at 3 and 7 days post-injury. In *5xFAD* mice, BACE1 siRNA-IONPs significantly reduced BACE1 protein expression at 7 days post-injury. These results support the therapeutic potential of NP-mediated delivery to modulate inflammation and amyloid pathology after TBI in two different genetically vulnerable models of AD. Future studies will explore the signaling pathways altered by these treatments and evaluate long-term cognitive outcomes. Together, these findings underscore the promise of NPs-based, brain-targeted strategies as precision therapeutics for TBI-exacerbated neurodegeneration in AD, providing a novel therapeutic avenue for neurodegenerative diseases.

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Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

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Program #/Poster #: PSTR267.21/D54

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Targeting UBE2K-mediated Ubiquitination activates autophagy and reduces Huntingtin aggregation: A novel therapeutic approach for Huntington's disease

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Abstract: Misfolded, toxic protein aggregates are hallmarks of many neurodegenerative disorders, including Huntington's disease (HD), where polyglutamine-expanded Huntingtin (mHtt) forms insoluble aggregates that contribute to cellular dysfunction. Leveraging the NAI Interrogative Biology Platform, an AI-driven, hypothesis-free systems medicine approach—we previously identified the E2 ubiquitin-conjugating enzyme UBE2K as a putative therapeutic target in HD. Earlier published work showed that UBE2K interacts with the Huntingtin protein, and it is known to be highly expressed in the brain. Here, we present mechanistic insights and preclinical validation supporting UBE2K inhibition as a potential disease-modifying intervention in HD. Using SH-SY5Y neuroblastoma cell lines with a stable shRNA-mediated UBE2K knockdown, we observed a drastic reduction in the accumulation of aggregated mutant Htt protein. This effect was blocked by proteasomal and lysosomal inhibitors (epoxomicin and bafilomycin A1, respectively) indicating involvement of proteostasis pathways. Proteomic and Gene Set Enrichment Analyses showed that decreasing UBE2K expression was associated with increased expression of lysosome-associated proteins, as well as an increase in expression of proteins linked to the early stages of autophagy activation and autophagosome generation. Genetic reduction of the levels of UBE2K led to significant increases in expression of p62/SQSTM1 and LC3B-II. Importantly, reducing UBE2K expression appeared to significantly enhance nuclear localization of TFEB, a master regulator of lysosomal protein synthesis and, by association, autophagy. Knowing that several pathways impinge on the control of TFEB (e.g. AMPK, mTOR, Calcineurin, calcium levels, etc.), we are currently examining the factors that change TFEB localization. To translate these findings pharmacologically, we have employed a small molecule tool compound that inhibits the ubiquitin-discharge function of UBE2K. Treatment with this compound recapitulated the biological effects of genetic UBE2K knockdown, including decreased mHtt aggregation and activation of lysosomal-autophagy pathways. Together, our data demonstrate that UBE2K regulates autophagic flux and aggregate clearance via TFEB-dependent mechanisms, and its inhibition, either genetically or

pharmacologically, may, through further investigation, potentially offer a novel strategy for therapeutic intervention in Huntington's disease.

Disclosures: **J. Lawrence:** A. Employment/Salary (full or part-time): BPGbio. **D.B. Oliver-Jolicoeur:** A. Employment/Salary (full or part-time): BPGbio. **G. Maor:** A. Employment/Salary (full or part-time): BPGbio. **J. Ranjan:** A. Employment/Salary (full or part-time): BPGbio. **T.V. Saunders:** A. Employment/Salary (full or part-time): BPGbio. **B.D. Dill:** A. Employment/Salary (full or part-time): BPGbio. **D. Chimmanamada:** F. Consulting Fees (e.g., advisory boards); Coorg Biosciences. **K.R. Nickerson:** A. Employment/Salary (full or part-time): BPGbio. **O. Zavidij:** A. Employment/Salary (full or part-time): BPGbio. **M.A. Kiebish:** A. Employment/Salary (full or part-time): BPGbio. **N.R. Narain:** A. Employment/Salary (full or part-time): BPGbio. **V.K. Vishnudas:** A. Employment/Salary (full or part-time): BPGbio. **S. Gesta:** A. Employment/Salary (full or part-time): BPGbio. **K.M. Rosen:** A. Employment/Salary (full or part-time): BPGbio.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.22/D55

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Development of novel small molecule EPHB3 inhibitors to treat neurodegenerative disease by targeting astrocyte-mediated disease mechanisms

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Abstract: Background: Neuroinflammation driven by brain glial cells has emerged as a key pathogenic mechanism in Alzheimer's disease (AD). We previously identified EPHB3 as a novel target that mediates cellular interactions between astrocytes and microglia in vivo to produce neuroinflammation and have now developed small molecule EPHB3 inhibitors with drug-like properties. We dosed these novel inhibitors in vivo to test our therapeutic hypothesis that EPHB3 inhibition will attenuate reactive astrocyte gene signatures associated with AD neuropathology, thereby decreasing neuroinflammation and rescuing associated behavioral deficits in mouse models of amyloidosis (5xFAD) and tauopathy (PS19).

Methods: We synthesized a collection of small molecule EPHB3 inhibitors and characterized them using both an in vitro kinase assay and also a cellular NanoBRET assay, both employing human EPHB3. We subsequently demonstrated PK/PD of select compounds in an acute LPS model with ICV dosing. In vivo efficacy of the top EPHB3 inhibitor that emerged from these efforts, VT-001, was further established in 14-day EAE, 2-month 5xFAD, and 3-month PS19 chronic dosing studies. Unbiased single-cell and single-nucleus RNA-seq (scRNA and snRNA-seq), proteomics, and immunofluorescence were used to assess VT-001 efficacy on astrocyte and

microglial mechanisms in total spinal cord (EAE), as well as neocortex and hippocampus (5xFAD). Relevant behavioral outcomes were monitored by clinical scoring (EAE), elevated plus maze (5xFAD, PS19), and Morris water maze (5xFAD, PS19).

Results: Our EPHB3 inhibitor VT-001 significantly rescued behavioral deficits in EAE, 5xFAD, and PS19 models, as well as attenuated neuroinflammation evoked by ICV LPS in mice. The rescue of EAE and 5xFAD behavioral deficits was associated with a robust attenuation in reactive astrocyte signature genes, neuroinflammation genes, as well as Abeta plaque-induced gene sets in astrocytes and microglia. Additionally, VT-001 was found to be well-tolerated with excellent CNS exposure across all in vivo studies. Finally, a next generation molecule, VT-002 with improved potency was identified and characterized in LPS and rat tolerability studies.

Conclusions: VT-001 emerged as a potent and selective small molecule EPHB3 inhibitor that displays striking in vivo efficacy in neuroinflammation and neurodegeneration disease models. EPHB3 inhibitors in general and VT-001 specifically, represent a highly promising opportunity for novel neurodegenerative disease therapeutics targeting astrocyte-mediated disease mechanisms.

Disclosures: **E. Lebois:** A. Employment/Salary (full or part-time): Violet Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Violet Therapeutics. **C. Kodira:** A. Employment/Salary (full or part-time): Violet Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Violet Therapeutics. **D. Schmidt:** A. Employment/Salary (full or part-time): Violet Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Violet Therapeutics.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

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Program #/Poster #: PSTR267.23/D56

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Stanley Medical Research Institute grants 03-484 and 06T-797
NIH NIMH/FIC/NCCAM grant R21MH095644
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Contract # HHSN-271- 2013-00017-C and HHSN- 271-2018-00023-C
(NIMH - PDSP)

Title: A proposal for the treatment of Huntington's disease symptoms based on phytopharmaceuticals

Authors: ***G. POLETTI**¹, C. GALLO¹, J. ALBAN², A. J. VAISBERG¹;

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Abstract: Traditional medicine knowledge can lead to the discovery of novel therapeutics for CNS disorders and help to unravel yet poorly understood biological processes behind the occurrence of these diseases. Rescuing traditional medicine practices is one of the possible avenues to identify novel pharmacological approaches for clinical conditions like Huntington's disease (HD), for which pharmacotherapies with better long-term outcomes and patient functionality are strongly needed.

Previous studies have led us to collect plants used for the treatment of CNS disorders in several Peruvian localities and geographical regions. We currently have a repository of ethanol extracts from 477 plant collections corresponding to 265 species from 87 different plant families.

Thanks to the support of the NIMH Psychoactive Drug Screening Program (PDSP) - University of North Carolina, Chapel Hill (UNC), we were able to screen these extracts for several G-Protein Coupled Receptors (GPCRs) and neurotransmitter transporters which have been linked to Huntington's disease.

The network of potential biological interactions of these proteins with Huntingtin (HTT) and its related proteins (Huntingtin Interacting Protein 1 (HIP-1), Huntingtin-associated protein 1 (HAP-1), Huntingtin interacting protein K (HYPK)), was worked out with the Search Tool for the Retrieval of Interacting Genes/Proteins (STRING) and its database of known protein interactions.

We have selected 131 extracts, having at least 3 combined activities that would be worth testing in animal models of the disease for a potential multi-target drug approach.

Disclosures: G. Poletti: None. C. Gallo: None. J. Alban: None. A.J. Vaisberg: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

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Program #/Poster #: PSTR267.24/D57

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Funding from Arvinas Operations, Inc.

Title: ARV-102, a PROteolysis TARgeting Chimera (PROTAC) leucine-rich repeat kinase 2 (LRRK2) degrader, modulates pathways associated with Parkinson's disease and progressive supranuclear palsy (PSP): proteomic analyses of cerebrospinal fluid (CSF) from non-human primates (NHPs) and healthy volunteers

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Abstract: LRRK2 is implicated in neurodegenerative diseases, including Parkinson's disease and PSP. In a phase 1 study in healthy volunteers, single and multiple oral doses of the brain-penetrant PROTAC LRRK2 degrader ARV-102 substantially reduced LRRK2 protein levels in peripheral blood mononuclear cells (PBMCs) and CSF, indicating ARV-102 induces peripheral and central LRRK2 degradation; single doses of ARV-102 reduced levels of the endo-lysosomal pathway biomarkers phospho-Rab10^{T73} in PBMCs and bis(monoacylglycerol)phosphate (BMP) in urine. Here, we explore the effects of ARV-102 on proteins downstream of LRRK2 that regulate disease-relevant pathways (eg, the endo-lysosomal system and neuroinflammation). In preclinical studies in NHPs, aptamer-based, unbiased CSF proteomic analysis was used to assess ARV-102-induced changes in LRRK2-associated proteins. A similar analysis was then performed on CSF samples from healthy volunteers in the phase 1 study. In NHPs, baseline CSF levels of the lysosomal pathway markers cathepsin B and glycoprotein non-metastatic melanoma protein B (GPNMB) and the microglia activation marker ionized calcium-binding adapter molecule 1 (IBA1) varied proportionally with baseline LRRK2 protein levels. Levels of cathepsin B, GPNMB, and IBA1 decreased proportionally following ARV-102-induced LRRK2 degradation. Proteomics in healthy volunteers were consistent with NHP data and included significant reductions in lysosomal and microglial pathway proteins that have been previously shown to be elevated in CSF in LRRK2 mutation carriers in a phenome-wide association study (PheWAS). Reductions in GPNMB and cathepsin H downstream of LRRK2 degradation were seen in healthy volunteers, with strong reduction of the microglial marker CD68, which was not observed in NHPs. Levels of other LRRK2 pathway-associated proteins previously shown by PheWAS to be elevated in LRRK2 gain-of-function mutation carriers were significantly lowered by ARV-102 in healthy volunteers, including the lysosomal regulator granulin (GRN), and the microglial regulators complement C1q tumor necrosis factor-related protein 1 (C1QTNF1) and ectonucleoside triphosphate diphosphohydrolase 1 (ENTPD1), indicating that LRRK2 degradation selectively regulates markers of pathways that link LRRK2 to neurodegenerative disease. In conclusion, the oral, brain-penetrant PROTAC LRRK2 degrader ARV-102 induced changes in neurodegenerative disease-relevant pathways regulated by LRRK2 in healthy volunteers. A phase 1 study assessing ARV-102 safety and biomarker engagement in patients with Parkinson's disease is ongoing.

Disclosures: **A.W. Hendricson:** A. Employment/Salary (full or part-time): Arvinas Operations, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas Operations, Inc. stock holder. **J.P. Corradi:** A. Employment/Salary (full or part-time): Arvinas Operations Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas Operations Inc. stock holder. **V. Guss:** A. Employment/Salary (full or part-time): Arvinas Operations Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas Operations Inc. stock holder. **K. Kelly:** A. Employment/Salary (full or part-time): Arvinas Operations, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas Operations, Inc. stock holder. **S.R. Soletsky:** A. Employment/Salary (full or part-time): Arvinas Operations, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas Operations, Inc. stock holder. **C. Polson:** A. Employment/Salary (full or part-time): Arvinas Operations, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent

holder, excluding diversified mutual funds); Arvinas Operations, Inc. stock holder. **M. MacDougall:** A. Employment/Salary (full or part-time): Arvinas, Inc (full-time). E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas, Inc. stock holder. **C. Lubeski:** A. Employment/Salary (full or part-time): Arvinas, Inc (full-time). E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas, Inc. stock holder. **L. Smits:** None. **S. Korsten:** None. **P. Kremer:** None. **A.M. Cacace:** A. Employment/Salary (full or part-time): Arvinas Operations, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Arvinas Operations, Inc. stock holder.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.25/D58

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NHMRC

Title: Pharmacological treatment of brain copper deficiency

Authors: ***A. A. LI**^{1,2}, **B. TRIST**³, **T.-T. HUNG**⁴, **M. GOTSBACHER**⁵, **C. POWER**⁴, **R. CODD**⁵, **K. DOUBLE**²;

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Abstract: Maintaining physiological levels of copper is crucial for cellular function. Imbalances in cellular copper levels lead to a range of disorders, including rare life-threatening genetic diseases such as Menkes disease and Wilson's disease, and common neurodegenerative disorders, such as Parkinson disease (PD) and amyotrophic lateral sclerosis (ALS). Copper dyshomeostasis represents an early change in PD and ALS, with restoration of physiological copper levels in the central nervous system (CNS) proposed to slow disease progression. The copper-containing ionophore diacetylbis(N(4)-methylthiosemicarbazonato)copper(II) (CuATSM) delivers copper to the CNS and may restore physiological brain copper levels in these, and other, disorders characterised by CNS copper deficiency. However, the dosage and treatment frequency required to achieve CNS copper restoration using CuATSM is unknown. We employed orally treated wilytype C57BL/6 mice and ctrl1+/- mice with 8 mg/kg of CuATSM or vehicle for either 10, 15 or 21 days. Treatments were performed daily, or 4 days per week, using micropipette-guided drug administration. Copper levels in the cerebellum, cortex, brainstem, deep brain nuclei, serum,

muscle, and liver of CuATSM-treated Ctr1^{+/-} mice were compared with vehicle treated Ctr1^{+/-} mice and wild-type C57BL6 mice. Via two way ANOVA test, we confirmed vehicle treated Ctr1^{+/-} mice have significantly lower levels of copper compared to wild-type C57BL6 mice in all CNS regions studied. Twenty-one days of 8 mg/kg daily CuATSM treatment restored Ctr1^{+/-} mice copper levels in all CNS regions studied to equivalent levels observed in wild-type mice. Data from this study could inform the design of copper supplementation in preclinical research and clinical trials.

Disclosures: A.A. Li: None. B. Trist: None. T. Hung: None. M. Gotsbacher: None. C. Power: None. R. Codd: None. K. Double: None.

Poster

PSTR267: Other Dementias, Proteinopathies, and Pathologies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR267.26/D59

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NS085770
DC008552
AG13854
R01AG091388

Title: Differential Loss of Nuclear TDP-43 in Pathological Subtypes of TDP-43 Proteinopathy

Authors: *I. A. AYALA¹, C. NELSON¹, T. GEFEN³, M.-M. MESULAM², C. GEULA⁴; ²Northeastern Univ., ¹Northwestern Univ., Chicago, IL; ³Cognitive Neurol. and Alzheimer's Dis. Ctr., Feinberg Sch. of Medicine, Northwestern Univ., Chicago, IL; ⁴Mesulam Cent for Cogn Neurol & Alzhei Dis, Northwestern Univ. Med. Sch., Chicago, IL

Abstract: A major question in relation to misfolded proteins deposited in neurodegenerative disorders is whether the consequences of the pathology are due to loss of the normal function of the deposited proteins or a toxic gain of function of misfolded proteins. In relation to TDP-43 proteinopathies, emerging evidence suggests that TDP-43 is lost from its normal nuclear localization, potentially resulting in loss of its many normal functions. TDP-43 proteinopathies can be classified under 3 main groups, TDP-43 type A which is characterized by inclusions predominantly in superficial cortical layers and short neurites, type B with more granular inclusions in both superficial and deep cortical layers, and type C characterized pathologically by long neurites, primarily in superficial cortical layers. This project investigates the incidence and frequency of loss of normal TDP-43 from neuronal nuclei in these 3 subtypes. Immunohistochemical techniques were implemented in a preliminary study to determine the presence or loss of nuclear wild-type TDP-43 in 6 participants (2 type-A, 1 type-B, 2 type-C, and 1 control). An antibody to wild-type human TDP-43 (Monoclonal AbNova TARDBP Mouse anti-human, 1:5000) was used to visualize the protein in 40 µm frozen sections of the inferior

frontal gyrus. Neurons with and without nuclear TDP-43 were counted using unbiased stereological techniques. The density of neurons without nuclear TDP-43 immunoreactivity, and the proportion of the density of such neurons as a ratio of total neurons, were calculated. The control participant displayed the lowest number and proportion of total neurons without nuclear TDP-43. In contrast, the density of neurons that displayed loss of nuclear TDP-43 was greater than control in all TDP-43 subtypes. The highest density and proportion of total density of neurons that had lost nuclear TDP-43 occurred in type-A participants (nearly 30% of total neuron density; 10 fold higher compared with control). In contrast, the lowest density in any participant with TDP-43 pathology was in a type-C. Overall, 15% of total density of neurons in type-C had lost their nuclear TDP-43. The density of such neurons in the type-B participant was intermediate, between type A and type-C. These preliminary results confirm loss of nuclear TDP-43 in participants with inclusions. They also indicate that participants with different subtypes of inclusions may display differential loss of nuclear TDP-43, with type-C displaying the lowest density of such neurons. Additional participants with TDP-34 proteinopathy and controls will be analyzed to confirm these preliminary findings.

Disclosures: I.A. Ayala: None. C. Nelson: None. T. Gefen: None. M. Mesulam: None. C. Geula: None.

Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.01/D60

Topic: C.03. Parkinson's Disease

Support: JST Moonshot R&D (JPMJMS2012)
JSPS KAKENHI 23K19933
JSPS KAKENHI 25K19984

Title: Beta burst duration in EEG reflects bradykinesia severity in Parkinson's Disease

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Abstract: Parkinson's disease (PD) is a neurodegenerative disorder characterized by the progressive loss of dopaminergic neurons, leading to hallmark motor symptoms such as bradykinesia, rigidity, and tremor. Early diagnosis remains challenging, as symptoms typically emerge only after significant neuronal loss. Recent research has focused on identifying neural biomarkers to aid in diagnosis and guide interventions like adaptive deep brain stimulation (DBS) and neurofeedback training (NFT). While studies using local field potentials from deep brain structures have shown that exaggerated beta-band activity correlates with motor symptom

severity, evidence for similar biomarkers using non-invasive techniques such as electroencephalography (EEG) is limited. To address this gap, we collected EEG data from 15 PD patients (mean age: 71 ± 10 years; 9 females; mean Movement Disorder Society-Unified Parkinson's Disease Rating Scale [MDS-UPDRS] Part III score: 34 ± 17) while on their regular levodopa medication (ON). A subset of 7 patients also underwent EEG recording while off levodopa (OFF; mean time since last administration 7 ± 5 hours). Recordings were obtained from sensorimotor regions under three conditions: resting, spiral drawing, and finger tapping. Disease severity was assessed by movement disorder specialists using the MDS-UPDRS. The symptom-dominant side was identified as the one with higher lateralized scores on Part III, and beta bursts were identified from EEG signals recorded over the ipsilateral hemisphere using the Fitting Oscillations and One-Over-F-based Burst Detection method (Seymour et al., 2022). Results revealed that the average duration of detected beta bursts during finger tapping of the symptom-dominant hand was significantly correlated with MDS-UPDRS Part III scores (Spearman $\rho = 0.741$, $p = 0.004$) and bradykinesia subscores (Spearman $\rho = 0.666$, $p = 0.011$) during the ON state, but not with other subscores or Parts I and II. These findings suggest that non-invasive EEG can capture abnormal neural activity associated with PD motor symptoms, particularly prolonged beta bursts related to bradykinesia during movement. This supports the potential of beta burst duration as a non-invasive, EEG-based biomarker for PD, forming a basis for the development of NFT protocols. Moreover, such biomarkers may enable more accurate diagnoses and personalized management strategies for individuals with PD.

Disclosures: **K. Adachi:** None. **S. Okusa:** None. **H. Saegusa:** None. **T. Watanabe:** None. **S. Iwama:** None. **M. Seki:** None. **J. Ushiba:** A. Employment/Salary (full or part-time): LIFESCAPES Inc..

Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.02/D61

Topic: C.03. Parkinson's Disease

Support: Korea Institute of Science and Technology Intramural Program (2E33701) K-brain Project (RS-2023-00262880) of National Research Foundation funded by the Ministry of Science and ICT, Korea

Title: In silico model incorporating topographic cortico-basal ganglia connectivity to uncover the comorbidity of motor and non-motor symptoms in Parkinson's disease

Authors: ***J. KIM**, T. KIM;

Brain Sci. Inst., Korea Inst. of Sci. and Technol., Seoul, Korea, Republic of

Abstract: The basal ganglia (BG) processes diverse inputs from motor and non-motor regions of the cerebral cortex via topographically organized cortico-striatal projections. While previous

research has focused primarily on fragmentary aspects of the BG feedback loop to the cortex via the thalamus, it has often overlooked the wide-range topographical organization within the BG and its direct influence on downstream targets. Considering that Parkinson's disease (PD) often accompanies non-motor symptoms such as executive dysfunction, in addition to the canonical motor symptoms, topographical organization of neuronal arrangements within each subnucleus in BG and connectivity between cortex and BG could underlie the comorbidity of distinct symptoms. To examine this hypothesis, we developed a computational spiking neural network (SNN) models of the cortico-basal ganglia circuit that explicitly incorporates topographic connectivity as well as the biologically plausible neuronal population ratio between subnuclei, dopaminergic projections, and other vital aspects of neuronal and synaptic model. We utilized various integrate-and-fire models that have been adapted to fit the firing characteristics of each neuronal type. First, we could successfully reconstruct the firing patterns and their changes by dopamine depletion in accord with the classical model explaining motor symptoms, referred as imbalance hypothesis between direct and indirect pathways. With the model incorporating topographic organization, we observed the emergence and propagation of pathological firing from the motor-related cortex-BG loop to other modality-specific loops. To this end, we seek to suggest the potential mechanism and the requisite of comorbidity of symptoms in PD using our SNN in silico model of cortex-BG.

Disclosures: J. Kim: None. T. Kim: None.

Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.03/D62

Topic: C.03. Parkinson's Disease

Title: Identifying region-specific imaging biomarkers for Parkinson's disease using T1-weighted MRI and machine learning approach

Authors: *R. TAI^{1,2}, J.-D. ZHU^{3,4};

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Abstract: Subtle Parkinsonian features in early Parkinson's disease (PD) often hinder accurate diagnosis. Although structural MRI is a tool for PD evaluation, the lack of specific alterations limits its diagnostic utility. Several MRI studies have employed machine learning approaches to classify PD patients from healthy controls (HCs). However, most classification models in these studies were built using whole-brain data, restricting the ability to examine region-specific contributions. This study aimed to construct classification models for individual brain regions to improve classification accuracy between PD and HC groups and to identify critical regions that

may serve as potential biomarkers for PD. We recruited data from the Parkinson's Progression Markers Initiative database, comprising 226 PD patients and 211 sex- and age-matched HCs. The dataset was randomly split into training and test datasets at a 7:3 ratio. Following T1-weighted MRI preprocessing, we built 116 classification models based on each brain region using the gray matter density map and automated anatomical labeling atlas within the training dataset. Feature selection was conducted using elastic net regression with alpha value set to 0.5, followed by five-fold cross-validation across multiple classification algorithms. The trained models were then applied to the test dataset. All procedures were repeated ten times. To identify the most relevant brain regions in PD patients, we selected the top ten brain regions ranked by mean test accuracy. Finally, we performed multiple linear regression using the gray matter volumes of the selected regions as predictors and the Unified Parkinson's Disease Rating Scale (UPDRS) Part III total score as the dependent variable to assess whether structural alterations in specific brain regions were related to motor symptom severity in PD. The results indicated that the logistic regression algorithm yielded the highest classification performance, with the mean test accuracies of the top ten brain regions ranging from 75.57% to 88.69%. Among these regions, only the medial part of the left superior frontal gyrus showed a significant negative correlation with the UPDRS Part III total score ($\beta = -2.712$, $p = 0.033$). Through the delicate MRI-based machine learning classification methods, ten crucial brain regions associated with Parkinson's disease were identified. Furthermore, one of these regions exhibited a significant negative correlation with UPDRS Part III scores, underscoring its potential as an early image biomarker for motor symptom severity in Parkinson's disease. Future studies should incorporate multi-center data to enable external validation.

Disclosures: R. Tai: None. J. Zhu: None.

Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.04/D63

Topic: C.03. Parkinson's Disease

Title: Evidence for sleep mediated homeostasis failure in levodopa induced dyskinesia

Authors: *G. LOMBARDI¹, L. FIORILLO^{2,3}, N. LA PORTA⁴, L. ARNAUD³, A. CASTELNOVO^{3,5}, A. KAEIN-LANG^{3,6}, S. GALATI^{3,5};

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Abstract: Introduction. Slow-wave activity (SWA) during slow-wave sleep (SWS) facilitates synaptic downscaling, crucial for brain plasticity. SWA, typically occurring early in the night, serves as a primary electrophysiological marker for the homeostatic process. Conversely, theta activity during wakefulness indicates synaptic potentiation, with SWA decreasing during sleep and theta increasing during wakefulness. **Objective.** Levodopa-induced dyskinesia (LID) in Parkinson's disease (PD) is linked to impaired synaptic homeostasis, but whether this stems from defective downscaling, impairing wake-related synaptic buildup, remains unclear. This study mainly focuses on analyzing theta activity during wakefulness to investigate the build-up process in resting state. **Methods.** We compared actigraphy and high-density EEG in 12 healthy volunteers and three PD cohorts: de novo (n=12), advanced non-dyskinetic (n=13), and dyskinetic (n=11). Participants underwent one-week actigraphy monitoring (i.e., GeneActiv sensors, with sleep metrics extracted via GGIR algorithms) and morning/evening resting-state EEG. Theta power (4-8 Hz) were analyzed using both linear models (LM) and linear mixed-effects models (LMM), adjusted for confounders (e.g., age, gender, LEDD and disease duration). To validate the spatial distribution of significant effects, we applied non-parametric permutation tests combined with Threshold-Free Cluster Enhancement (TFCE). We finally assessed associations between theta and clinical variables (LEDD, DD) via partial correlation. **Results.** DYS patients exhibited reduced actigraphy-estimated sleep efficiency ($p<0.001$) and prolonged sleep latency ($p<0.05$) versus controls. Theta power was higher in dyskinetic patients across all cortical regions during morning sessions (LM-TFCE-corrected $p<0.05$), with absent diurnal scaling across prefrontal, temporal, and occipital regions (LMM-TFCE-corrected $p<0.05$) - contrasting CTL and non-dyskinetic groups showing progressive theta accumulation. While LEDD did not differ significantly between ADV and DYS, higher LEDD was associated with increased theta only in DYS. **Conclusion.** These findings demonstrate for the first time a bidirectional synaptic homeostasis failure in LID: impaired SWA-mediated downscaling coexists with saturated morning theta activity, indicating defective wake-related synaptic potentiation. Our results highlight sleep modulation as a therapeutic priority. Enhancing slow-wave sleep, can we restore the synaptic downscaling, and potentially break the cycle of theta saturation and dyskinesia initiation and/or progression?

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.05/D64

Topic: C.03. Parkinson's Disease

Support: NINDS UH3NS103468
UH3NS129898

Title: Beta phase-locking value, not beta amplitude, best encodes bradykinesia in Parkinson's disease.

Authors: *S. L. SCHMIDT¹, R. RODRIGUEZ CAPILLA⁴, H.-J. LEE^{1,2}, J. PETERS¹, D. A. TURNER⁴, W. M. GRILL³;

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Abstract: Deep brain stimulation (DBS) is an effective treatment for the motor symptoms of Parkinson's disease. Because DBS does not entirely replace medication, the effects of continuous (c)DBS are limited by the onset of side effects during the peak of the medication cycle. Subsequently, when medication washes out, patients receive less DBS than is optimal for symptom reduction. Adaptive DBS aims to improve upon cDBS by adapting to patients' symptoms across the medication cycle. To do this effectively, a biomarker that tracks symptoms, rather than tracking the level of DBS, is required. We conducted chronic local field potential (LFP) recordings of the subthalamic nucleus (STN) and globus pallidus (GP). We examined the utility of amplitude of beta oscillations in the STN (STN Amp), and two measures of intrahemispheric coherence: phase-locking value (PLV) and magnitude squared coherence (MS Coh) between the STN and GP in the beta band as biomarkers. We conducted a series of cDBS experiments in a cohort of 6 participants receiving dual-target DBS. All study activities were approved by the FDA and Duke IRB, and all participants provided informed consent. We assessed the suitability of the three biomarkers by their relation to hand grasp speed during 300 s experiments with graded levels of cDBS amplitudes. We observed that intrahemispheric STN Amp, PLV, and MS Coh all correlated with hand grasp speed. After accounting for the effect of DBS amplitude in a linear mixed effects model, all three metrics remained significant predictors of grasp speed (median effect size [95% CI]: -0.0392 [-0.0447 to -0.0347] Hz/dBmV for STN Amp, -0.701 [-0.778 to -0.598] Hz for PLV, and -0.617 [-0.693 to -0.484] Hz for MS Coh). These significant effect sizes suggest each feature of the beta LFP was a biomarker for symptoms. Mediation analysis revealed that PLV mediated a large fraction of the total change in STN Amp and MS Coh due to DBS (median proportional effect: 0.319 [0.309 to 0.327] for STN Amp and 0.47 [0.474 to 0.483] for MS Coh). Because STN Amp and MS Coh encoded akinetic information, this finding supports communication through coherence within the basal ganglia. We applied mediation analysis to determine the extent to which each biomarker mediated the effect of DBS on grasp speed. PLV mediated significantly more of the impact of DBS than STN Amp or MS Coh (median proportional effect: -0.0675 [0.0508 to 0.0744] for STN Amp, 0.128 [0.114 to 0.140] for PLV, and 0.0634 [0.0542 to 0.0707] for MS Coh). The larger effect size of PLV compared to STN Amp and MS Coh, combined with the concept of communication through coherence, demonstrates for the first time in humans that the information lesion is a mechanism of DBS for PD.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.06/E1

Topic: C.03. Parkinson's Disease

Support: KID grant, Karolinska Institutet
ASAP-020370

Title: Anxa1+ dopamine neuron vulnerability defines prodromal Parkinson's disease bradykinesia.

Authors: *V. SKARA¹, I. MANTAS², A. CONTESTABILE³, K. MELETIS³;
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Abstract: Progressive degeneration of dopamine neurons (DANs) is a defining feature of Parkinson's disease (PD), yet the identity and role of the most vulnerable DAN populations during the prodromal phase remain poorly understood. Here, we identify a subpopulation of substantia nigra DANs expressing Annexin A1 (Anxa1) as selectively vulnerable across multiple prodromal PD models and significantly reduced in patient- derived DANs. Anxa1+ DANs display a distinct functional profile that does not encode for reward or drive reinforcing and motivated behaviors. Instead, activity of Anxa1+ DAN axons correlates with vigorous movements during self-paced exploration. Silencing these neurons disrupts specific action sequences that recapitulate the PD bradykinesia profile. These findings highlight Anxa1+ DANs as an early-vulnerable population whose function may underlie prodromal motor deficits in PD.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

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Program #/Poster #: PSTR268.07/E2

Topic: C.03. Parkinson's Disease

Support: APDA P09
APDA 1053521
Udall Parkinson's Foundation Research Center of Excellence
NIH (NINDS) T32 NS115724

Title: Deep brain stimulation local evoked potentials as state-independent biomarkers for precision subthalamic nucleus targeting

Authors: *E. BENCE¹, E. GADZIC¹, W. ENGELHARDT⁴, N. MANTENA¹, S. MIOCINOVIC⁶, P. BRUNNER⁷, J. T. WILLIE⁵, A. HESTON², K. CHOU², E. L. LEVIN⁸, D.

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Abstract: Despite deep brain stimulation's (DBS) efficacy for Parkinson's disease (PD), optimal subthalamic nucleus (STN) targeting remains challenging. DBS local evoked potentials (DLEP) may address these limitations by providing objective biomarkers for targeting and therapeutic outcomes. In 16 PD patients undergoing STN-DBS implantation (all recorded awake, with 12 also studied under anesthesia), we recorded local field potentials at rest and during 130 and 11 Hz stimulation. DLEP amplitudes were quantified as RMS envelopes (5-20 ms post-stimulation, artifact-corrected). Beta power (13-35 Hz) was measured within a 5 Hz bandwidth centered at the electrodes' peak beta frequency. Using Lead-DBS-derived coordinates, we modeled the 3D spatial distributions of DLEPs and beta oscillations in MNI space via linear mixed-effects models incorporating quadratic terms and subject/hemisphere random effects. To evaluate the potential link between these distributions to functional neuroanatomy and clinical outcomes, we assessed spatial concordance between DLEP amplitudes and intraoperative MER-defined STN boundaries, and compared maximal-amplitude contact locations to postoperative therapeutic stimulation thresholds. Quadratic models significantly outperformed linear fits for both biomarkers ($p < 0.01$), with x and z coordinates showing strong influence on spatial distributions ($p < 0.05$), confirming non-linear 3D organization. Awake patients exhibited stronger DLEPs for intra-STN versus above-STN stimulation across frequencies ($p < 0.05$), while anesthetized patients showed this differentiation only for dorsal contacts ($p < 0.01$). DLEP-maximizing contacts exhibited excellent agreement with therapeutic electrodes (82-90% concordance, Cohen's $\kappa > 0.7$), outperforming beta power (50-65% agreement, Cohen's $\kappa < 0.5$). These results demonstrate that DLEPs provide precise, state-independent spatial localization within the STN that strongly correlates with both microelectrode recording-defined anatomy and clinically effective stimulation sites. The robust performance of DLEPs across physiological states suggests they could serve as valuable biomarkers for optimizing both intraoperative targeting and postoperative programming in STN-DBS for PD.

Disclosures: E. Bence: None. E. Gadzik: None. W. Engelhardt: None. N. Mantena: None. S. Miocinovic: None. P. Brunner: None. J.T. Willie: None. A. Heston: None. K. Chou: None. E.L. Levin: None. D.K. Leventhal: None. E. Opri: None.

Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.08/E3

Topic: C.03. Parkinson's Disease

Support: American Academy of Neurology
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Stanford Knight Initiative for Brain Resilience
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Department of Defense Vannevar Bush Faculty Fellowship

Title: Striatal Neural Ensemble Codes for Normal, Parkinsonian, and Dyskinetic Movements

Authors: *G. CHATTREE^{1,2,3}, R. CHRAPKIEWICZ^{1,3}, Y. ZHANG^{1,3,4}, J. LI^{1,3,5}, F. DINC⁶, M. J. SCHNITZER^{1,3,4,5,7};

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Abstract: Classical models of movement control posit that striatal spiny projection neurons of the basal ganglia's direct and indirect pathways (dSPNs and iSPNs) respectively promote and suppress movement. Supporting this view, physiological recordings have revealed imbalanced dSPN and iSPN activity levels during hypokinetic and hyperkinetic movement conditions. However, in normal brain states, dSPN and iSPN ensembles have approximately equal activation amplitudes and time courses, jointly encoding specific actions. How pathological movement conditions alter such action coding remains poorly understood. Here we imaged the concurrent dynamics of dSPNs and iSPNs in behaving mice across normal, parkinsonian (hypokinetic), and dyskinetic (hyperkinetic) conditions, before and after administration of drug treatments used clinically. Analyses focused on rest periods and those immediately preceding movement, examining how SPNs encoded upcoming actions. In parkinsonian states, the dSPN population was hypoactive relative to the iSPNs, consistent with prior reports. Further, individual dSPNs and iSPNs that encoded upcoming locomotion exhibited a reduced measure of activity compared to the normal state; the extent of this reduction predicted the degree of decline in the occurrence of locomotion. Levodopa (L-DOPA) and amantadine treatments both improved locomotion frequency but acted via distinct mechanisms. L-DOPA rebalanced the activity of the dSPN and iSPN populations, whereas amantadine boosted the activity of individual locomotion-related dSPNs and iSPNs. In hyperkinetic states modeling L-DOPA-induced dyskinesia, dSPN populations were hyperactive relative to iSPNs. Involuntary dyskinetic movements engaged individual dSPNs and iSPNs distinct from those encoding voluntary locomotion. Amantadine treatment reduced the resting activity of dyskinesia- but not locomotion-related SPNs without improving the overall dSPN and iSPN imbalance. These findings highlight the importance of SPN action coding, not merely the extent of activity balance, for normal and pathological movements. The results delineate two distinct therapeutic mechanisms, one that rebalances the activity of the direct and indirect pathways and another that selectively potentiates or depresses the activity of SPN populations encoding voluntary or involuntary actions. Overall, this study refines the understanding of striatal dysfunction in movement disorders, demonstrates that distinct neural populations underlie normal voluntary locomotion and involuntary dyskinetic movements, and defines two complementary routes for the development of symptomatic treatments.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR268.09/E4

Topic: C.03. Parkinson's Disease

Support: PAPIIT IN 205023
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SECIHTI 1103623

Title: Parkinsonism induced by acute blockade of dopamine receptors or by a 6-OHDA lesion in the Substantia Nigra pars compacta leads to alterations in cortical oscillations and network dynamics

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Abstract: The primary motor cortex (M1) plays a central role in motor planning and execution, integrating inputs from premotor regions and projecting glutamatergic outputs to the basal ganglia (BG) and spinal cord. In Parkinson's disease (PD), dopaminergic depletion in the BG leads to the emergence of beta-band oscillations (13-40 Hz), typically interpreted as pathological synchronization. Here, we used a hemiparkinsonian mouse model induced by unilateral 6-hydroxydopamine (6-OHDA) lesion of the substantia nigra pars compacta (SNc), which spares the ventral tegmental area (VTA) and thus preserves dopaminergic modulation in the cortex. Brain slices containing M1 were recorded in vitro using microelectrode arrays (MEAs), allowing simultaneous analysis of extracellular action potentials and local field potentials (LFPs). We show that DA depletion restricted to the BG is sufficient to induce aberrant beta synchronization in cortical circuits in the absence of intact BG-thalamo-cortical connectivity. Beyond spectral changes, analysis of spiking activity revealed reorganized neuronal ensembles and altered network dynamics. Using recurrence analysis and network topology metrics, we observed that M1 dynamics under DA depletion deviated from criticality: although recurrent structures persisted, the system exhibited increased determinism and reduced trajectory divergence, consistent with a shift away from flexible, metastable dynamics. These features were reversed by acute L-DOPA application. Comparisons with DA receptor antagonism revealed distinct patterns of beta synchronization and ensemble reconfiguration. Our findings suggest that cortical beta

oscillations can arise independently of subcortical loops, and that dopamine loss in the BG alone alters cortical circuit dynamics, pushing them away from criticality.

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Poster

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Topic: C.03. Parkinson's Disease

Support: R01NS037019
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MnDrive Postdoctoral Fellowship

Title: Neural population dynamics in Parkinsonian motor cortex during reaching

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Abstract: Bradykinesia, one of the cardinal motor signs of Parkinson's disease (PD), is characterized by slowness of movement. While the primary motor cortex (M1) has been implicated in bradykinesia, prior studies focusing on single-neuron activity have yielded inconsistent findings and failed to account for the considerable trial-by-trial variability in motor behavior. In the present study, we investigated latent neural dynamics within M1 by recording population-level neuronal activity in non-human primates during both naive and parkinsonian states. By applying Principal Component Analysis (PCA), we identified that slow-varying dynamical motifs (i.e., monotonic and monophasic) remained preserved in the PD state despite the presence of bradykinesia. Canonical correlation analysis (CCA) on shared neural modes between naive and PD states further supports the preservation of these slow-varying neural dynamics. However, we observed a coordinated oscillatory mode (COM) in the low-beta frequency band (10-20 Hz) that emerged during reach in the parkinsonian state but was absent at rest, suggesting a functional role in movement execution. To investigate this role, we developed a neural overlap index by quantifying the angles between single-trial COM and the invariant monotonic dynamics. We found that the neural overlap index positively correlated with reach duration. The positive correlation between the neural overlap index and reach duration suggests a functional role for invariant monotonic dynamics and a pathological role for the COM. Our findings support the coexistence of preserved and pathological neural dynamics in M1 in the

parkinsonian condition and provide novel insights into the neural mechanisms underlying bradykinesia.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

Location: SDCC Halls B-H

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Program #/Poster #: PSTR268.11/E6

Topic: C.03. Parkinson's Disease

Support: Th Red Gates Foundation

Title: Dopaminergic signaling, oxidative stress and antioxidant defense in striatal vs. cortical DA target neurons

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Abstract: In Parkinson's Disease (PD), basal ganglia vs. cerebral cortical dysfunction have different onsets—earlier vs. later—and distinct endpoints—paucity of movement vs. cognitive decline. We asked whether these differences reflect divergent DA signaling, mitochondrial function, and antioxidant defense in striatal vs. cerebral cortical DA-target neurons. We analyzed these differences in typical aging mice and those with a heterozygous deletion orthologous to that in human 22q11.2 Deletion Syndrome, which recent evidence suggests accounts for a 5-to-6-fold increase in PD diagnosis. Using spatial transcriptomics, we found distinct sets of mitochondrial and antioxidant defense genes expressed in Darpp-32+ striatal vs. cortical neurons in typical aging mice, subsets of which are differentially expressed in striatal vs. cortical DA-target neurons in aging 22q11-deleted mice. These differences are paralleled by disparate densities of DA innervation for striatal (high) vs. cortical (lower) DA-target neurons within and between genotypes. Divergent antioxidant defense signatures for densely vs. sparsely DA-innervated striatal vs. cortical neurons are paralleled by distinct levels of striatal vs. cortical DA release, detected using electrodes for which DA sensitivity is determined a priori using concentration-based machine learning models. Transiently increased DA is accompanied by

transiently increase reactive oxygen species (ROS, measured as H₂O₂ using parallel individual electrode models), suggesting that DA transmission increases local oxidative stress. Both DA release and transient ROS are increased in 22q11-deleted mice, suggesting that increased oxidative stress may contribute to PD risk. Thus, divergent PD motor vs. cognitive pathology may reflect altered capacity of striatal vs. cortical DA-target neurons to manage oxidative stress that accompanies varying DA innervation, transmitter release and signaling. Distinct antioxidant defense signatures in striatal vs. cortical DA target neurons may provide novel targets to modulate motor vs. cognitive decline in individuals with PD. Finally, our results suggest that 22q11-deleted mice provide a valid model for understanding PD pathogenesis.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Topic: C.03. Parkinson's Disease

Support: 1R01NS122226-01

Title: Prophylactic effects of genetic knockdown of the serotonin transporter on L-DOPA-induced behaviors and striatal organization in a rat model of Parkinson's disease

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Abstract: Parkinson's disease (PD) is caused by the loss of dopamine (DA) neurons in the substantia nigra pars compacta (SNpc) resulting in symptoms such as resting tremor, rigidity, akinesia, and postural instability. DA replacement therapy with L-3,4-dihydroxyphenylalanine (L-DOPA) is highly effective at alleviating the hypokinetic motor symptoms of PD in early stages of the disease. Unfortunately, the majority of PD patients will develop treatment-induced side effects typified by choric abnormal involuntary movements (AIMs) in a syndrome termed L-DOPA-induced dyskinesia (LID). Though the specific mechanisms underlying the development and expression of LID remain enigmatic, accumulating research has implicated aberrant neuroplasticity within raphe-striatal serotonin (5-HT) circuits. While this compensatory neuroplasticity may support L-DOPA's initial motor benefit, continued 5-HT gain-of-function through later stages of PD may lead to the hyperinnervation of raphe-striatal circuits, which precipitate LID. The 5-HT transporter (SERT) has been identified as an attractive antidyskinetic candidate as it demonstrates affinity for DA uptake and is upregulated in the brains of dyskinetic patients and in animal models of LID. Preclinical studies have supported beneficial effects of SERT modulation with selective serotonin reuptake inhibitors (SSRIs) and genetic SERT knock-down (SERT-KD). However, it is possible that modulating SERT may also prevent or blunt the

emergence of LID. The current study employs a validated genetic SERT-KD to investigate its role in LID development. Adult male and female 6-hydroxydopamine-lesioned Sprague-Dawley rats received infusions either adeno-associated virus 9 (AAV9) short-hairpin RNA driven SERT-KD (SERT-shRNA) or control scrambled shRNA AAV9-GFP (SCR-shRNA) into the dorsal raphe nucleus (DRN). Rats were then evaluated for motor performance and LID development across 21 days of sub-chronic L-DOPA treatment. We hypothesized that this genetic intervention would attenuate the development of LID by limiting the gain-of-function and associated aberrant neuroplasticity within raphe-striatal circuits. Behavioral data showed a marked reduction in the severity of LID development in SERT-KD animals compared to SCR-shRNA controls. This suggests that a prophylactic SERT-KD may inhibit the reorganization of pro-dyskinetic circuitry, which buttresses support for SERT as an important mechanism in the development of LID. This offers a promising avenue for developing novel therapies to optimize PD treatment.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Program #/Poster #: PSTR268.13/E8

Topic: C.03. Parkinson's Disease

Title: Influence of 5-HT Circuitry on Visual Prepulse Inhibition in a Unilateral Parkinsonian Rat Model

Authors: H. PRISINZANO¹, *S. VENKATESH¹, N. LIPARI¹, G. MCMANUS¹, J. NARDIELLO¹, C. R. BISHOP²;

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Abstract: Parkinson's Disease (PD) is a progressive neurodegenerative disorder characterized by dopamine (DA) depletion in the substantia nigra pars compacta (SNc). Along with characteristic motor deficits, PD often presents with debilitating non-motor symptoms (NMS). One such NMS is PD-associated psychosis (PDAP), which can include illusions, delusions, and visual hallucinations that contribute significantly to caregiver burden and increased institutionalization. Visual hallucinations are one of the most prevalent and detrimental symptoms experienced by PDAP patients, however, the mechanisms behind this condition are not fully understood. One proposed mechanism underlying PDAP is cortical serotonergic dysregulation, particularly the upregulation of 5-HT_{2A} receptors (5-HT_{2A}R). To investigate this proposed mechanism, the present study utilized a novel visual-auditory prepulse inhibition (PPI) assay. PPI measures sensorimotor gating, a commonly disrupted feature in psychiatric conditions and one with close association to hallucinations. In Experiment 1, unilateral 6-hydroxydopamine lesioned and sham rats were assessed for differences in sensorimotor gating function through PPI. Experiment 2 used the same multimodal PPI assay to investigate the role of 5-HT in the expression of PDAP following acute administration of a partial 5-HT_{2A}/2CR agonist, $\pm$ 2,5-

Dimethoxy-4-iodoamphetamine hydrochloride (DOI). Findings revealed that 6-OHDA-lesions did not change PPI compared to non-lesioned animals. However, DA-lesioned rats displayed sensorimotor gating deficits when administered DOI compared to shams. This was most pronounced at low doses (0.25 mg/kg) while the higher dose (1mg/kg) resulted in disruption in PPI at shorter prepulse intervals. In addition to PPI, the Head Twitch Response (HTR) and rearing tests were used to compare differences in 5-HT_{2A}R activation specific to the medial prefrontal cortex (mPFC). No significant differences were found between sham and DA-lesioned animals from these behavioral proxies, suggesting a distinction between PPI and HTR in 5-HT circuitry. These PPI findings are the first to model and validate maladaptive upregulation of 5-HT_{2A}R occurring in the striatum, prefrontal and temporal cortices, that are commonly associated with visual sensory processing and PD. This innovative visual paradigm not only deepens our understanding of the nonmotor features of PD but also opens avenues for developing therapeutic strategies for patients suffering from visual hallucinations while addressing a critical gap in current research.

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Poster

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Topic: C.03. Parkinson's Disease

Support: NIH Grant R01NS058487

Title: Functional task-based connectivity in Rapid Eye Movement Behavior Disorder

Authors: *J. P. SIMPSON¹, E. R. TOBIN⁵, X. LOU², M. PRESTI³, A. KRAUS⁶, R. BERRY³, M. JAFFEE⁴, E. A. CHRISTOU¹, D. E. VAILLANCOURT¹;

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Abstract: Rapid Eye Movement (REM) Behavior Disorder (RBD) is a strong prodromal marker of Parkinson's disease (PD) and Parkinsonism, with up to 50-60% of RBD patients converting to PD in 8-12 years. Here, we investigated the early-stage functional changes associated with RBD, using a novel form of functional brain connectivity to examine the dynamic patterns of connectivity during a robust visually guided grip force task. The rationale to use task-based connectivity is that deficits associated with RBD will be most active during a sensorimotor task. Thirty-one healthy controls were -age and -sex matched to RBD (n=41) and PD (n=39). All RBD individuals had confirmed video polysomnography diagnoses. All PD individuals were diagnosed by a movement disorder specialist and negative for atypical parkinsonism using the

Automated Imaging Differentiation for Parkinsonism. We used a unimanual grip task requiring repetitive force contractions and visual feedback to drive changes in functional connectivity. Since the putamen is affected in early PD and has been implicated in RBD previously, seeds were placed in the bilateral anterior dorsal putamen, anterior ventral putamen, posterior dorsal putamen, and posterior ventral putamen to examine the functional connectivity between these seeds and cortical regions. We also explored the relation between functional connectivity and clinical measures of cognitive, depressive, and motor symptoms. With seeds located in the bilateral posterior dorsal putamen, right posterior ventral putamen, and left anterior dorsal putamen, the PD group had reduced functional connectivity to visual and parietal regions than controls. No significant clinical measures correlated with these findings. For the RBD group, we observed far more significant seeds located in the bilateral anterior/posterior and dorsal/ventral putamen that had reduced functional connectivity to visual and parietal regions than controls. The RBD group also had a significant negative relation between functional connectivity and UPDRS-3 ($r = -0.392$) and significant positive relation between functional connectivity and Purdue Pegboard Test ($r = 0.381$). Our findings show reduced functional connectivity between the putamen and visual parietal regions in both RBD and PD patients. The RBD group had greater deficiencies relating to the anterior and posterior dorsal putamen seeds than the PD group, indicating early-stage deficits in visuospatial processing and integration during the sensorimotor task. RBD also showed unique relations with these cortico-striatal networks and clinical visuomotor tests, which may be specific to RBD and later change with Parkinsonism onset.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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VCU Parkinson's and Movement Disorders Pilot Grant.

Title: Spectral Characterization of Local Field Potentials in the Globus Pallidus Internus for Parkinson's Disease during Standing and Walking

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Abstract: Freezing of gait (FoG) is a debilitating and poorly understood symptom of Parkinson's disease (PD), often resistant to conventional therapies and difficult to quantify objectively. While the subthalamic nucleus (STN) has been extensively studied in this context, the spectral dynamics of the globus pallidus internus (GPi), another key target for deep brain stimulation (DBS), remain relatively unexplored during naturalistic motor behaviors. This study aims to characterize GPi local field potential (LFP) activity during standing and walking in individuals with PD to identify neurophysiological biomarkers relevant to gait disturbances to inform future adaptive DBS (aDBS) strategies. LFPs were recorded from four PD patients implanted with Percept PC neurostimulators targeting the bilateral GPi. Participants completed five validated obstacle courses designed to elicit FoG while wearing inertial measurement units (IMUs); sessions also included relaxed standing. Power spectral densities (PSDs) of the LFPs were estimated and decomposed into periodic and aperiodic components using the fitting oscillations & one over f (FOOOF) algorithm. Canonical frequency band powers were extracted from the FOOOF-adjusted spectra, and spectral exponent and offset parameters of the aperiodic fit were quantified. Results revealed consistent differences in spectral parameters between standing and walking, with aperiodic features showing greater reliability and robustness across participants and hemispheres. The elevated exponent during walking suggests a shift in excitation-inhibition balance, potentially reflecting suppression of GPi output and disinhibition of thalamocortical pathways essential for locomotion. Increased offset values may indicate enhanced synaptic drive and greater sensorimotor engagement during walking. In contrast, narrowband oscillatory activity varied idiosyncratically and lacked consistent state-dependent patterns. These findings support the potential of aperiodic GPi LFP features as stable, generalizable biomarkers of motor state. This work highlights the viability of leveraging aperiodic dynamics for future aDBS systems aimed at alleviating gait disturbances, including FoG, in Parkinson's disease.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Topic: C.03. Parkinson's Disease

Title: Assessing structural brain changes due to chronic L-DOPA administration in the unilateral 6-OHDA rat model

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WI; ⁷Psychology, Binghamton Univ., Binghamton, NY; ⁸Bioengineering and Neurolog. Surgery, Univ. of Wisconsin Madison, Madison, WI

Abstract: Dopaminergic replacement therapy via the dopaminergic (DA) precursor L-DOPA is the major medical therapy for Parkinson's disease (PD) resulting significant symptom control. However, long-term L-DOPA administration is known to cause uncontrolled dyskinetic movements (LID). It, however, remains unclear if the maladaptive changes in brain structure/function that give rise to LID are localized within the nigrostriatal DA system or impact various regions throughout the brain. In this study, we employed diffusion magnetic resonance imaging (dMRI) to investigate structural brain networks associated with chronic L-DOPA treatment and development of LID in a unilateral 6-OHDA rat model. Fifty four (54) male, Long Evans rats were used in this study. All procedures were approved by the Institutional Animal Care and Use Committee at UW-Madison. On day 0, while under isoflurane anesthesia, rats received an injection of 6-OHDA (n=48; 2µg/µl) or saline (n=6) into the left median forebrain bundle (MFB). Fourteen (14) days following surgery, limb use asymmetry and apomorphine induced rotation tests were performed to verify the PD phenotype. Lesioned rats (PD cohort, n=22) that exhibited more than 7 rotations per minute during the apomorphine test were included in the treatment phase. On day 16, PD rats were randomly assigned into two groups: the first group (n=15) received 1x daily L-Dopa treatment (6 mg/kg) and the second (n=7) received saline as a treatment. On day 30, the severity of LID was measured using the Abnormal Involuntary Movements (AIMs) scale at 20-minute intervals post-injection for 180 minutes. L-DOPA treated animals were assigned into two groups based on the severity of LID, measured as the area under the curve (AUC) of the AIMs scores over the 180 minute observation period. On day 31, rats were transcardially perfused with 4% paraformaldehyde and prepared for dMRI. Examination of dMRI using correlational tractography identified broad, bilateral brain statistical maps both positively and negatively associated with disease state and AIMs (FDR < 0.05). Interestingly, fibers within the MFB were found to be negatively associated with disease state consistent with degeneration of DA neurons caused by the 6-OHDA lesion. Furthermore, tracks connecting the sensorimotor cortex to the striatum and the striatum to the substantia nigra showed strong positive and negative association with AIMs, respectively. MFB fibers associated with disease state and AIMs were distinct. These results suggest that tractography based approaches are a valuable tool for investigating the neural mechanisms of PD and for evaluating the efficacy of therapeutic interventions.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Topic: C.03. Parkinson's Disease

Support: MN partnership grant for biotechnology and 353 medical genomics (MNP2142)

Title: Spike Shape-Based Classification of Electrophysiological Borders in GPi Deep Brain Stimulation Surgery

Authors: *M. R. BAKER¹, B. KLASSEN², K. J. MILLER³;
²Neurol., ³Neurosurg., ¹Mayo Clin., Rochester, MN

Abstract: Accurate identification of anatomical and functional borders within the internal globus pallidus (GPi) is critical for optimizing outcomes in deep brain stimulation (DBS) surgery for movement disorders. While microelectrode recordings (MER) are widely used to assess neural activity intraoperatively, interpretation of raw electrophysiological signals remains subjective and highly dependent on surgical expertise. In this study, we apply a novel spike shape parameterization and clustering framework to analyze intraoperative single-unit activity across patients undergoing GPi DBS. Using a canonical response parameterization approach, we quantify the similarity of action potential morphologies and classify spike waveforms using unsupervised hierarchical clustering. By comparing these clusters to clinically determined anatomical segmentations within the GPi, we evaluate whether distinct electrophysiological patterns correspond to borders between sensorimotor, associative, and limbic subregions. Our findings demonstrate that spike shape-based classification may be useful to distinguish functionally and anatomically relevant borders, offering a potential objective tool for intraoperative functional mapping and refinement of DBS targeting.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Program #/Poster #: PSTR268.18/Web Only

Topic: C.03. Parkinson's Disease

Title: Association between white matter lesions and parkinson's disease in the mexican population

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Abstract: Parkinson's disease (PD) is one of the most common neurodegenerative disorders worldwide, with an estimated prevalence of 6.1 million individuals in 2016. In recent years, a significant increase in the number of cases has been observed, particularly in countries such as China and in high-income nations in Europe. PD is characterized by the progressive loss of dopaminergic neurons in the substantia nigra, a process associated with abnormal accumulation

of α -synuclein, neuroinflammation, mitochondrial dysfunction, and lysosomal alterations. This neurodegeneration leads to the manifestation of motor symptoms (resting tremor, rigidity, bradykinesia, speech disturbances, hypersalivation) and non-motor symptoms (depression, anxiety, cognitive impairment, hyposmia, sleep, urinary, and gastrointestinal disorders). Although PD has traditionally been considered a condition primarily involving gray matter alterations, recent studies suggest a potential association between white matter lesions and the presence or severity of both motor and non-motor symptoms. Given the limited data on this phenomenon in the Mexican population, the aim of this article is to analyze the relationship between white matter lesions and the clinical symptoms of Parkinson's disease in this population group.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Topic: C.03. Parkinson's Disease

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Edward and Barbara Bell Endowed Chair.

Title: Cortico-subthalamic dynamics during freezing of gait in parkinson's disease

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Abstract: Freezing of gait (FOG) in Parkinson's disease (PD) is a complex motor symptom hypothesized to result from disrupted communication between cortical and subcortical networks. To investigate its neural basis, we simultaneously recorded subthalamic nucleus (STN) local field potentials (LFPs) and 64 channels electroencephalography (EEG) during three behaviorally relevant modules—Physical, Cognitive, and Anxiety—in 8 PD participants. Data were preprocessed using artifact subspace reconstruction, independent component analysis, and current source density transformation. Power spectral density (1-30 Hz) was estimated using the multitaper method, and cortico-subthalamic functional connectivity was assessed using the weighted phase-lag index (WPLI). Cluster-based permutation testing was used for statistical evaluation. Compared to walking, FOG episodes exhibited increased theta and alpha power (3.4-

12.4 Hz) over right parietal and occipital regions across all modules. Module-specific patterns revealed: (1) Anxiety - elevated delta-theta power (1.4-6.4 Hz) and increased STN-cortical connectivity in right parieto-occipital areas; (2) Cognitive - increased theta power (4.9-8.4 Hz) over midline and posterior electrodes, with a trend toward enhanced STN-cortical coupling in prefrontal and sensorimotor regions; (3) Physical - increased delta-theta power (1.4-8.4 Hz), reduced beta activity (22.9-29.9 Hz), and significantly elevated STN-cortical connectivity in prefrontal and sensorimotor areas. These results indicate that FOG, compared to walking, is associated with consistent low-frequency power increases and module-specific connectivity changes, implicating dysfunctional frontoparietal and motor circuits. This work provides insight into the pathophysiology of FOG and informs neuromodulation-based therapeutic strategies. Funding: MJFF-020020 and Edward and Barbara Bell Endowed Chair.

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Poster

PSTR268: Parkinson's Disease: Circuit Mechanisms and Movement Disruptions

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Program #/Poster #: PSTR268.20/E14

Topic: C.03. Parkinson's Disease

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Title: Spatial Clustering of Cortical Glutamatergic Inputs in the Zona Incerta: A Comparison Between Healthy and Parkinsonian Monkeys

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Abstract: The zona incerta (ZI), a small structure underneath the thalamus, receives topographically organized inputs from nearly the entire cortex and functions as an integrative hub mediating diverse behaviors including escape, feeding, and social interactions through its widespread projections. Despite evidence for topographic spatial organization of cortical afferents, it remains unexplored whether cortical inputs converge into clustered functional domains within the ZI. This exploratory study examines VGLUT1 immunolabeling, a highly specific marker for cortical glutamatergic terminals, with the goal of mapping the spatial distribution of all cortical inputs in the ZI. We observed non-uniform VGLUT1 distribution, raising the possibility that cortical inputs establish ZI functional territories and heterogeneously

shape activity across the ZI. This is particularly relevant in Parkinson's disease, where changes in cortical inputs could potentially contribute to motor and non-motor symptoms. Using immunoperoxidase and light microscopy techniques, we systematically characterized VGLUT1 clustering patterns in coronal ZI sections from healthy (n=4) and MPTP-treated parkinsonian (n=4) rhesus monkeys. Using ImageJ and MATLAB algorithms, we first isolated VGLUT1-labeled particles from digital images and then calculated Voronoi tessellations to conduct a quantitative clustering analysis. This analysis creates geometric density maps for each ZI section, where each detected VGLUT1 centroid point has its own surrounding polygonal territory that represents its spatial domain of influence. VGLUT1 clusters at different ZI levels showed varying densities, with small polygon areas indicating dense regions and large areas indicating sparse regions. Initial global analysis combining all rostral to caudal sections of the ZI revealed no significant difference in median Voronoi areas between groups ($337.82 \pm 317.95 \mu\text{m}^2$ (mean $\pm$ SD) and $283.39 \pm 315.1 \mu\text{m}^2$ in healthy and parkinsonian monkeys respectively; $p=0.2214$ Mann-Whitney U test). Both groups exhibited considerable variability, likely reflecting natural VGLUT1 density variation across coronal planes. Ongoing analysis will determine whether observed clusters differ significantly from spatial randomness using Monte Carlo simulations and investigate region-specific changes within functional domains that global analysis may mask. This work establishes the first quantitative map of cortical input clusters within the ZI and encourages robust spatial analysis beyond traditional densitometry for understanding topographical heterogeneity in immunostained tissue.

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Poster

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Title: Progressive Changes in Cortical Noradrenergic Innervation in the MPTP-treated non-human primate model of Parkinson's Disease

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Abstract: Parkinson's Disease (PD) is a common movement disorder characterized by motor symptoms such as tremor, rigidity, impaired balance, and slowness of movements induced by degeneration of the nigrostriatal dopamine system. However, early non-motor symptoms including executive dysfunctions in attention, cognitive flexibility and working memory often precede motor deficits and significantly impact quality of life. The chronic low-dose 1-methyl-4-phenyl-1, 2, 3, 6-tetrahydropyridine (MPTP) model of PD in non-human primates presents with early cognitive deficits in executive functions prior to motor symptoms. In humans, some of these early cognitive impairments may be due to dysfunction and/or loss of brainstem noradrenergic neurons in the locus coeruleus (LC) and their projections to frontal cortices. Given evidence that LC neurons are key regulators of arousal, attention, mood and autonomic functions, early dysregulation of these cells could elicit a broad range of non-motor features commonly seen in early PD. To further validate the use of chronic MPTP intoxication as a tool to induce progressive loss of LC noradrenergic projections in monkeys, this study aims at assessing changes in noradrenergic innervation of various cognitive and sensorimotor cortical regions in MPTP-treated monkeys with partial (~30-50%) or severe (>70%) striatal dopamine denervation. To date, the brain tissue from 2 control and 3 MPTP-treated parkinsonian monkeys has been collected and immunostained for norepinephrine transporter (NET). Series of digital images of brain sections were analyzed to compare the intensity of NET immunostaining in different layers of sensorimotor and cognitive frontal cortices (Brodmann's area 4, 6, 9, 46, 24, and 25) between control and MPTP-treated monkeys. Initial analyses revealed a 60-80% decrease in NET labeling intensity measurements from all cortical areas examined in MPTP-treated parkinsonian monkeys compared to controls. The extent of LC neuronal loss is being assessed and cortical tissue from motor asymptomatic MPTP-treated monkeys with partial striatal denervation is being analyzed. We hypothesize that motor asymptomatic MPTP-treated monkeys with partial striatal dopamine denervation will display a significant loss of cortical NET immunostaining and mild degeneration of LC neurons suggesting retrograde degeneration of cortical NE innervation as proposed in PD patients. The results of this study will provide greater insights into the possible use of the MPTP-treated monkey model to understand the pathophysiology and develop therapeutics for early non-motor PD symptoms.

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Poster

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France Parkinson (SUMMA GAO 2019)
Agence Nationale de Recherche (ANR-19-CE17-0031 - SUMMA)

Title: Strain specific α Synuclein effects on brain function and pathology in non human primates

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Abstract: Synucleinopathies are characterized by the pathological aggregation of α -synuclein (α -syn), a normally monomeric presynaptic protein. In Parkinson's disease (PD), these aggregates form Lewy bodies and neurites, contributing to neurodegeneration. Compelling evidence shows that α -syn aggregates propagate in a prion-like manner, recruiting monomers and spreading between cells. This was first observed in fetal neuronal grafts implanted in PD brains, where host-derived α -syn inclusions appeared. Experimental models using viral vectors to overexpress wild-type or mutant α -syn in rodents and primates, as well as inoculation of brain homogenates from transgenic mice or synucleinopathy patients, or synthetic α -syn preformed fibrils (PFF), have demonstrated α -syn propagation and dopaminergic degeneration. Importantly, α -syn can adopt structurally distinct fibrillar forms or "strains" that differ in their biochemical and pathological profiles. In vivo studies show that these strains induce distinct patterns of α -syn pathology and neurotoxicity, supporting the hypothesis that strain-specific properties contribute to clinical heterogeneity in synucleinopathies. Here we performed bilateral inoculation of 18 primates both in the substantia nigra (AAV-CBA- α SynA53T or PBS) and putamen (PFF, patient-derived α -syn or PBS). All NHPs underwent motor and cognitive assessment longitudinally as well as PET imaging at baseline, 6, and 12 months post-injection (moPI). Post-mortem analysis included tyrosine hydroxylase immunocytochemistry to detect α -syn-mediated nigrostriatal alterations. We assessed the seeding propensity of the various α -syn inocula by quantifying phosphorylated and aggregated α -syn. Preliminary results show motor and cognitive deficits of different severity according to the inocula, and suggest that the combination of AAV injection in the SN and PFFs or patient α -syn in the putamen significantly exacerbates the phenotype and accelerates the appearance of disease symptoms. This was further confirmed by PET imaging, looking at the activity of the dopamine transporter (18F-FE-PE2I) and the D2 receptor 18F-Fallypride, as well as a radioligand binding α -syn. Post-mortem and biomarker studies are currently ongoing. Our findings support the relevance of α -syn strain diversity and dual-hit models in reproducing the complexity of synucleinopathies and, to the best of our knowledge, offer a robust and valuable platform for therapeutic and biomarker testing.

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Poster

PSTR269: Huntington's Disease: Preclinical Studies

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Program #/Poster #: PSTR269.01/E17

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NSF Grant 82394422
NSF Grant 82371874

Title: Gene therapy for huntington's disease using large animal models

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Abstract: Huntington's disease (HD) is caused by the expansion of CAG repeat sequences in the first exon of the huntingtin (HTT) gene. Mutant huntingtin (mHTT) with expanded PolyQ preferentially causes striatal neurodegeneration. Currently, there is no effective cure for HD. The HD knock-in pig model, which mimics striatal neurodegeneration seen in HD patients, provides a valuable platform for studying HD pathogenesis and developing therapeutic strategies. Our research revealed that aberrant spliced HTT exon1 is also present in the brains of HD pigs, but it is expressed at a much lower level than the normally spliced HTT exon products, indicating that mutant HTT exon 1 is a major toxic product in HD. Consistently, Disrupting or repairing mutant HTT exon 1 in HD knock-in pigs using CRISPR/Cas9 could alleviate neuropathology and movement abnormalities. However, non-allele-specific targeting with CRISPR/Cas9 may inadvertently affect the wild-type HTT, potentially impacting its normal function. Considering that HTT's function is age-dependent, we investigated whether CRISPR/Cas9-mediated HTT depletion in the adult monkey brains posed harm to neuronal cells. Our findings demonstrated no significant effects of HTT deletion on neuronal, glial cells in the brain striatum of adult cynomolgus monkeys for up to one months. Further long-term studies are ongoing to explore whether simple disrupting exon 1 of HTT in the striatum of adult primates could serve as a viable intervention strategy for HD.

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Poster

PSTR269: Huntington's Disease: Preclinical Studies

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Program #/Poster #: PSTR269.02/E18

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NIH Grant R01NS123557

Title: Focused Ultrasound-Mediated Delivery of Adeno-Associated Virus Vectors Expressing Short Hairpin RNAs for the Silencing of Msh3 in a Mouse Model of Huntington's Disease

Authors: *B. S. OWUSU-YAW¹, Y. ZHANG¹, R. MOURO PINTO², N. TODD¹;
¹Radiology, Brigham and Women's Hosp., Boston, MA; ²Ctr. for Genomic Med., Massachusetts Gen. Hosp., Boston, MA

Abstract: Huntington's disease (HD) is a fatal, autosomal dominant, neurodegenerative disorder caused by a cytosine-adenine-guanine (CAG) trinucleotide repeat expansion in exon 1 of the *HTT* gene that encodes the huntingtin (HTT) protein. There are currently no cures for HD and for several years research has focused on HTT lowering strategies, involving the use of RNA interference (RNAi) and antisense oligonucleotides. But emerging scientific research suggests that targeting somatic repeat expansion instability could delay the onset of disease and slow down disease progression in HD. Studies in HD mouse models have demonstrated that knocking down MSH3 using RNA interference can block somatic repeat expansion. Adeno-associated virus (AAV) vectors can be used to deliver RNAi constructs, but effective delivery is hindered by the blood-brain barrier (BBB). Direct methods of injection are invasive, require skilled neurosurgeons and carry the risk of infection. Whereas intravenous administration requires higher doses to reach therapeutic concentrations in the brain and higher doses are associated with systemic toxicity. Focused ultrasound (FUS) with microbubbles is an alternative approach that can be used to temporarily disrupt the BBB to deliver AAV vectors and other therapeutics to specific brain regions in a non-invasive manner. Here, we tested the delivery of an scAAV9 containing a short hairpin RNA (shRNA) targeting mouse *Msh3* using focused ultrasound combined with microbubbles to the right striatum of 12-week-old Hdh^{Q111} mice. T1-weighted contrast enhanced MRI images were acquired before and after gadolinium injections to confirm and visualize the location and extent of BBB opening. Our results show successful BBB opening across treatment groups demonstrated by a significant increase in BBB permeability in the sonicated striatum compared to the untreated hemisphere in Hdh^{Q111} mice following FUS-mediated BBB opening. Mice will be sacrificed at 30 weeks post treatment to assess CAG repeat length, DNA vector copy number and MSH3 protein expression.

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Poster

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Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NIH T32 AG052374

Title: Zip delivery: a novel peptide delivery system for the brain

Authors: *B. ROSSMILLER¹, L. M. ELLERBY²;

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Abstract: One of the major challenges in developing therapeutics for neurological disorders remains widespread delivery of therapeutics to the brain. We addressed this challenge by developing a novel cell penetrating peptide based on the Zika virus, called “ZIP” we have tested this technology in neuronal stem cells, striatal organoids and murine models. We have fused ZIP to both CRISPR and to EGFP to evaluate its ability to transport cargo both into neuronal cells and past the blood brain barrier. Our initial experiments focused on using Huntington’s disease models to explore a potentially therapeutic mechanism for decreasing the number of CAG repeats in the Exon 1 CAG expansion. Huntington’s disease is a rare, devastating, neurodegenerative disorder with no cure and minimal treatments. It has been previously shown that a reduction in the expression levels of the expanded CAG repeat in Exon 1 would lead to an amelioration of symptoms. A major challenge for HD treatments in decreasing the CAG expansion has been the delivery of relevant therapeutics. This challenge was why we specifically on various HD models for our studies. We were able to show a contraction of the Exon 1 expansion in both neuronal stem cells and striatal organoids. Impressively, we have been able to show that the “ZIP” has been able to transport Cas9 past the adult murine BBB and into neurons and astrocytes with extremely high efficiency, including into the hippocampal and striatal regions, from a single intravenous injection. We have further tested the ability of ZIP to transport different cargos by adding a EGFP instead of a Cas9, again showing transport into human striatal organoids and past the adult murine brain. In an effort to move this technology toward potential human use we have explored the ability to have the ZIP-Cas9 commercially synthesized and we are testing this new construct to evaluate recombination efficiency at therapeutically relevant levels. Our presentation will highlight the advances ZIP technology continues to make as a potential therapeutic delivery platform for neurological diseases and disorders

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Poster

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Program #/Poster #: PSTR269.04/E20

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Title: Context-dependent translation inhibition as a novel therapeutic mechanism to address CAG repeat (poly-Q) expansion disorders including Huntington’s Disease

Authors: *A. P. SCHULLER, M. HOLM, P. V. SAUER, H. K. ADAMS, G. W. WIENKER, T. Z. LEHMBERG, L. FERGUSON, P. D. DIAMOND, M. MEYERS, Z. KONST, S. BAHMANYAR, L. G. HAMANN;
Interdict Bio, South San Francisco, CA

Abstract: Huntington's Disease (HD) is a neurodegenerative disorder with >1,000 new diagnoses annually in the US caused by an expanded CAG trinucleotide repeat in *HTT* gene that encodes the protein huntingtin. Inserted into Exon 1 of *HTT*, these repeats encode long poly-glutamine (poly-Q) stretches into the N-terminus of the huntingtin protein that lead to protein aggregation and disrupt huntingtin function (Bates, *et al.* 2015). Interdictors are sequence-selective inhibitors of translation that bind to the human ribosome and act in a sequence preferential manner to inhibit translation elongation (Diamond, *et al.* 2025). As heterobifunctional small molecules, interdictors bind to the ribosome at the peptidyl-transferase center where the growing polypeptide chain adopts a uniformly linear conformation and make selective interactions with the nascent polypeptide chain as it is being synthesized to create friction that ultimately slows the rate of protein synthesis. Using structure-guided drug design, we designed and synthesized interdictors that selectively inhibit the translation of long poly-Q peptides, such as those at the N-terminus of mutant huntingtin, as a novel approach to reduce the cellular burden of toxic huntingtin protein caused by CAG repeat expansion. Using an *in vitro* assay to measure translation kinetics, we observe that designed interdictors are more than 10-fold selective for inhibiting the translation of a long poly-Q peptide (Q₁₄₄) representative of the pathological form of mutant huntingtin, compared to short poly-Q (Q₂₄). These data suggest a synergistic effect of inhibiting successive poly-Q incorporation by the ribosome with our interdictor, possibly through induction of ribosome quality control mechanisms. Furthermore, we used cryo-electron microscopy (cryo-EM) to determine the structure of an interdictor-bound, poly-Q ribosome nascent chain complex to 2.2 Å resolution where we observe the network of hydrogen bonds that drive selective interactions between the interdictor and the poly-Q peptide being synthesized. Together, our data establish context-dependent translation inhibition as a novel approach to inhibit the expression of poly-Q expansion containing proteins, including mutant huntingtin, and reverse the progression of HD.

Disclosures: **A.P. Schuller:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **M. Holm:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **P.V. Sauer:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **H.K. Adams:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **G.W. Wienker:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **T.Z. Lehmberg:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **L. Ferguson:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **P.D. Diamond:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Interdict Bio, Inc. **M. Meyers:** A. Employment/Salary (full or part-time): Interdict Bio, Inc.. E. Ownership

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Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR269.05/E21

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NIH Grant R01NS040068
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CHDI Foundation Grant A3917

Title: Cross-biofluid miRNA biomarker validation in Huntington's disease

Authors: J. D. CLEMSEN¹, H. J. BOCKHOLT¹, C. SCHERZER², *J. S. PAULSEN³;
¹Georgia State Univ., Atlanta, GA; ²Neurol., Yale Sch. of Med., New Haven, CT; ³Univ. of Wisconsin, Madison, Madison, WI

Abstract: MicroRNAs (miRNAs) are promising biomarkers for neurodegenerative diseases due to their stability and accessibility in biofluids. In Huntington's disease (HD), miRNA signatures have been profiled in cerebrospinal fluid (CSF) and plasma, but cross-biofluid validation remains unexplored. We aimed to identify and evaluate overlapping miRNA species detectable in both CSF and plasma from gene-positive HD participants, supporting robust, non-invasive biomarker development. We reanalyzed CSF miRNA data from PREDICT-HD (n=55) using the NanoString nCounter® Human v3 platform [1], profiling >800 miRNAs via direct hybridization and digital barcoding. RNA was extracted from 200 µL CSF using phenol-chloroform and assessed by NanoDrop and Bioanalyzer. We compared these findings to plasma miRNA data (n=99) from the Scherzer Lab [2], where RNA from EDTA plasma was isolated using the Qiagen miRNeasy Kit and profiled via Illumina NGS. Differential expression was analyzed with DESeq2 and validated in two independent cohorts. *miR-30d-5p* and *miR-323b-3p* were unique to plasma [2] while *miR-132-3p*, *miR-106a-5p*, and *miR-135b-3p* were unique to CSF [1]. These may reflect biofluid-specific regulatory roles. *miR-10b-5p* emerged as a strong candidate for cross-biofluid validation, appearing in both datasets. It is implicated in neuronal plasticity and

immune signaling, and co-expressed in CSF with *miR-132-3p* and *miR-135b-3p*, converging on inflammatory pathways [5]. Cross-biofluid analysis highlights *miR-10b-5p* as a candidate HD biomarker. Integrating CSF and plasma miRNA profiles may improve robustness and reduce invasiveness. These findings support longitudinal validation of peripheral miRNAs as proxies for central inflammation in HD. References: [1] PMID: PMC5778676; [2] PMID: PMC8682947; [3] PMID: PMC5513519; [4] PMID: PMC3380106; [5] PMID: PMC9993119

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Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

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Program #/Poster #: PSTR269.06/E22

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Title: Immune checkpoint blockade as a therapeutic approach in Huntington's disease

Authors: *A. KERTSER¹, S. BRAIMAN¹, R. LAVENDER¹, M. SCHWARTZ², K. BARUCH¹;

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Abstract: Huntington's disease (HD) is an inherited neurodegenerative disorder in which mutant huntingtin (mHTT) aggregates drive progressive motor, cognitive, and psychiatric decline. HD involves chronic inflammation within the central nervous system (CNS), driven in part by resident glial cells that adopt a persistent activated state, perpetuating rather than resolving tissue damage. In multiple models of dementia, mobilizing the peripheral immune system via transient PD-1/PD-L1 immune checkpoint blockade has been shown to reduce neuroinflammation and mitigate disease progression. Here, we tested whether this immune-based approach could benefit HD. Female R6/2 mice received a single intraperitoneal injection of anti-PD-L1 antibody at the stage of early symptom onset (7 weeks of age), resulting in transient elevation of circulating PD-1⁺CD44⁺ memory CD4 T cells. Motor assessments showed that the treated mice had fewer clasping events, longer wire-hang endurance at 10 weeks, and fewer slips on a beam-walk test at 11 weeks of age. Brain tissue analyses four weeks post-injection revealed reduced astrogliosis, increased striatal neuronal density, and a shift of mHTT from aggregated to soluble forms. These findings support the notion that intermittent blockade of the inhibitory PD-L1 checkpoint activates a common immune-based mechanism driving a therapeutic effect in neurodegenerative diseases, independent of disease primary etiology.

Disclosures: A. Kertser: A. Employment/Salary (full or part-time): ImmunoBrain. S. Braiman: A. Employment/Salary (full or part-time): ImmunoBrain. R. Lavender: A. Employment/Salary (full or part-time): ImmunoBrain, Weizmann Institute of Science. M.

Schwartz: A. Employment/Salary (full or part-time): ImmunoBrain, Weizmann Institute of Science. **K. Baruch:** A. Employment/Salary (full or part-time): ImmunoBrain.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

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Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: OHSU PCO Pilot Grant

Title: Time-restricted eating improves mitochondrial bioenergetics in early-stage Huntington's disease: Mechanistic insights from a 12-week interventional trial

Authors: ***R. WELLS**¹, L. NEILSON², A. MCHILL³, A. HILLER²;

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Abstract: Huntington's disease (HD) is a progressive neurodegenerative disorder marked by abnormal motor control, cognitive impairment, and psychiatric disturbances. Despite intensive efforts, no disease-modifying therapies exist, underscoring the need for novel strategies. Time-restricted eating (TRE), a form of intermittent fasting, has emerged as a potential therapeutic candidate in HD through its activation of adaptive neuroprotective stress response mechanisms. Although TRE has shown potential in other neurodegenerative disease contexts and in HD animal models, it has yet to be analyzed in the clinical setting. To address this gap, we conducted an open-label, single-arm, interventional trial which evaluated the effects of an 8-hour daily TRE regimen for 12 weeks in 20 people with early-stage HD with no prior TRE experience. We previously reported that TRE was well-tolerated, feasible, and appeared safe in this population. Participants exhibited significant improvements in motor symptoms, cognitive performance, and the composite Unified Huntington's Disease Rating Scale (cUHDRS) score. Specifically, Total Motor Score, Verbal Fluency Test, and Symbol Digit Modalities Test scores improved by 12% (p=0.03), 17% (p=0.0004), and 7% (p=0.02) from baseline to follow-up, respectively. The cUHDRS improved by 0.5 ± 0.7 points compared to pre-treatment (p=0.005). Notably, this score typically declines by about 1 point each year in early-stage HD. Here, we sought to explore potential underlying mechanisms by analyzing mitochondrial function in peripheral blood mononuclear cells using Seahorse XF Extracellular Flux Analyzer - a plate-based real-time approach. We found significant enhancements in bioenergetic profiles compared to pre-intervention in basal respiration (29%; p=0.01), maximal respiration (24%; p=0.01), ATP-linked respiration (20%; p=0.03), and a trending improvement in spare capacity (18%; p=0.19). Considering the evidence implicating mitochondrial dysfunction and bioenergetic inefficiencies in HD, our findings suggest TRE may rescue metabolic disturbances. Next, we plan to analyze peripheral biomarkers of neurodegeneration and conduct targeted neuropathology gene expression analysis to further elucidate the cellular and physiologic effects of TRE in this population. Despite limitations of this unblinded study, we establish a mechanistic and clinical

foundation for further investigation of TRE as a neurorestorative intervention in HD and lay the foundation for exploring this in related neurodegenerative diseases.

Disclosures: R. Wells: None. L. Neilson: None. A. McHill: None. A. Hiller: None.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR269.08/E24

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NIH CHDI

Title: Augmenting Nrf2 Signaling is neuroprotective in Huntington's Disease

Authors: *D. N. HOOPES¹, B. D. PAUL²;

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Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disease which has been found to result in heightened oxidative stress and neuronal cell death. The oxidative stress response in the cell is mediated by nuclear factor erythroid 2-related factor 2 (Nrf2), and H₂S signaling which is blunted in HD. H₂S is known to regulate Nrf2 signaling in neurodegenerative states. The Nrf2/H₂S axis is known to mitigate neuroinflammation, oxidative stress, and neurodegeneration in HD, AD, and PD. Here we show that augmenting H₂S signaling in both cell culture (Q7 and Q111) and mouse models (R6/2 and zQ175) of HD confers therapeutic benefits.

Disclosures: D.N. Hoopes: None. B.D. Paul: None.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR269.09/E25

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Title: Anle138b ameliorates pathological phenotypes in mouse and cellular models of Huntington's disease

Authors: *M. PADILHA;

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Abstract: Huntington's disease (HD) is a devastating neurodegenerative disorder characterized by the progressive loss of motor function and by cognitive decline. HD is caused by the expansion of the CAG tract in the Huntingtin gene (HTT) above a threshold of 35 repeats. Currently there are no disease-modifying therapies available for HD, making the search for effective treatments a critical enterprise. Here we show that administration of the small oligomer modifying molecule anle138b, ameliorates disease phenotypes in cellular and mouse models of HD. Anle138b reduced mutant HTT aggregate formation and ameliorated toxicity in mouse primary neurons and decreased aggregate load in human neuronal progenitor cells differentiated from HD patient-derived induced pluripotent stem cells (iPSCs). Moreover, R6/2 HD model mice treated with anle138b performed significantly better in a battery of motor assays (rotarod, grip strength, open field and hindlimb clasping test) when compared to littermate R6/2 mice treated with placebo. In addition, anle138b-treated R6/2 mice had less pronounced brain atrophy and increased lifespan. No adverse effects of anle138b administration were observed in wildtype littermates. Further investigation into the possible mechanisms underlying these improvements revealed that anle138b decreased intranuclear mutant HTT aggregate load, rescued neurochemical imbalances, mitigated dendritic spine loss in striatal spiny projection neurons and reduced neuroinflammation. Administration of anle138b in a second HD mouse model, the knock-in zQ175DN model, confirmed the small molecule's ability to decrease mutant HTT aggregate load and rescue neurochemical changes. Altogether these results suggest that anle138b is an effective approach in the treatment of HD.

Disclosures: M. Padilha: None.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR269.10/E26

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NIH R21NS132089
NIH R01NS120832
NSF NeuroNex-1707352

Title: Pre-symptomatic corrective circuit manipulation through optical synapse targeting in Huntington's disease mice

Authors: *C. BERMUDEZ¹, M. TREE², G. L. DUNBAR, Sr.⁵, J. ROSSIGNOL³, U. HOCHGESCHWENDER⁴;

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Univ., MT Pleasant, MI; ⁵Psychology/Program in Neurosci., CENTRAL MICHIGAN UNIVERSITY, MOUNT PLEASANT, MI

Abstract: Huntington's disease (HD) is characterized by a cytosine-adenine-guanine (CAG) triplet expansion in exon 1 of the huntingtin gene, resulting in the late-stage symptoms of motoric, psychiatric, and cognitive deficits. In presymptomatic HD patients and mouse models, excitatory overdrive of cortical output onto striatal neurons has been observed. Our central hypothesis is that manipulating the firing activity within selected microcircuits before the onset of symptoms will slow HD disease progression. We previously utilized bioluminescent optogenetics (BL-OG) to counteract the increased activity of cortical pyramidal neurons (CPNs) and the decreased activity of parvalbumin interneurons (PNs) in R6/2 HD mice. Luminopsins (LMOs), fusion proteins of a light-emitting luciferase and a light-sensing opsin, were expressed in CPNs (inhibitory LMOs) or in PNs (excitatory LMOs). Daily luciferin application during weeks 5 - 8 in R6/2 HD mice dampened the excitatory cortical drive and significantly slowed progression of motor deficits as seen in improved motor coordination and gait parameters. We now want to explore the possibility of self-regulating pathological corticostriatal communication by applying an "Interluminescence" strategy to convert the excitatory overdrive of CPNs into dampening the activity of striatal target neurons. This is achieved through the "optical synapse" where a vesicle-targeted luciferase is released into the synaptic cleft when the presynaptic CPN neuron fires; in the presence of luciferin the bioluminescence emitted will activate an inhibitory optogenetic channel expressed in postsynaptic striatal neurons. AAVs of a Cre-dependent luciferase (AAV2/9 Efla DIO hPOMC1-26 sbGluc dTomato) were used to transduce the M1 motor cortex of 3-week-old Emx1-Cre::R6/2 mice and AAVs of an inhibitory opsin (AAV2/9 hSyn hGtACR1 EYFP) to transduce neurons in the striatum. Intraperitoneal application of luciferin once daily during weeks 5 - 8 is expected to convert the overexcitation of CPNs into dampening of striatal neuronal activity and HD motor recovery in the HD mice. Mice are undergoing various motor-based tests such as balance beam and ladder tests, as well as rotarod and grip strength to assess recovery in motor function.

Disclosures: C. Bermudez: None. M. Tree: None. G.L. Dunbar: None. J. Rossignol: None. U. Hochgeschwender: None.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

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Program #/Poster #: PSTR269.11/E27

Topic: C.04. Movement Disorders Other Than Parkinson's Disease

Support: NSFC Grant 2021YFA0805200
NSFC Grant 2021ZT09Y007
NSFC Grant 2020B121201006

Title: Retrograde AAV-mediated huntington's disease monkey model recapitulates core neuropathology

Authors: *Y. QIN¹, S. YANG¹, X.-J. LI²;

¹Jinan Univ., Guangzhou, China; ²Jinan Univ., Guangdong, China

Abstract: Huntington's disease (HD) is a neurodegenerative disorder caused by abnormal trinucleotide repeat expansion in the HTT gene. Research on its pathogenic mechanisms and therapeutic strategies largely relies on animal models, yet existing models have significant limitations: mouse models often lack prominent neuronal loss, while transgenic monkey models face reproductive challenges. Conventional viral-mediated primate models also suffer from technical drawbacks such as uneven transfection efficiency and limited viral spread. In this study, we successfully established a novel HD monkey model using a retrograde adeno-associated virus combined with a medium spiny neuron (MSN)-specific promoter. By injecting the retrograde AAV into the substantia nigra of adult cynomolgus monkeys, we achieved widespread expression of the mutant HTT exon 1 fragment throughout the striatum. The monkey model recapitulated key neuropathological features of HD, including: (1) impaired striatal dopaminergic neuron function, (2) formation of mutant HTT nuclear inclusions, (3) marked glial activation, and (4) significant neuronal loss. These results demonstrate that this model accurately reproduces the neuropathological hallmarks of HD patients, providing a robust platform for mechanistic studies and drug development.

Disclosures: Y. Qin: None. S. Yang: None. X. Li: None.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR269.12/E28

Topic: C.05. Tauopathies, Synucleinopathies, and Other Related Diseases

Title: In vitro and in vivo comparison of three small molecule splicing modulators: Branaplam, PTC-518 and CHDI-113

Authors: *P. MITCHELL¹, M. E. HERVA MOYANO², D. MAGNANI³, D. F. FISCHER⁴;

¹Charles River Labs, Saffron Walden, United Kingdom; ²Discovery. Biol., ³Discovery UK, Charles River Labs., Saffron Walden, United Kingdom; ⁴Discovery, Charles River, Saffron Walden, United Kingdom

Abstract: Lowering the expression of pathogenic huntingtin protein (mHTT) is a leading therapeutic approach for the treatment of Huntington's Disease (HD). Two small molecule HTT splicing modulators (Branaplam and PTC-518) have been advanced to the clinic and successfully demonstrated CSF mHTT lowering in HD patients. Both molecules lower *HTT* expression by promoting the inclusion of a pseudo exon i49 containing a premature termination codon that

leads to transcript degradation and HTT lowering via nonsense mediated decay. The Branaplam VIBRANT-HD trial was terminated due to signs of neurotoxicity including peripheral neuropathy in some treated patients. However, PTC-518 was granted fast track designation after showing promising results from the 12-month interim data readout of the PIVOT-HD study and no evidence of peripheral neuropathy in patients. Here we report *in vitro* and *in vivo* profiling of Branaplam, PTC-518 and a structurally distinct HTT splicing modulator (CHDI-113). *In vitro* all three compounds had similar HTT lowering potency in HD stem cell models. We observed no toxicity with PTC518 up to 30 μ M, whereas Branaplam and CHDI-113 reduced cell viability at high concentrations. Interestingly, all three compounds induced PBMC cell cycle arrest at similar concentrations via a mechanism that is not clear. In terms of selectivity for HTT over other less desirable splicing targets CHDI-113 displayed marginally more selectivity over *MADD*, while PTC-518 shows greater selectivity over *FOXMI*. However, PTC-518's lower aqueous solubility (<5 μ M) impacts accurate selectivity and toxicity assessments *in vitro*. When orally dosed for 21 days BACHD mice, Branaplam showed the greatest peripheral tissue HTT lowering (compared to PTC-518 and CHDI-113) at doses resulting in 50% CNS mHTT lowering. The therapeutic Index (defined as the ratio of highest doses tolerated vs dose required to reach 50% mHTT lowering in CNS) was similar and very narrow (2-fold) for all three compounds

Disclosures: P. Mitchell: None. M.E. Herva Moyano: None. D. Magnani: None. D.F. Fischer: None.

Poster

PSTR269: Huntington's Disease: Preclinical Studies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR269.13/E29

Topic: C.05. Tauopathies, Synucleinopathies, and Other Related Diseases

Support: NRF Grant RS-2023-00241411

Title: Exosomes as a potential messenger unit for amelioration of Huntington's disease

Authors: *M. LEE¹, M. KIM²;

¹Seoul Natl. Univ. Hospital/ Biomed. Res., Seoul, Korea, Republic of; ²Seoul Natl. Univ. Hosp, Seoul, Korea, Republic of

Abstract: Huntington's disease (HD) is characterized by progressive neurodegeneration that begins well before clinical symptoms appear. Despite extensive research, effective therapies to halt or reverse disease progression remain elusive. Emerging evidence suggests that exosomes—nanoscale extracellular vesicles involved in intercellular communication—may play a pivotal role in modulating neurodegenerative processes. However, the identity and function of specific messenger units that transfer beneficial factors in systemic circulation remain poorly understood. Here, we demonstrate that exosomes isolated from young blood serum mediate neuroprotective effects in HD, suggesting their potential as therapeutic vectors. Using the R6/2 mouse model of

HD, we investigated the effects of exosomes derived from young versus old wild-type mice. Neural stem cells differentiated from the subventricular zone (SVZ) of the brain were employed as an in vitro cellular model of HD. Treatment with exosomes isolated from young serum significantly ameliorated mutant Huntingtin aggregation (EM48, $p < 0.05$), enhanced mitochondrial biogenesis (p-CREB/CREB, $p < 0.005$), reduced apoptotic markers including p53 ($p < 0.05$), Bax ($p < 0.005$), cleaved caspase-3 ($p < 0.05$), and increased expression of the anti-apoptotic protein Bcl-2 ($p < 0.05$). Furthermore, cell proliferation was significantly improved, as assessed by WST-1 assays ($p < 0.005$). HD mice model exposed to young blood circulation via heterochronic parabiosis exhibited improved weight maintenance and survival. Corresponding reductions in mutant Huntingtin aggregation (EM48, $p < 0.005$), mitochondrial dysfunction (PGC-1 α , $p < 0.05$; p-CREB/CREB, $p < 0.005$), apoptotic cell death (p53, Bax, cleaved caspase-3; all $p < 0.05$), and improvements in cognitive markers (DCX, $p < 0.05$) were observed. Collectively, these findings implicate exosomes as critical mediators of the rejuvenating effects observed in heterochronic parabiosis. Our results provide compelling evidence that exosomes derived from young blood carry neuroprotective factors capable of mitigating HD pathology. These findings underscore the therapeutic potential of exosome-based interventions for neurodegenerative diseases such as HD.

Disclosures: M. Lee: None. M. Kim: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.01/E30

Topic: C.06. Neuromuscular Diseases

Title: Acetylated α -Tubulin as a clinical plasma biomarker of disease severity in CMT1A

Authors: *C. J. MALTBY, J. PINTO, M. BIANCHI;
Ulysses Neurosci. Ltd, Dublin, Ireland

Abstract: Charcot-Marie-Tooth disease type 1A (CMT1A) is the most common form of inherited peripheral neuropathy, caused by a 1.5 Mb duplication on chromosome 17p11.2 encompassing the *PMP22* gene. This duplication leads to *PMP22* overexpression, progressive demyelination, and peripheral nerve dysfunction. Clinically, CMT1A presents with distal muscle weakness, atrophy, sensory deficits, foot deformities, and gait disturbances, often beginning in childhood. While several clinical and electrophysiological scales exist to monitor disease progression, they are limited by subjectivity and an insensitivity to gradual and slow disease progression. Efforts to identify circulating biomarkers have included neurofilament light chain (Nf-L), which reflects axonal injury, but has shown limited correlation with CMT1A disease severity or progression, thus constraining its use in disease progression measures and clinical trials. To address this, we evaluated acetylated α -tubulin—a microtubule-stabilising post-translational modification—as a candidate blood-based biomarker in the plasma of 46 genetically

diagnosed CMT1A patients and 31 age-matched healthy controls. Acetylation at lysine 40 (K40) of α -tubulin, catalysed by α TAT1 and removed by HDAC6, promotes microtubule stability and efficient axonal transport, processes essential for neuronal health and long-term neuronal stability. Dysregulation of α -tubulin acetylation has been implicated in neurodegeneration and axonal dysfunction, so we hypothesized that reduced tubulin acetylation may couple with impaired axonal integrity in CMT1A. In this study, we first confirmed that Nf-L is significantly increased in the plasma of CMT1A patients, but show that these increases in Nf-L do not correlate with increases in clinical outcome and severity measures for CMT1A. Furthermore, we demonstrate that plasma levels of acetylated α -tubulin are significantly decreased in individuals with CMT1A compared to healthy controls, with acetylated α -tubulin levels showing significant inverse correlation with both the Overall Neuropathy Limitations Scale (ONLS) and the Charcot-Marie-Tooth Examination Score (CMTES), suggesting a strong relationship between biomarker level and clinical impairment of the patients assessed. These findings indicate that impaired axonal integrity observed in CMT1A patients are coupled with a decrease in microtubule stability. Acetylated α -tubulin is a downstream target of HDAC6 inhibitors that are currently under investigation for the treatment of CMT, and thus serves as a novel and disease-relevant biomarker that reflects both molecular pathology and clinical severity in CMT1A.

Disclosures: **C.J. Maltby:** A. Employment/Salary (full or part-time): Ulysses Neuroscience Limited. **J. Pinto:** A. Employment/Salary (full or part-time): Ulysses Neuroscience Limited. **M. Bianchi:** A. Employment/Salary (full or part-time): Ulysses Neuroscience Limited.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.02/E31

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant K22NS125057
JHU Merkin Center Grant

Title: Pmp22 dosage disrupts myelin subdomain architecture in cmt1a and hnpp: evidence for primary myelin pathogenesis in the most prevalent inherited peripheral neuropathies

Authors: ***M. A. AROWOLO**¹, D. R. GUTIERREZ², A. HOKE², K. MOSS³;
¹NextGen Precision Hlth., Univ. of Missouri, Columbia, MO; ²Dept. of Neurol., Johns Hopkins Univ., Baltimore, MD; ³Physical Med. and Rehabil., Univ. of Missouri, Columbia, MO

Abstract: Charcot-Marie-Tooth disease type 1A (CMT1A) and Hereditary Neuropathy with Liability to Pressure Palsies (HNPP) are the most prevalent inherited peripheral neuropathies, caused by duplication and deletion of the Peripheral Myelin Protein 22 (PMP22) gene, respectively. While secondary axonal degeneration has long been considered the primary driver

of pathology in patients, our previous work revealed neuromuscular deficits in CMT1A mice without overt axon loss, suggesting a primary role for myelin dysfunction. In this study, we investigated how altered PMP22 dosage affects the molecular organization of peripheral nerve myelin. Using high-resolution confocal imaging of teased sciatic nerve fibers from CMT1A and HNPP mouse models, we observed structural abnormalities at key myelin subdomains: Schmidt-Lanterman incisures (SLIs) and nodes of Ranvier. We identified disorganization of adherens junctions, mislocalization of Connexin29, and altered distribution of nodal ion channels in CMT1A and HNPP model myelin, with several defects more pronounced in CMT1A, aligning with clinical severity. Notably, Kv1.2 and Caspr mislocalization along the internode and Nav nodal widening suggest disruption of axoglial domains essential for saltatory conduction. Together, these phenotypes support a model in which PMP22 likely regulates myelin architecture through adherens junctions, consistent with prior reports linking PMP22 to cell adhesion both structurally and functionally. Disruption of these myelin domains is predicted to impair metabolic support and maintenance of axonal ion flow required for efficient nerve conduction, thereby compromising the structural and functional integrity of the myelin sheath and contributing directly to disease pathogenesis. These findings shift the pathogenic paradigm for CMT1A and HNPP from axonal degeneration to primary myelin failure and highlight actionable molecular targets for therapeutic intervention. This study offers mechanistic insight into CMT1A and HNPP and provides a conceptual framework with broad relevance to other dysmyelinating disorders.

Disclosures: M.A. Arowolo: None. D.R. Gutierrez: None. A. Hoke: None. K. Moss: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.03/E32

Topic: C.06. Neuromuscular Diseases

Support: Charcot-Marie-Tooth Association

Title: Organelle transport deficits drive multisystem dysfunction in an MFN2 rat model of Charcot-Marie-Tooth disease

Authors: *W. MATYSIK¹, J. CICERO², C. WU², K. SUNG², E. OROZCO², K. PAK², K. HITCHCOCK², T. VELASQUEZ², U. MANOR²;

¹UCSD, San Diego, CA; ²Univ. Of California San Diego, La Jolla, CA

Abstract: Charcot-Marie-Tooth disease type 2A (CMT2A) is a dominantly inherited peripheral neuropathy caused by mutations in MFN2, a gene critical for mitochondrial fusion and organelle trafficking. Despite clear progressive motor and sensory deficits in patients, the link between subcellular organelle dysfunction and systems-level phenotypes remains poorly understood. Using a heterozygous MFN2 R364W rat model, we reveal a compelling multi-scale narrative

that connects impaired organelle transport to axonal degeneration and behavioral decline. We observe progressive motor coordination deficits via grip strength testing at 16, 22, and 30 weeks, paralleled by auditory brainstem response (ABR) abnormalities, including reduced wave I amplitudes and change in threshold, indicative of auditory nerve and synaptic dysfunction. These findings reflect a length-dependent vulnerability across peripheral circuits. At the cellular level, live-cell high-resolution fluorescence microscopy imaging of dorsal root ganglion (DRG) neurons shows disrupted mitochondrial morphology, reduced motility, and altered lysosomal distribution in mutant animals, highlighting defects in organelle dynamics. By integrating subcellular imaging with a phenotyping study, this research bridges molecular pathology with functional outcomes. These data establish organelle trafficking deficits as a central mechanism of CMT2A and lay the foundation for targeted, mechanism-based therapeutic strategies.

Disclosures: W. Matysik: None. J. Cicero: None. C. Wu: None. K. sung: None. E. Orozco: None. K. Pak: None. K. Hitchcock: None. T. Velasquez: None. U. Manor: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.04/E33

Topic: C.06. Neuromuscular Diseases

Title: Involvement of motor neuron mitochondria in multiple models of charcot marie-tooth type 2d (CMT2D)

Authors: *J. FUNKE, R. W. BURGESS;
The Jackson Lab., Bar Harbor, ME

Abstract: Mutations in genes encoding mitochondrial proteins often cause peripheral neuropathy, including multiple subtypes of Charcot-Marie-Tooth (CMT). Mutations in *GARS* cause CMT type 2D. *GARS* encodes glycyl tRNA-synthetase, which is bifunctional: the gene encodes for both the cytosolic and mitochondrial versions. Mouse models of CMT2D exhibit a robust phenotype that appears to impact mitochondria in a cell-type specific manner. We used biochemical assays, immunofluorescence, and electron microscopy to assess mitochondrial quantity and morphology in neuronal cell bodies, axons and nerve terminals of affected motor neurons, and in unaffected cell types/tissues, of *Gars* mice . Additionally, compartmentalized hiPSC-derived motor neuron cultures will be used to measure mitochondrial phenotypes, such as respiration, in both neuronal cell bodies and axons/terminals. Mitochondria will also be isolated for proteomics. The findings from *in vitro* work will be extended *in vivo* by targeting mitochondria in motor neurons using a system that allows the cell-type-specific isolation of mitochondria via a Cre-inducible GFP-tagged outer membrane protein. We found that the distal axon, and the neuromuscular junction (synapse), of motor neurons in *Gars* mice exhibit significantly reduced mitochondrial number as compared to WT. As well, motor neuron soma also have reductions in mitochondrial quantity - indicating that the entire motor neuron is

impacted. Unaffected cell types in dorsal root ganglion neurons and other tissues did not display this mitochondrial reduction. *In vitro* mitochondrial phenotyping and *in vivo* immunoisolation of motor neuron mitochondria is currently in progress. Mitochondria are affected in a cell-type specific manner in *Gars* mice, like cell-type specific alterations involving cytosolic translation in these mice. Additionally, motor neurons are affected across all cellular compartments, indicating that mitochondria may be impacted directly due to gene function. As well, this could partially explain length dependent neuropathy and synaptic deficits observed in *Gars* mice.

Disclosures: J. Funke: None. R.W. Burgess: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.05/E34

Topic: C.06. Neuromuscular Diseases

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Companion Animal Fund, UW Madison, School of Veterinary Medicine
American College of Veterinary Surgeons Research Foundation
Wisconsin Alumni Research Foundation

Title: A naturally occurring canine model of peripheral neuropathy: genetic basis, transcriptomic profiling, and in vitro modeling through direct reprogramming

Authors: *R. S. ANDERSON¹, M. MOMEN¹, A. CHU¹, M. PATTERSON¹, S. ALE¹, A. CORONA¹, M. SUZUKI², J. P. SVAREN³, S. J. SAMPLE¹;

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Abstract: Hereditary peripheral neuropathies (HPNs) are debilitating disorders of the peripheral nervous system with no FDA-approved treatments. Late-onset peripheral neuropathy (LPN) is a prevalent canine HPN overrepresented in Labrador Retrievers. LPN is a life-limiting, length-dependent axonopathy presenting with upper respiratory obstruction due to laryngeal paralysis, esophageal dysmotility, and pelvic limb weakness. Identifying the genetic and molecular basis of LPN is key to establishing it as a large animal model for human HPNs. We hypothesize that Labrador LPN is an autosomal dominant Mendelian disease, as supported by pedigree analysis. A GWAS and fine-mapping of 137 LPN affected and 56 control Labradors identified an associated locus in a novel HPN candidate gene ($P=1.13E-7$). Long-read whole-genome sequencing (6 cases, 6 controls) revealed two structural variants (217 bp insertion and 42 bp deletion) and several single nucleotide variants segregating with phenotype. PCR genotyping (65 cases, 63 controls) showed the 217 bp insertion strongly associates with disease (OR=10.23;

$P < 0.0001$). To examine gene expression, RNA was extracted from spinal cord at L5, dorsal root ganglia (DRG), and sciatic nerve of cases (heterozygotes/homozygotes) and controls. The candidate gene is highly expressed in spinal cord and DRG, but no significant differential expression was seen between groups. DEXSeq and rMATS-turbo detected no significant exon usage or splicing changes. DESeq2 analysis revealed 3 differentially expressed genes in both spinal cord and DRG. In sciatic nerve, 185 differentially expressed genes were found; gene set enrichment indicated suppression of pathways involving actin cytoskeleton, extracellular matrix, and focal adhesion regulation, which are pathways intricately related to the candidate gene. As a next step, we developed a direct reprogramming system to generate canine-induced motor neurons from control and affected fibroblasts via *Ngn2*, *Sox11*, *Isl1*, and *Lhx3* overexpression with dorsomorphin and forskolin. Reprogrammed neurons express HB9, TUBB3, MAP2, ChAT, and SYN1, show ~50% induction efficiency, and are electrically active after 6 weeks. This methodology enables investigation of gene-specific cellular phenotypes crucial in understanding the disease. Our data support autosomal dominant inheritance of LPN with a strong association to a novel candidate gene showing mild mRNA alterations. Transcriptomic profiling highlights disrupted extracellular matrix and cytoskeletal pathways. Ongoing work uses induced-motor neurons to model disease mechanisms *in vitro*.

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Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.06/E35

Topic: C.06. Neuromuscular Diseases

Support: NIDCD R01DC011311

Title: Canine Model of Repetitive Pressure Trauma to the Left Recurrent Laryngeal Nerve

Authors: R. PANIELLO¹, A. FORREST², N. MYERS³, *J. M. BARKMEIER-KRAEMER⁴, J. VANDE GEEST²;

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Abstract: Vocal fold paralysis affects approximately 100,000 people each year in the US. In about 1/4 of these cases, a specific cause of the paralysis is never identified (referred to as “idiopathic”). The left side is injured ~70% of the time compared to the right, likely due to the different paths taken by the two recurrent laryngeal nerves (RLNs). Previously, our group found that some patients with idiopathic vocal fold paralysis (iVFP) have increased compliance of the aortic wall. We hypothesized that repetitive trauma caused by motion of a hypercompliant aortic

wall, impacting the left RLN with every heartbeat, could cumulatively lead to dysfunction of the nerve, and thereby cause paralysis of the left vocal fold. We sought to develop an animal model of such injury for further study. Our design involved three components: 1) a 3D-printed nerve sheath with two interlocking pieces; 2) a balloon catheter, implanted alongside the nerve within the sheath and extending externally; and 3) a servo actuator that alternately advanced and withdrew a syringe plunger, inflating and deflating the balloon. A pocket was sewn onto a canine anxiety jacket to hold the external device with its electronics and battery. Following early studies in 4 canines, we determined the optimal nerve sheath length (2.0 cm) and positioning (at the distal left vagus where the RLN branches). We then proceeded to implant and test the device on 14 additional dogs in 4 study groups: G1) 3.0mm diameter nerve sheath, endpoint at VFP onset (n=5); G2) same as G1, but balloon never inflated (control group for the setup alone) (n=3); G3) same as G1, but canine monitored for 90 days with device off to determine whether VFP would recover (n=3); G4) same as G1, but with 3.5mm diameter nerve sheath, reducing the pressure applied with each inflation (n=3). A thoracotomy was performed to place the balloon catheter and nerve sheath in the chest. A tracheostomy was performed to allow visualization of spontaneous vocal fold movement via a scope viewing from below. In G1 and G3, all 8 dogs developed VFP in 6.7 ± 2.4 (mean $\pm$ SD) days of device exposure. None of the 3 dogs in G3 had recovery of motion after 95.0 ± 10.8 days of observation. In G2, none of the 3 dogs developed VFP after 18.7 ± 3.5 days of device exposure, validating sheath and balloon placement alone did not lead to VFP. In G4, only one dog developed VFP, on day 17, while two continued to have movement despite 12.4 and 26.2 days of device exposure. These results indicate that the balloon catheter-nerve sheath arrangement in G1 and G3 provide a reasonable “semi-chronic” model of repeated pressure trauma to the left RLN, leading to VFP.

Disclosures: **R. Paniello:** A. Employment/Salary (full or part-time): Washington University, St Louis. 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.; NIDCD R01DC011311. **A. Forrest:** A. Employment/Salary (full or part-time): University of Pittsburgh. **N. Myers:** None. **J.M. Barkmeier-Kraemer:** A. Employment/Salary (full or part-time): University of Utah. 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.; NIDCD R01DC011311. **J. Vande Geest:** A. Employment/Salary (full or part-time): University of Pittsburgh. 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.; NIDCD R01DC011311.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.07/E36

Topic: C.06. Neuromuscular Diseases

Support: NIH 5T32 HD049336

Title: Sex-dependent efficacy of gonadal hormone treatment on behavioral recovery and reorganization of motoneuron morphology and spinal circuitry following peripheral nerve injury

Authors: *E. A. HIBBARD, D. R. SENGELAUB;
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Abstract: Reinnervation of distal targets after peripheral nerve injury (PNI) through the slow and imprecise regeneration of axons typically results in poor behavioral recovery. Axonal regeneration is outpaced by the rate of muscle and motoneuron atrophy, potentially resulting in irreversible deficits. Treatment with gonadal hormones has been reported to accelerate axonal regeneration, and may also encourage adaptive plasticity in critical spinal circuitry. For example, treatment with gonadal hormones after spinal cord injury attenuates injury-induced dendritic atrophy. In the current study, we examined the potential effects of sex and hormone treatment on both sensory and locomotor recovery after PNI, as well as the consequences they have for motoneurons and their premotor afferents. Young adult male and female Lewis rats were given a unilateral sciatic nerve transection repaired with either microsutures or Nerve Tape (BioCircuit Technologies) and immediately implanted with subcutaneous Silastic capsules filled with either estradiol, dihydrotestosterone or left blank. Locomotor and sensory behavioral testing were performed weekly. At either 8- or 12-weeks post-injury, motoneurons whose axons had regenerated into the anterior tibialis muscle were retrogradely labeled with cholera toxin-conjugated horseradish peroxidase (BHRP), allowing for assessment of spinal location and dendritic morphology. Premotor neuron distribution in the lumbar spinal cord was assessed using injection of pseudorabies virus (PRV) strain 152 into the flexor digitorum brevis muscle. Locomotor function of hormone-treated males was improved compared to that of untreated males, whereas females consistently improved with or without treatment. In both sexes, hormone treatment did not induce pro-nociceptive effects compared to untreated animals. BHRP labeling showed that motoneuron axons had regenerated into either the correct or incorrect muscle, and in both cases, motoneurons exhibited dendritic reorganization. Injury and/or regeneration-induced changes in dendritic organization were reflected in the redistribution of PRV-labeled premotor neurons. This reorganization differed by sex, with premotor neuron density in L3-4 increasing in females but decreasing in males. Regardless of sex or treatment, animals with poor locomotor recovery exhibited larger increases in premotor neuron density in L3-4 than well-recovered animals, suggesting the gain of density in this region is a maladaptive plasticity. Overall, sex and/or hormone-mediated plasticity in spinal circuitry after axonal regeneration may contribute to behavioral deficits or recovery after PNI.

Disclosures: E.A. Hibbard: None. D.R. Sengelaub: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.08/E37

Topic: C.06. Neuromuscular Diseases

Support: NSF CBET2144472
NIH R15DC017866

Title: Noninvasive force measurement in mice for quantitative and longitudinal assessment of nerve injury recovery

Authors: G. LICWINKO¹, A. YEN¹, J. POTHUGANTI¹, P. CHAN², W.-M. YU¹, *V. CHEN¹;
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Abstract: Peripheral nerve injuries often result in incomplete motor recovery, and preclinical models could be useful for evaluating new and novel regenerative strategies. As the functional recovery process is typically assessed through behavioral metrics that are indirect, the sensitivity and reproducibility in the motor assessments are usually limiting. To address this, we developed and validated a mouse sciatic nerve crush model with in vivo isometric force output as a direct readout of functional recovery. This method could provide objective physiological data and allow for within-subject longitudinal assessment. The force measurement in mice was particularly challenging due to their low force output, but mouse models offer valuable opportunities for genetic knockout studies. The sciatic nerves of shaved male and female CD1 mice (18-24 weeks old) were located using tweezer probes and marked with a permanent marker. The mice were then attached to customized 3D-printed fixation apparatus with a customized force sensor module to measure isometric plantarflexion force by stimulating the sciatic nerve noninvasively over the marked skin using wire electrodes which were securely fixed on the 3D-printed apparatus. The mice then underwent the standard procedure of unilateral sciatic nerve crush injury as reported in an earlier study. Post-injury measurements were collected longitudinally up to 28 days. All experiments were conducted under isoflurane anesthesia and the contralateral sides of the mice served as controls. Following nerve crush, mice exhibited a significant decrease in isometric force output and gradual recovery was observed over time through force measurements collected with our fixation apparatus and force sensor module. We observed that the recovery of neuromuscular force is closely correlated with nerve regeneration and remyelination, as demonstrated by histological analyses using various markers to assess axonal growth and myelin sheath formation. The contralateral force showed no significant changes across time and the force measurement system demonstrated high intra-animal reproducibility. This study presents a noninvasive isometric force measurement method to assess motor function recovery in mice following nerve injury. Our in vivo approach enables quantitative, longitudinal tracking of functional recovery without sacrificing animals during the investigation process. This model enhances the translational relevance of peripheral nerve injury research and provides a sensitive, low-variability platform for testing pro-regenerative therapies.

Disclosures: G. Licwinko: None. A. Yen: None. J. Pothuganti: None. P. Chan: None. W. Yu: None. V. Chen: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

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Program #/Poster #: PSTR270.09/E38

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant R21NS072955-01A1

Title: Sox2 expression in long-gap peripheral nerve repair with growth factor-releasing conduits remyelination

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Abstract: Long- gap (>3cm) peripheral nerve injuries affect thousands each year, and currently have few surgical options for repair and functional recovery. Autografts remain the gold standard despite associated morbidity. Nerve conduits are desirable off-the-shelf alternatives but offer very limited functional recovery in long gap repairs. Those that incorporate growth factors such as glial-derived nerve growth factor (GDNF) and brain-derived neurotrophins have shown promising results. We have reported that pleiotrophin (PTN) and GDNF synergistically promote axon regeneration across a 4 cm gap, but most axons remained unmyelinated and wrapped into Remak bundles. In a recent study we evaluated a 4 cm nerve gap injury in the rabbit (n=21), repaired with either an autograft, an empty conduit, a conduit filled with slow-release Neuregulin 1 Type III (NRG) or a combination of PTN and NRG. We found that both the NRG and NRG+PTN groups increased axon regeneration and provided signaling needed for effective Remak unbundling by axon sorting, but re-myelination remained limited. SOX2 is a known inhibitor of nerve of Schwann cell differentiation and myelination typically expressed during the initial stage of injury during debris clearance, and SOX10, a positive regulator of myelination also expressed in mature Schwann cells. We reasoned that the activation of genes that mediate Schwann cell differentiation from repair to re-myelination phenotypes such as SOX2 and SOX10 remained altered in our long-gap conduit repair explaining the low re-myelination rate, despite growth factor release and axonal regeneration. Using immunocytochemistry and quantitative analysis we are currently evaluating the expression of SOX2 in the empty and growth-factor regenerated nerves and comparing to the autograft group. This study will shed light on their possible association with re-myelination of regenerated nerves across a long gap injury.

Disclosures: K. Jackson: None. M.I. RomeroOrtega: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); CTO for Pioneer Neurotech Inc.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.10/E39

Topic: C.06. Neuromuscular Diseases

Title: Base exchange inhibitors of SARM1 form mononucleotide adducts and activate SARM1 in vivo.

Authors: H. T. REARDON¹, C. GUIJAS¹, J. R. CLAPPER¹, ***J. CALLENDER**¹, E. KOZINA¹, F. SARABIA¹, C. HUTTON¹, D. HERBST¹, L. PEREHINETS¹, D. KUMMER¹, J. L. BLANKMAN¹, J. WIENER¹, T. SAMAD²;

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Abstract: Activation of the NAD hydrolase SARM1 causes neuronal degeneration. NAD binding to an allosteric site maintains SARM1 autoinhibition while NMN binding enables activation. We evaluated SARM1 base exchange inhibitors (BEI) that exchange with nicotinamide in the SARM1 catalytic site to form inhibitor adducts. We found that BEI paradoxically activated SARM1 in some contexts, increasing both the SARM1 product cADPR and ratio to substrate, cADPR/NAD, in uninjured sciatic nerve and in healthy animals. Knock-in mice containing a SARM1 mutation that lacks NADase activity (E642A) were protected from elevated cADPR and cADPR/NAD in sciatic nerve after BEI dosing. BEI formed mononucleotide (MN) adducts in vitro and in vivo, similar to the activating forms of the neurotoxins Vacor and 3-acetylpyridine. These findings highlight the low specificity of nicotinamide-related metabolism and SARM1's susceptibility to activation by pyridine derivatives.

Disclosures: **H.T. Reardon:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **C. Guijas:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **J.R. Clapper:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **J. Callender:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **E. Kozina:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **F. Sarabia:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **C. Hutton:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **D. Herbst:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **L. Perehinets:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **D. Kummer:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **J.L. Blankman:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **J. Wiener:** A. Employment/Salary (full or part-time): Lundbeck La Jolla Research Center, Inc. **T. Samad:** A. Employment/Salary (full or part-time): H. Lundbeck A/S.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.11/E40

Topic: C.06. Neuromuscular Diseases

Title: Identification of a non-covalent, allosteric SARM1 inhibitor that can prevent axonal damage in vitro and in vivo

Authors: *W. FAN^{1,2}, Y. XU², L. LIU², Q. WANG², L. YU¹, S. AGARWAL²;

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Abstract: Axonal degeneration is a hallmark pathological feature of numerous neurodegenerative diseases, including amyotrophic lateral sclerosis (ALS), Parkinson's disease, and chemotherapy-induced peripheral neuropathy (CIPN). The sterile alpha and TIR motif-containing 1 (SARM1) protein is a NADase highly expressed in the nervous system and serves as a central executioner of Wallerian degeneration, a conserved axon degeneration pathway. In healthy neurons, SARM1's NADase activity is tightly suppressed by an intrinsic auto-inhibitory mechanism regulated by NAD⁺. Pathological conditions that disrupt soma-to-axon protein trafficking—such as mechanical axonal injury—can lead to the depletion of the labile enzyme NMNAT2 within axons. Loss of NMNAT2 causes an increase of local NMN/NAD⁺ ratio, which in turn activates SARM1. Rodents completely lacking the *Sarm1* gene develop normally and exhibit remarkable resistance to neuronal damage in a variety of neurodegenerative models, including ALS, CIPN, traumatic brain injury, glaucoma etc.. Moreover, gain-of-function mutations in *SARM1* have been identified in ALS patients. Herein we report the discovery of a non-covalent, brain penetrant, allosteric inhibitor of SARM1—Compound A—that does not engage the enzyme's catalytic site nor rely on base-exchange activity for its mechanism of action. **Methods:** Compound's in vitro activity was tested in primary mouse DRG or human iPSC derived motor neuron that underwent mechanical damage. Compound's effect on axon protection was measured by degenerative index of the axon. Compound A's in vivo activity was tested in models of sciatic nerve axotomy (SNA), neurotoxin-induced acute neuronal injury and in paclitaxel induced peripheral neuropathy model. Compound A was administered through oral gavage, Compound's effect on SARM1 activity was measured through the cADPR level in the SNA model and by mouse mortality in the neuron toxin model. **Results and conclusions:** In vitro, Compound A effectively prevented axonal degeneration in primary mouse dorsal root ganglion (DRG) neurons (IC₅₀=140nM) and in human induced pluripotent stem cell (iPSC)-derived motor neurons (IC₅₀=8nM) following mechanical injury. In vivo, orally dosing of Compound A provided robust neuroprotection in multiple models of axonal damage.

Disclosures: W. Fan: A. Employment/Salary (full or part-time): SIRONAX. Y. Xu: A. Employment/Salary (full or part-time): SIRONAX. L. Liu: A. Employment/Salary (full or part-time): SIRONAX. Q. Wang: A. Employment/Salary (full or part-time): SIRONAX. L. Yu: A. Employment/Salary (full or part-time): SIRONAX. S. Agarwal: A. Employment/Salary (full or part-time): SIRONAX.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.12/E41

Topic: C.06. Neuromuscular Diseases

Support: CSIR-SRF Fellowship
IA/TSG/20/1/600137

Title: Microtubule-based approach to understand the mechanistic onset of paclitaxel-induced peripheral neuropathy

Authors: *A. BARDHAN¹, M. MAGIERA², C. JANKE², P. PULLARKAT^{3,4}, A. GHOSE¹;
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Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a progressive degeneration of peripheral nerves in patients receiving chemotherapy, forcing discontinuation of otherwise successful therapy. The lack of well-structured assessments and a thorough understanding of the mechanistic onset of CIPN leaves very little possibility to intervene throughout the disease's progression, which frequently affects the patient's quality of life. Paclitaxel (PTX), a widely used drug for treating breast and ovarian cancers, presents a high prevalence of CIPN in cancer patients. It predominantly affects long sensory neurons presenting itself as a “glove and stocking” neuropathy, indicating a role for microtubule (MT)-mediated axonal transport in the onset of neuropathy. Even though we know that PTX binds and stabilizes microtubules, resulting in cell cycle arrest in cancerous cells, the cellular mechanisms underlying its neurotoxicity are not known. We therefore use primary adult murine dorsal root ganglion neuronal cultures, cell types affected in CIPN patients, to understand the pathogenic effect of PTX in neurons. Chronic *in vitro* treatment with PTX induced focal axonal swellings (FAS) and lysosomal stalling, indicative of neurotoxicity, which is concomitant with increased levels of several tubulin post-translational modifications (PTMs). Delta2 ($\Delta 2$) modification, an irreversible PTM, showed the most pronounced elevation. Increasing $\Delta 2$ levels by overexpressing either recombinant $\Delta 2$ or CCP1, an enzyme that drives tubulin $\Delta 2$ modification, was sufficient to induce axonopathy. Pharmacological reduction of the $\Delta 2$ levels reduced PTX-induced FAS in the neurons, thereby indicating functional relevance of $\Delta 2$ in causing neuropathy. Specifically, shRNA-mediated knockdown of CCP1 reduced $\Delta 2$ levels and alleviated swellings and loss of lysosomal motility induced by PTX. Importantly, CCP1 knockdown did not interfere with the therapeutic efficacy of PTX in cancer cells. Altogether, our findings indicate that the pathogenic role of PTX converges on impaired axonal transport associated with distinct neurodegenerative phenotypes, and CCP1 could serve as a potential mechanistic target of PTX in orchestrating peripheral neuropathy.

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Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.13/E42

Topic: C.06. Neuromuscular Diseases

Support: NIH/NIDDK R01DK135970
NIH/NIDDK Diacomp 21AU4165

Title: Impaired Mitochondria-derived Microvesicle Transfer from Schwann Cells Drives Axonal Dysfunction in Diabetic Neuropathy

Authors: *X. LIU¹, M. CHOPP^{2,3}, B. FAN², L. WANG², Y. ZHANG², J. LANDSCHOOT-WARD², Z. ZHANG²;

²Neurol., ¹Henry Ford Hosp., Detroit, MI; ³Dept. of Physics, Oakland Univ., Rochester, MI

Abstract: Mitochondrial-derived microvesicles (MDVs) enriched with mitochondrial components are emerging as important mediators of intercellular communication, particularly in tissues with high metabolic demands. Schwann cells (SCs) play a vital role in supporting metabolic homeostasis and axonal function. Mitochondrial dysfunction in SCs contributes to the pathogenesis of diabetic peripheral neuropathy (DPN). However, the specific role of SC-derived MDVs (SC-MDVs) in DPN remains elusive. Here, we tested the hypothesis that SC-MDVs are essential to sciatic nerve function by transferring mitochondrial components to dorsal root ganglion (DRG) neurons. SC-MDVs and exosomes were isolated from SC-conditioned media via differential ultracentrifugation and characterized with nanoparticle tracking analysis (NTA), transmission electron microscopy (TEM), and immunoblotting. Mitochondrial function within SC-MDVs was assessed by measuring mitochondrial membrane potential ($\Delta\Psi_m$), ATP, and reactive oxygen species (ROS) levels. Primary DRG neurons cultured in a microfluidic device were treated with SC-MDVs and axonal growth was evaluated. NTA showed that the size of SC-MDVs (200-400 nm) was larger than exosome sizes (30-150 nm). SC-MDVs were enriched with mitochondrial proteins and contained functional mitochondria as measured by flow cytometry. Compared to SC-MDVs from cultured SCs under normal glucose level, SC-MDVs isolated from SCs treated with high glucose (HG, 25 mM) exhibited a 50% reduction in TOM20 ($p < 0.05$) and loss of $\Delta\Psi_m$ ($p < 0.01$), 40% decrease in ATP production ($p < 0.01$) and 2.1-fold increased ROS content ($p < 0.001$). Strikingly, MDVs isolated from the sera of diabetic patients and high-fat diet (HFD)/streptozotocin (STZ)-induced diabetic mice exhibited a 60-70% reduction in Argonaute2 (Ago2) expression ($p < 0.01$), a core factor of the RNA-induced silencing complex. Furthermore, when these dysfunctional MDVs derived from SC-specific Ago2-deficient sciatic nerve tissues were applied to the DRG neurons, a significant suppression of axonal growth was observed ($p < 0.01$). In contrast, administration of naïve SC-MDVs effectively restored axonal mitochondrial function, increased ATP levels by 2.5-fold ($p < 0.01$), while reduced HG-induced NLRP3 (NOD-like receptor protein 3) inflammasome activation by 55% measured by immunoblotting in DRG neurons under HG conditions. Our results show that SC-MDVs function as critical mediators of SC-axon metabolic coupling and hyperglycemia-induced SC-MDVs could contribute to DPN pathogenesis. Thus, developing MDV-based therapies to target mitochondrial transfer provides a new treatment for DPN.

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Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.14/E43

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant R01NS069726
NIH Grant R01NS0945639
AHA Grant 16GRNT31280014

Title: Characterizations of motor neurons derived from iPSCs of a novel sensory motor neuropathy caused by a NAMPT mutation

Authors: *Z. ZHANG¹, B. DING², S. DING³;

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Abstract: In mammalian cells, NAD⁺ is primarily synthesized through the salvage pathway, where nicotinamide phosphoribosyltransferase (NAMPT) is the rate-limiting enzyme. Increasing evidence suggests a neuronal protective role of NAMPT in a range of neurological diseases such as ischemic stroke, Alzheimer's disease (AD), and amyotrophic lateral sclerosis (ALS). In our recent study, we discovered a novel sensory motor neuropathy attributed to a homozygous missense mutation (c.472.C>G, p.P158A) in the coding region of the NAMPT gene. To study the functional change of motor neurons caused by the P158A mutation, in current study, we reprogramed the fibroblasts collected from the patient into induced pluripotent stem cells (iPSCs) and subsequently differentiate them into motor neurons (MNs). Using a variety of approach, we investigated the bioenergetic and metabolic effects of P158A mutation in NAMPT on iPSC derived MNs. Our results demonstrated that the P158A mutation in NAMPT disrupts cellular metabolism and causes mitochondrial dysfunction in iPSC-derived MNs. The P158A mutation also leads to aberrant synaptic development and Ca²⁺ signaling and induces cell senescence. Overall, our study reveals an undiagnosed motor neuropathy caused by P158A mutation in NAMPT, confirms NAMPT as a key enzyme in neuronal metabolism and functions and indicates that targeting NAMPT might be a novel therapeutic strategy for motor neuron disease.

Disclosures: Z. Zhang: None. B. Ding: None. S. Ding: None.

Poster

PSTR270: Peripheral Nerve Traumas and Neuropathies

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR270.15/E44

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Raft into trouble: connexin 43 redistribution into lipid rafts in the nodose ganglion as a potential mechanism for neuroimmune crosstalk in allergic airway inflammation

Authors: *A. ALVARADO VAZQUEZ¹, A. PATEL², Y. MILLER²;

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Abstract: Vagal sensory neurons innervating the lungs actively participate in modulating airway inflammation, yet how peripheral immune signals reshape neuronal function remains poorly understood. In the nodose ganglion, expression of Connexin 43 (Cx43), a gap junction protein involved in intercellular communication, modulation of neuronal activity, and inflammatory responses, is normally restricted to satellite glial cells but not neurons. Lipid rafts are specialized membrane microdomains enriched in cholesterol, sphingolipids, and signaling molecules that serve as platforms for organizing and amplifying cellular signaling. The localization of proteins such as Cx43 into lipid rafts facilitates more efficient and selective signal transduction, particularly in response to extracellular stimuli. Here, we investigated whether exposure to house dust mite (HDM), a clinically relevant aeroallergen, alters the subcellular distribution of Cx43 in the nodose ganglion. We used a mouse model of allergic airway inflammation induced by repeated intranasal instillations of HDM extract. Nodose ganglia were collected, fixed in 4% paraformaldehyde, cryosectioned, and processed for analysis. A proximity ligation assay (PLA) was employed to detect co-localization of Cx43 with cholera toxin B, a well-established lipid raft marker. Satellite glial cells were identified by immunostaining for fatty acid binding protein 7 (FABP7). Our findings show that HDM exposure significantly increased the localization of Cx43 to lipid rafts without altering total Cx43 expression levels. Furthermore, we observed enhanced Cx43-lipid raft associations in both FABP7-positive satellite glial cells and glia-negative regions, potentially neurons. These observations suggest that HDM-induced redistribution of Cx43 into lipid rafts could enhance neuronal and glial responsiveness to pro-inflammatory signals, thereby priming vagal sensory neurons and their associated glia for heightened neuroimmune reactivity in the context of allergic airway inflammation.

Disclosures: A. Alvarado vazquez: None. A. Patel: None. Y. Miller: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.01/E45

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Complementary iPSC-derived neuronal assays for modelling neurodegeneration and repair

Authors: M. HEMBURY¹, J. VAN OFFENBEEK¹, *C. J. M. LOOMANS¹, B. TORROBA¹, D. KETSKEMETY¹, M. PLIEGO CABALLERO¹, K. LO¹, M. MUSTERS¹, M. BSIBSI², M. VLAMING²;

²Biotherapeutics Drug Discovery, ¹Charles River Labs., Leiden, Netherlands

Abstract: Neurite degeneration is a hallmark of early-stage neurodegenerative disorders such as Alzheimer's disease (AD), Parkinson's disease (PD), and amyotrophic lateral sclerosis (ALS). Therefore, developing disease-relevant phenotypic assays is essential for identifying novel drug candidates targeting neurodegeneration and promoting neuronal repair. The aim of this study was to develop neurite outgrowth assays using human induced pluripotent stem cell (iPSC)-derived neurons to model both neurodegenerative and regenerative processes. We established two complementary assays. In the first, iPSC-derived dopaminergic neurons were cultured as three-dimensional aggregates and exposed to chemically defined triggers mimicking disease-relevant molecular mechanisms. These cultures were evaluated and validated using immunocytochemistry, multi-parametric high-content imaging, multivariate data analysis, and quantification of Neurofilament light chain (NfL) release as readouts for neurodegeneration. Live fluorescent dyes enabled multiplexed assessment of neurite features, including length, density, branching, morphology and cell viability. High-content imaging was performed using high-resolution imager (Yokogawa CV8000) and image datasets were analyzed via an in-house algorithm using advanced image analysis tools (Arivis). The use of 3D neuronal aggregates enhances physiological relevance and network complexity and enables robust detection of disease phenotypes and drug responses. To model neuronal repair, we established an axotomy assay using both wild-type and disease-relevant mutant iPSC-derived neurons, such as TDP43-M337V motor neurons, associated with ALS. Cells were cultured in eNUVIO OMEGA96™ dual-chamber microfluidic devices, allowing for selective axon severing and real-time monitoring of axonal regrowth via live imaging. Quantitative metrics, including regeneration rate, length, and morphological complexity, were extracted to evaluate regenerative capacity under disease-relevant conditions. Together, these two assays offer a powerful platform for phenotypic screening, capturing both neurodegeneration and regenerative responses. Their scalability and complementary readouts support early-phase drug discovery in Parkinson's disease, amyotrophic lateral sclerosis, and related disorders.

Disclosures: M. Hembury: None. J. van Offenbeek: None. C.J.M. Loomans: None. B. Torroba: None. D. Ketskemety: None. M. Pliego Caballero: None. K. Lo: None. M. Musters: None. M. Bsibsi: None. M. Vlaming: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.02/E46

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Fundação de Amparo à Pesquisa de São Paulo (FAPESP) 2016/19391-8
Fundação de Amparo à Pesquisa de São Paulo (FAPESP) 2021/14831-8
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Fundação de Amparo à Pesquisa de São Paulo (FAPESP) 2024/05640-2

Title: From Glycation to Neurodegeneration: The Impact of Advanced Glycation End-Products on Neuronal Structure and Function

Authors: *G. DE ASSIS SILVA¹, M. CAPRIO SCHIESS¹, M. BUFALO², M. MEDINA DE SOUZA², A. CHUDZINSKI-TAVASSI³, V. OLZON ZAMBELLI¹;

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Abstract: Advanced glycation end products (AGEs) are formed through the irreversible attachment of sugar molecules to proteins such as collagen. Studies have shown that AGEs are associated with the development of peripheral neuropathy and pain. Despite this evidence, the mechanisms underlying AGE-induced neuronal hypersensitization and axonal degeneration remain unknown. Thus, this study aimed to evaluate the role of AGEs in the neuronal degeneration processes associated with hypernociception. SH-SY5Y cell line differentiated into sensory-like neurons incubated with glycated collagen (GC) was used as a model to investigate the effect of AGEs on cytoskeletal morphology, by evaluating actin filament and β III-tubulin expressions; pro-nociceptive signaling pathways, by evaluating sensitivity to PGE2-induced calcium influx, and mitochondrial function, by evaluating ROS production, mitochondrial fragmentation, and membrane potential. Finally, we assessed the effect of AGEs on axonal degeneration using a mouse *ex vivo* model of peripheral nerve explant. The axonal integrity was evaluated by α -tubulin, acetylated tubulin, and myelin basic protein (MBP) expression and histology. Normal collagen (NC) was used as a control. Although we did not observe changes in cell viability, treatment with GC decreased the expression of structural proteins, such as actin filaments ($30.7 \pm 2.3\%$) and β III-tubulin ($25.2 \pm 7\%$), compared to the control, indicating damage to the cytoskeletal protein structure. Moreover, GC treatment hypersensitized cells to calcium influx by 130% when exposed to PGE2, compared to the control. In addition, GC altered mitochondrial function by increasing ROS production ($20 \pm 5.37\%$), mitochondrial fragmentation (increased counting and decreased mean area), and reducing the membrane potential ($79.28 \pm 10.64\%$). Mitochondrial dysfunction was reversed by RAGE antagonist (FPS-ZM1). Sciatic nerve explant model analysis showed that GC decreases the MBP levels ($24.64 \pm 6.64\%$), as well as the acetylated tubulin/ α -tubulin ratio ($49.2 \pm 13.66\%$), compared to NC. The histological analysis revealed that GC increased morphological changes in fibers, demonstrated by infoldings and loss of axonal myelin. These data indicate that glycation induces neuronal hypersensitization and impairment in mitochondrial functions. Moreover, GC induces disruption of structural proteins in sensory neurons in culture and in axonal fibers *ex vivo*. Taken together, our data may contribute to the establishment of a new study platform for neurodegenerative diseases, as well as to the understanding of the role of AGEs in this process.

Disclosures: G. De Assis Silva: None. M. Caprio Schiess: None. M. Bufalo: None. M. Medina de Souza: None. A. Chudzinski-Tavassi: None. V. Olzon Zambelli: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.03/E47

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: AHA predoctoral fellowship 24PRE1188417

Title: Reexamining mechanisms of axon degeneration vs resilience: the role of mitochondrial NAD⁺

Authors: ***L. BARBAR**¹, Y. SASAKI², J. BLOOM³, J. D. MILBRANDT⁴, A. DIANTONIO⁵;
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Abstract: Loss of nicotinamide adenine dinucleotide (NAD⁺) is a hallmark of axon degeneration and neurodegenerative disease. NAD⁺ is an essential metabolite, necessary for fundamental cellular functions including ATP synthesis. NAD⁺ pools are compartmentalized into the nucleus, cytosol, and mitochondria, with different roles in each compartment. Loss of cytosolic NMNAT2, the major NAD⁺-synthesizing enzyme in axons, leads to the activation of SARM1, an NAD⁺-cleaving enzyme, which results in catastrophic NAD⁺ depletion and axon loss. Knockout of *Nmnat2* in mice shows widespread axonopathy and perinatal lethality. In the absence of SARM1, mice lacking NMNAT2 live a normal lifespan with no overt phenotype, indicating that cellular and metabolic processes are maintained despite the lack of cytosolic NAD⁺ synthesis by NMNAT2. Given the central role of NAD⁺ in cellular and metabolic processes, this suggests that other subcellular compartments contribute to the cytosolic NAD⁺ pool. Mitochondria hold the largest NAD⁺ pool in neurons. While NAD⁺ transporters facilitating mitochondrial import in mammalian cells were recently identified, the possibility of mitochondrial NAD⁺ export to the cytoplasm remains unexplored. We find that loss of mitochondrial NAD⁺-synthesizing NMNAT3, in *Nmnat2:Sarm1* double knockout mice, leads to nerve conduction deficits and paralysis, suggesting the presence of a mechanism for mitochondrial NAD⁺ export to the cytosol. Our study further investigates NAD⁺ exchange between mitochondria and cytosol as a candidate mechanism for axon resilience through different *in vivo* and *in vitro* approaches.

Disclosures: L. Barbar: None. Y. Sasaki: None. J. Bloom: None. J.D. Milbrandt: None. A. DiAntonio: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.04/E48

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NSFC Grant 32325016
NSFC Grant 32270812
NSFC Grant 22337005
Basic Research Pioneer Project of Science and Technology Commission
of Shanghai Municipality

Title: Fbxo21 deficiency confers neuroprotection in nerve injury via regulating the ubiquitination and turnover of nmnat2

Authors: *Y. (. FANG;

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Abstract: NMNAT2 is an essential but labile protein required for axon integrity. It is rapidly degraded after nerve injury, promoting axon degeneration. However, the mechanisms regulating NMNAT2 ubiquitination and turnover in neurons remain unclear. In this study, we identify the F-box protein FBXO21 as an NMNAT2-binding protein and its deficiency confers axonal protection via increasing NMNAT2 abundance. FBXO21 recruits SKP1, CUL1 and RBX1 to form an SCF^{FBXO21} complex, which promotes NMNAT2 ubiquitination *in vivo* and *in vitro*. SCF^{FBXO21} ubiquitinates NMNAT2 at K155 within an isoform-specific targeting and interaction domain of the family of NMNATs, which underlies the unique labile nature of NMNAT2. The ubiquitination-deficient NMNAT2-K155R exhibits substantially reduced protein turnover and enhanced axon-protective capacity. Finally, in *Fbxo21* knockout mice, NMNAT2 levels are markedly increased and the survival of injured sciatic nerves is significantly prolonged. Collectively, our findings reveal a crucial role of FBXO21 in axon degeneration, highlighting the SCF^{FBXO21} complex as a potential target for modulating NMNAT2-dependent axon survival.

Title: Upstream open reading frame-mediated translational regulation of DLK in response to axonal injury

Authors: *Y. OH, Y. CHO;

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Abstract: Dual leucine zipper kinase (DLK), encoded by *Map3k12*, serves as a master regulator of both regenerative and degenerative pathways in neurons, acting as a double-edged sword in axonal responses. Given its opposing roles, DLK protein levels must be tightly regulated, and most studies have focused on protein degradation pathways. However, how DLK expression is regulated at the translational level remains poorly understood. In this study, we found that DLK protein levels significantly increase following axonal injury in mouse sciatic nerves. Notably, DLK is upregulated under both pro-regenerative and degenerative stresses, but with distinct spatial patterns, as its increase is localized to axons after axotomy, whereas it is primarily observed in the cell body following vincristine treatment or NGF deprivation. To understand the basis of this context-specific translational regulation, we analyzed Map3k12 transcript variants with and without upstream open reading frames (uORFs), which are known to modulate translation efficiency under cellular stress conditions. In naïve embryonic neurons, we found evidence suggesting that uORFs modulate DLK translation. Furthermore, the injury-induced increase in DLK protein levels was suppressed by PERK inhibition, implicating a role for signal-dependent alternative translation mechanisms. Our ongoing work aims to elucidate the molecular mechanisms governing uORF-dependent translational control of DLK after injury. Ultimately, this study seeks to uncover how stress-specific translational regulation of DLK contributes to the balance between axon regeneration and degeneration, potentially offering a therapeutic strategy to modulate its dual functions by targeting specific uORF-containing transcripts.

Disclosures: Y. Oh: None. Y. Cho: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.06/E50

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Sarm1 promotes schwann cell-mediated nerve repair mechanisms via transcriptional reprogramming

Authors: *M. KANG^{1,2}, S. JANG¹, H. PARK^{1,2}, J. SHIN^{1,2};

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Abstract: Axon regeneration occurs after axonal injury in the peripheral nervous system (PNS). Axon degeneration precedes axon regeneration and is essential for robust regeneration because delayed degeneration hinders the regeneration process. Reprogramming of Schwann cells (SC)

into repair SC also contributes to axon regeneration by providing mechanically and chemically supportive environment. Yet, the molecular mechanism orchestrating these related events is unclear. In this study, we hypothesized that a SARM1-dependent pathway executing the axonal death initiates crosstalk between axons and SC for successful axon regeneration. To investigate SARM1-dependent differentially expressed genes (DEG) under axonal injury conditions, we performed transcriptome analysis using SARM1 knockout (KO) mice, where significantly delayed axon degeneration and regeneration were observed. RNA-sequencing data from the sciatic nerves present that genes associated with demyelination and SC reprogramming are SARM1-dependently regulated; at seven days after injury, neurotrophic factor secretion and membrane remodeling are highly expressed in the SARM1-dependent DEGs. Whereas regeneration-associated genes were expressed in SARM1-independent manner in dorsal root ganglia (DRG) until seven days after nerve crush, the activation of immune system showed a SARM1-dependency. To sum up, these results propose that SARM1-dependent axon degeneration induces the SC reprogramming and the immune cell activation for regeneration. Furthermore, we suggest that this mechanism is related to changes in local immunity in the DRG during persistent axon regeneration.

Disclosures: M. Kang: None. S. jang: None. H. Park: None. J. Shin: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.07/E51

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Dose-dependent and robust target engagement of a novel brain penetrant SARM1 inhibitor (SIR2501) in healthy adult participants

Authors: P. J. RYAN¹, D. GRACHEV², W. FAN³, L. SONG³, *R. SUTHERLAND², S. AGARWAL², Y. SHEN³;

¹Florey Inst. of Neurosci. and Mental Hlth., Parkville, Australia; ²Sironax, Waltham, MA;

³Sironax, Beijing, China

Abstract: SARM1 is a nicotinamide adenine dinucleotide (NAD)ase that is predominantly expressed in neurons and can be activated by various pathological insults. The activation of SARM1 in neurons leads to a rapid loss of NAD⁺, which is an essential component for the generation of adenosine triphosphate (ATP). Loss of ATP results in axonal and neuronal death. Animal studies have demonstrated that inhibiting SARM1 activity provides neuron protective effects in disease models such as traumatic brain injury, chemotherapy induced peripheral neuropathy (CIPN), amyotrophic lateral sclerosis (ALS), and other neurodegenerative disorders. An FIH Phase I study to investigate the safety, pharmacokinetics, and target engagement of investigational product SIR2501 in healthy adult participants was conducted in Australia and China. Results from Australian cohorts showed that the SIR2501 was generally safe and well

tolerated when administered orally as a single dose (up to 600 mg) or multiple doses (up to 150 mg QD) in healthy adult participants; general dose proportionality to systemic exposure in single ascending dose (SAD; from 10 to 600 mg) and multiple ascending dose (MAD; from 30 to 150 mg, QD) studies; limited food effect; potential permeability across the blood-brain barrier; no impact on CYP3A activity; and robust PD effects with greater than 90% target engagement (inhibition of cADPR in stimulated peripheral blood mononuclear cells [PBMCs]) achieved at the 30-mg QD dose. The I_{max} PD model has demonstrated a concentration-dependent target engagement of SIR2501. The results indicated that a 50% reduction of cADPR, identified as the half-maximal inhibitory concentration (IC₅₀), was attained at a SIR2501 plasma concentration of 55.2 ng/mL. Additionally, the IC₈₀ and IC₉₀ were reached at 170 ng/mL and 329 ng/mL, respectively. Results from the China cohort are currently under analysis. This observation supported the notion that SIR2501 interacted with SARM1, as intended, inhibiting its NADase activity. SIR2501 demonstrated dose-dependent and robust target engagement *ex vivo* in stimulated PBMCs, indicating it is a potent inhibitor of SARM1 and suggesting its therapeutic potential to modulate SARM1 NADase activity in humans to protect against axonal degeneration. These findings support the further evaluation of SIR2501 in clinical development, including the treatment of ALS, and prevention or lessening the severity of CIPN.

Disclosures: **P.J. Ryan:** 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.; Sironax. **D. Grachev:** F. Consulting Fees (e.g., advisory boards); Sironax. **W. Fan:** A. Employment/Salary (full or part-time); Sironax. **L. Song:** A. Employment/Salary (full or part-time); Sironax. **R. Sutherland:** A. Employment/Salary (full or part-time); Sironax. **S. Agarwal:** A. Employment/Salary (full or part-time); Sironax. **Y. Shen:** A. Employment/Salary (full or part-time); Sironax.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.08/E52

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Discovery of Novel Small Molecule SARM1 Inhibitors Blocking NAD⁺ Hydrolysis to Prevent Axon Degeneration

Authors: ***J. HSU**, T. BURDETT, J. DANA O, X. WANG, J. BECKSTEAD, R. RIETMEIJER, Y. KIM, R. BRITTON, S. SAMBASHIVAN;
Nura Bio, South San Francisco, CA

Abstract: Axon degeneration is a hallmark of neurological diseases, leading to neural circuit disconnection and subsequent nervous system dysfunction. SARM1 has been identified as a key driver of programmed axon death that is activated upon local metabolic dysregulation. Genetic

knockout of SARM1 is protective across multiple preclinical models of neurodegenerative injury and disease, including, but not limited to traumatic brain injury (TBI), chemotherapy-induced peripheral neuropathy (CIPN), amyotrophic lateral sclerosis (ALS), and ocular neuropathy. Here, we describe the discovery of novel, selective, orally bioavailable, brain-penetrant small molecule SARM1 inhibitors that function by intercepting NAD⁺ hydrolysis. These inhibitors potently block axon degeneration in primary neurons damaged by physical or chemical insults. SARM1 inhibitors also prevent axon degeneration and improve functional outcomes in preclinical models of neurological disease. Our results provide strong support for the broad therapeutic potential of SARM1 inhibition in the treatment of a range of neurological diseases.

Disclosures: **J. Hsu:** A. Employment/Salary (full or part-time): Nura Bio Inc. **X. Wang:** None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.09/Web Only

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: JP22ek0610023h0003 (Japan Agency for Medical and Research Development))
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JP23ek0610031h0001 (Japan Agency for Medical and Research Development)
JP24ek0610031h0002 (Japan Agency for Medical and Research Development)
JP25ek0610031h0003 (Japan Agency for Medical and Research Development)

Title: Neuroprotective therapy using SARM1 inhibitor compounds

Authors: ***T. ARAKI**¹, S. TOKUNAGA²;

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Abstract: Neurons are distinguished by their unique morphology, particularly their long axonal projections that facilitate intercellular communication. In many neurological disorders, axonal degeneration often precedes neuronal cell death. Increasing evidence suggests that axonal degeneration is an active, enzyme-regulated process, distinct from classical apoptosis and governed by its own molecular mechanisms. In neurodegenerative conditions, a decrease in the axonal NAD-synthesizing enzyme NMNAT2 and the activation of SARM1, which degrades

NAD, are believed to lead to energy depletion within the axon, triggering destabilization of axonal structures. Notably, gain-of-function mutations that enhance the NADase activity of SARM1 have been found at higher frequencies in patients with amyotrophic lateral sclerosis (ALS), highlighting the importance of SARM1 activity regulation in the pathogenesis of neurodegenerative diseases. We have focused on compounds related to nicotinamide, a substrate of the NAD biosynthetic pathway, we have identified nicotinamide derivatives that exhibit potent axon-protective effects in a Wallerian degeneration model using cultured neurons. These compounds also demonstrated significant protective effects when administered orally in mouse models. In this presentation, we will highlight the therapeutic efficacy and partially elucidated mechanism of action of these compounds, and explore the broader potential of targeting axonal NAD metabolism as a neuroprotective strategy for treating neurodegenerative diseases.

Disclosures: **T. Araki:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Meiji Seika Pharma, Jiksak Bioengineering. **S. Tokunaga:** A. Employment/Salary (full or part-time): Meiji Seika Pharma.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.10/E53

Topic: D.04. Brain Injury and Trauma

Support: NIH R01NS114397

Title: Triple pharmacological targeting of Wallerian degeneration pathways preserves sciatic and optic axons in ex vivo models of central and peripheral nerve injury

Authors: ***T. ASIMAKOPOULOS**¹, A. S. ALEXANDRIS¹, E. KOKKINO³, A. M. LABUZ⁴, T. FAZE², V. E. KOLIATSOS⁵;

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⁴Johns Hopkins Univ., Baltimore, MD, ; ⁵Neuropathology, Neurology, Psychiatry, Johns Hopkins Univ., Baltimore, MD

Abstract: Wallerian degeneration is a programmed process in which the distal axon segment degenerates after injury, driven by NAD⁺ depletion, MAPK activation, and collapse of axonal metabolism, all converging on SARM1 activation. Pharmacological strategies that delay or halt SARM1 activation may offer therapeutic potential in peripheral and central nervous system injuries. A triple combination therapy targeting key nodes in this cascade—a MAPK inhibitor (GNE-3511), a NAMPT inhibitor (FK866), and nicotinic acid riboside (NaR), a precursor of NAD⁺—has been effective in vitro in suppressing SARM1 activity and axonal degeneration. We investigated this drug combination in ex vivo sciatic (peripheral) and optic (central) nerves. Axonal degeneration was induced by axotomy in sciatic and optic nerves from young adult male

YFP-16 mice. Nerves were cultured with vehicle or the triple-drug combination, then fixed and cleared using the SHIELD protocol after 2, 4, or 7 days. Sciatic nerves were analyzed for intact axon percentage using YFP fluorescence and immunostaining for neurofilament light chain (NFL) and its degenerative form (dNFL). Optic nerves were assessed by total fluorescence intensity and presence of axonal spheroids. Triple-drug treatment reduced axonal degeneration in sciatic nerves at all time points, with intact axons at 2 days post-injury being 60% in treated nerves versus 23% in controls ($p = 1.6 \times 10^{-6}$), and similarly at 4 days (56% vs. 16%, $p = 0.0020$) and 7 days (36% vs. 0%, $p = 0.0013$). In optic nerves, treated samples showed a 35.6% higher fluorescence signal at 7 days (41.3 vs. 26.6, $p = 0.039$), and a modest but non-significant trend at 2 days. We are also performing targeted metabolomics to quantify NMN, NAD⁺, and cADPR in treated and control nerves. This ex vivo study demonstrates that a targeted triple-drug combination can robustly mitigate Wallerian degeneration and promote axonal preservation, supporting multi-target neuroprotective strategies and highlighting the ex vivo nerve culture model for preclinical therapeutic screening.

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Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

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Program #/Poster #: PSTR271.11/E54

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant HL093056

Title: Transient Cardiac Sympathetic Denervation in AngII Hypertension: Mechanisms of Axon Degeneration

Authors: *A. P. SEK¹, B. A. HABECKER²;

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Abstract: Hypertension is a major risk factor for cardiovascular diseases, including heart failure, and drives pathological cardiac remodeling. Despite its prevalence, the effects of hypertension on cardiac sympathetic innervation remain poorly understood. After 4 weeks of angiotensin II(AngII)-induced hypertension, mice begin to experience heart failure and regional loss of sympathetic nerves. To assess changes in sympathetic innervation during hypertension, we infused mice with AngII via osmotic minipump across multiple time points. We used echocardiography to measure cardiac function and quantified sympathetic nerves with immunohistochemistry for tyrosine hydroxylase (TH). We observed significant sympathetic denervation after 7-days of AngII-hypertension, which regenerated by 14-days and stabilized through 4 weeks. Notably, the 7-day AngII mice exhibited impaired cardiac function that was not

seen in the 14-day AngII mice. Treatment with marimastat, a matrix metalloproteinase (MMP) inhibitor, attenuated sympathetic denervation, suggesting a role for proteolytic pathways. The p75 neurotrophin receptor (p75NTR) promotes apoptosis and neurodegeneration through proteolytic cleavage of its intracellular domain—a process non-selectively inhibited by marimastat. Prior studies have implicated p75NTR in sympathetic axon degeneration and denervation of damaged cardiac tissue following a heart attack. We hypothesize that p75NTR contributes to hypertension-induced cardiac sympathetic denervation. To test this, we treated 7-day AngII mice with either LM11A-31, a small-molecule p75NTR inhibitor, or vehicle control, and quantified cardiac sympathetic innervation. Through this study, we will identify p75NTR as a potential therapeutic target to preserve sympathetic innervation and prevent cardiac dysfunction.

Disclosures: A.P. Sek: None. B.A. Habecker: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

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Title: Neuroprotective Mechanisms of Adenosine in Vincristine-Induced Peripheral Neuropathy.

Authors: *H. KIM¹, J. SHIN²;

¹Dong-A University, Col. of Med. / Dept. of Mol. Neuroscience, Peripheral Neuropathy, Busan, Korea, Republic of; ²Dept. of Mol. Neurosci., Dong-A Univ., Busan, Korea, Republic of

Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a common side effect of anticancer drugs. Though CIPN often leads to dose limitations, no effective treatments or biomarkers have been developed for its diagnosis and management. To develop a CIPN treatment, it is essential to study the mechanisms through which anticancer drugs induce axon degeneration and pain. Adenosine, a purine nucleoside, is involved in various physiological processes and has been studied for its therapeutic potential in treating central nervous system diseases. In the peripheral nervous system, adenosine has been shown to protect damaged axons, with effects lasting longer than those of supplementing NAD⁺, which regulates the activity of SARM1, the primary regulator of axon degeneration. However, the molecular mechanism of action of adenosine remains unclear. In this study, we determine the molecular mechanism of the peripheral neuropathy induced by vincristine, a well-known anticancer drug, and the neuroprotective effect of adenosine by modeling vincristine-induced peripheral neuropathy

(VIPN) using cultured dorsal root ganglia neurons. In addition, a VIPN mouse model was used to assess neuropathic pain and loss of intra-epidermal nerve fibers. We find that adenosine rescues the disruption of mitochondrial membrane potential and NAD⁺ reduction caused by vincristine treatment in vitro. We suggest that elucidating the neuroprotective mechanism of adenosine will help to pinpoint druggable targets for VIPN treatment.

Disclosures: H. Kim: None. J. Shin: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

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Program #/Poster #: PSTR271.13/E56

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R01 CA205255

Title: Chemotherapeutic injury induces local axonal translation in dorsal root ganglion sensory neurons

Authors: *J. ZUNIGA PAIZ¹, M. F. PAZYRA-MURPHY², R. A. SEGAL³;

¹Harvard Univ., Boston, MA; ²Dana Farber Cancer Inst., Boston, MA; ³Harvard Med. Sch. Grad. Program In Neurosci., Boston, MA

Abstract: Axon degeneration marks the common physiological event in most, if not all, neurodegenerative pathologies. Within the long axons of the peripheral nervous system (PNS), nerve injury rapidly and locally initiates cell-autonomous degenerative mechanisms, referred to as Wallerian degeneration, that are either succeeded by a commitment to cell death or reversal towards regeneration. PNS nerve crush injury models show the activation of protein translational regulation immediately near the site of injury are required for determining cell fate, specifically, via activation of the mTORC pathway. However, it is still not known how protein synthesis is locally regulated in axons after injury and if translational regulation is a key mechanistic step in the degenerative cascade. To answer these questions, we utilize chemotherapy-induced peripheral neuropathy (CIPN), a chronic neuronal injury model, which initiates a retrograde axonal degenerative cascade in dorsal root ganglion (DRG) neurons. To address how axonal translation is locally regulated following neuronal injury, we cultured primary DRG neurons in compartmentalized Xona microfluidic devices and investigated the effects of paclitaxel (120 nM) as an inducer of chronic injury using immunofluorescence imaging techniques. We treated cells with puromycin (2 μ M) and measured changes in the abundance and localization of nascent, puromycinylated peptides along the distal axon compartment of paclitaxel and control-treated neurons. We found a significant increase in the signal intensity and percent area of puromycinylated peptides within DRG axons with paclitaxel-induced degeneration, implying an upregulation in axonal translation (n=3 sets). Specifically, nascent peptides are more localized to branch points, growth cones, and areas of axonal swelling. Furthermore, we visualized changes

in abundance, organization, and localization of ribosomes in DRG axons following paclitaxel treatment. Using a ribosomal RNA signal intensity, we determined that the abundance and distribution of ribosomes in axons increased as measured with percent area and signal intensity (n=3 sets). Interestingly, ribosomes are similarly localized to regions of increased nascent proteins, such as branch points, growth cones, and swellings, linking axonal ribosome organization to changes in translational status. We are currently investigating the upstream mechanisms involved in regulating injury-induced translation. to identify if inhibition alleviates axonal degeneration in CIPN.

Disclosures: J. Zuniga Paiz: None. M.F. Pazyra-Murphy: None. R.A. Segal: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.14/E57

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01NS043314-19

Title: Using the ketogenic diet as a therapy for bortezomib-induced peripheral neuropathy

Authors: *L. L. HESLOP;

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Abstract: Bortezomib-induced peripheral neuropathy (BIPN) is a condition where peripheral nerves degenerate after exposure to the neurotoxic chemotherapy agent, Bortezomib (BTZ). BIPN affects up to 75% of cancer patients who receive it, causing symptoms of pain, numbness, and tingling in the distal limbs. Therapeutic options for BIPN are limited, leading us to explore new avenues focused on dietary interventions, specifically the ketogenic diet. To study the interplay between BTZ and the ketogenic diet, we conducted a ten-day study, administering intraperitoneal injections for five consecutive days of either vehicle or BTZ, followed by a five-day washout period in C57B/L6 male mice that received either a chow diet or a ketogenic diet throughout the entirety of the study (n = 8-12 per group). Measured outcomes included mechanical sensitivity, cold sensitivity, intraepidermal nerve fiber density (IENFD), dorsal root ganglion (DRG) neurite outgrowth, DRG extracellular acidification rate, mitochondrial oxygen consumption rate and mitochondrial reactive oxygen species production. Our *in vivo* results show that BTZ significantly reduced the IENFD in mice fed a chow diet, but mice given BTZ and fed a ketogenic diet were protected from IENFD reductions. Further results also showed that BTZ-treated mice developed cold sensitivity, but a ketogenic diet mitigated this behavior in BTZ-treated mice. These results suggest that elevations in ketones from a ketogenic diet can prevent epidermal fiber loss in BIPN and also play a role in alleviating painful symptoms of neuropathy. In addition, our *in vitro* results reveal differences in various metabolic parameters within neurons. The differences in cellular metabolism suggest that BTZ and a ketogenic diet

might lead to opposing metabolic programs within DRG neurons. Further *in vitro* studies using primary DRG culture will be completed to explore benefits of *in vitro* ketone administration and also confirm results seen *in vivo*.

Disclosures: L.L. Heslop: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.15/E58

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Animalfree Research Switzerland
Charité 3R — Replace — Reduce — Refine
Else-Kröner-Fresenius-Stiftung (2020_EKEA.80 to PH)

Title: Modulation of neuronal calcium sensor 1 protein interactions shows promising translational potential in the prevention of paclitaxel-induced neurotoxicity

Authors: *W. BOEHMERLE¹, C. SCHINKE¹, B. E. EHRLICH², S. MAIERHOF¹, L. NOLTE¹, M. ENDRES³, P. HUEHNCHEN⁴;
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Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a common adverse effect of the cytotoxic chemotherapy drug paclitaxel, causing sensory loss and neuropathic pain, substantially reducing patients' quality of life. The pathophysiology remains incompletely understood; previous research implicated neuronal calcium sensor 1 (NCS-1), which interacts with the inositol 1,4,5-trisphosphate receptor (InsP3R), disrupting calcium homeostasis. We hypothesized that modulating NCS-1 interactions would protect neurons from paclitaxel toxicity. Human induced pluripotent stem cell-derived sensory neurons (iPSC-DSN) were generated from two female, one male donor 26-48 yr and a matched NCS-1 knockout (CRISPR-Cas9) line. iPSC-DSN were matured ≥ 60 d and treated with paclitaxel (100 pM-10 μ M) which includes the clinically relevant dose range. Cellular viability, calcium imaging, patch-clamp electrophysiology, and multielectrode arrays assessed neuronal integrity and function. NCS-1 knockout significantly increased neuronal survival (IC₅₀: 1.1 μ M vs. 263 nM in wild-type) and attenuated electrophysiological impairment in paclitaxel treated cells. We identified lithium ions and ezetimibe as modulators of the NCS-1-InsP3R interaction. Co-incubation with both drugs improved neuronal viability dose-dependently, effects replicated by a synthetic blocking peptide, confirming interaction specificity. A retrospective analysis of anonymized German health insurance records showed a 35.3% relative risk reduction for CIPN diagnoses among ezetimibe users. Likewise, we will present data for the effects of a lithium co-medication in paclitaxel

treated patients from the PREPARE trial (DRKS00027165). Rigorous methodologies, including blinded experimental replications (minimum 2 biological replicates, 6 technical replicates per condition), control treatments, and appropriate statistical analyses, ensured robust conclusions. In summary, these bench-to-bedside findings highlight NCS-1 as a therapeutic target, suggesting lithium ions and ezetimibe as promising co-medications to mitigate paclitaxel-induced neuropathy, warranting further clinical investigation.

Disclosures: **W. Boehmerle:** None. **C. Schinke:** None. **B.E. Ehrlich:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); CSO Osmol Therapeutics. **S. Maierhof:** None. **L. Nolte:** None. **M. Endres:** None. **P. Huehnchen:** None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.16/F1

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: College of Medicine Summer Fellowship to JP
College of Medicine CoMRAD to AK
Natural Sciences and Engineering Research Council (NSERC) USRA to JP
Natural Sciences and Engineering Research Council (NSERC) Discovery Grant to AK

Title: Exploring the interactome of mesencephalic astrocyte-derived neurotrophic factor (manf) using bio-id method

Authors: ***J. PATEL**¹, **B. SIVAKUMAR**², **V. MARTINEZ**³, **A. KUMAR**³, **A. KRISHNAN**²;
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Abstract: Bio-ID is a well-established technique that utilizes a biotin ligase enzyme to biotinylate proteins that are in close proximity to a target protein, allowing the detection of transient protein-protein interactions via pulldown of biotinylated proteins. Mesencephalic Astrocyte-Derived Neurotrophic Factor (MANF) is a neurotrophic factor that has previously been studied extensively in the central nervous system. We identified the expression and growth-promoting properties of MANF in the peripheral nervous system (PNS). However, the receptor and the interactome of MANF for its growth-promoting properties have not been elucidated. Insights into this would further our understanding of the molecular operations of MANF. Therefore, we performed Bio-ID experiments to identify MANF's interactome and receptor. We developed a Dox-inducible lentivirus that encodes the biotin ligase BirA (L-Dox-BirA) as

control and a Dox-inducible lentivirus that encodes the fusion protein BirA-MANF (L-Dox-BirA-MANF). These viruses were transduced in rat Schwann cell (SC) line S16 and primary SCs. Stably transduced cells were selected with puromycin, and gene expression was induced by supplementation of Dox. Following this, we supplemented biotin to the stably transduced cells expressing BirA and MANF-BirA fusion protein to biotinylate proteins that are closely associated with MANF. These biotinylated proteins were then pulled down using streptavidin beads for LC-MS/MS analysis for the detection of interactome. Western Blot and qRT-PCR assays showed successful induction of BirA and BirA-MANF in SCs. Biotinylation assay showed that BirA in control SCs and BirA-MANF expressing SCs is functional. LC-MS/MS analysis of the pulled-down biotinylated proteins is ongoing, and these results will also be presented in the poster.

Disclosures: **J. Patel:** None. **B. Sivakumar:** None. **V. martinez:** None. **A. Kumar:** None. **A. Krishnan:** None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.17/F2

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Federal Ministry of Education and Research Germany 01KC2002

Title: Results of the preclinical multicenter randomized controlled paclitaxel-induced neuropathy prevention replication study (PINPRICS)

Authors: ***P. HUEHNCHEN**^{1,2}, **M. ENDRES**², **W. BOEHMERLE**²;

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Abstract: Objective: Chemotherapy-induced peripheral neuropathy (CIPN) is a frequent and serious side effect of many cytotoxic drugs, including paclitaxel. Despite the identification of treatment options in animal models, clinical trials for the treatment or prevention of CIPN have been negative. Major challenges for successful clinical translation of preclinical data include a lack of reproducibility and randomization, small sample sizes and insufficient statistical tests. We therefore conducted a confirmatory, preclinical multicenter randomized controlled replication trial to test the safety and efficacy of three drugs for preventing paclitaxel-induced polyneuropathy: (1) nilotinib, (2) lithium carbonate and (3) interleukin-6-neutralizing antibodies. We preregistered the data analysis plan as well as the two-step study protocol: the optimal doses of the three compounds were assessed first and then tested in a mouse breast cancer xenograft model to compare safety and efficacy.

Results: Unfortunately, toxicity of intraperitoneally administered nilotinib in combination with paclitaxel was observed, and higher-than-expected tumor growth resulted in a lack of power

when the trial was analyzed. Thus, although lithium carbonate and IL-6-neutralizing antibodies tended toward neuroprotection, the differences between these groups were not statistically significant. However, the PINPRICS study ultimately still provides important lessons with regard to the planning and conduction of multicenter preclinical trials.

Disclosures: P. Huehnchen: None. M. Endres: None. W. Boehmerle: None.

Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.18/F3

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 5R25GM060507

Title: Omega-3 fatty acid mitigate neuropathic pain by modulating ferroptosis, glutamate, and lipid remodeling

Authors: *V. DINH¹, A. DURAN², M. DESCORBETH⁶, F. ZAMORA³, J.-W. LIU⁴, J. D. FIGUEROA⁵, M. DE LEON⁷;

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Abstract: Neuropathic pain (NP) is a debilitating condition resulting from nerve injury and is a common co-morbidity of type 2 diabetes. Dietary omega-3 (OMG3) fatty acids have been shown to alleviate hyperalgesia and NP, likely by modulating key pathological mechanisms, including neuroinflammation, oxidative stress, mitochondrial dysfunction, and ion channel dysregulation. Understanding how OMG3 fatty acids regulate NP could help identify molecular targets for therapy. This study aimed to elucidate the primary cellular pathways by which dietary OMG3 fatty acids attenuate NP. We analyzed multi-omics data from both preclinical rodent models of chronic constriction injury (CCI) and human cohorts with painful diabetic neuropathy, all exposed to OMG3-rich diets. In the preclinical study, 40 rats were fed diets enriched with either fish oil (OMG3) or soy oil (omega-6) for four weeks prior to sciatic nerve CCI. Pain behaviors were assessed using the Hargreaves and CatWalk tests, and dorsal root ganglia tissue was harvested for RNA sequencing. Quantitative Real-Time PCR was used to confirm expression of promising RNA-seq transcripts. The clinical study enrolled 40 patients with type 2 diabetes and neuropathy who received OMG3 supplementation for three months. Plasma samples were analyzed via metabolomics and lipidomics. In rats, OMG3 significantly reduced hyperalgesia and allodynia and improved locomotion by day 7 post-CCI ($p < 0.05$). RNA-seq data revealed downregulation of pain-associated genes including **Scn10a** ($\log_2FC = -1.3$, $p = 0.006$), **Trpa1** ($\log_2FC = -1.4$, $p < 0.001$), and **P2rx7** ($\log_2FC = -0.73$, $p = 0.056$), alongside modulation

of the ferroptosis pathway gene **Gpx4** ($\log_2FC = 1.1$, $p = 0.01$). Markers of satellite glial cells, such as **Gfap** ($\log_2FC = 1.64$, $p = 0.0015$) and **Glul** ($\log_2FC = 0.90$, $p = 0.023$), were upregulated, suggesting enhanced neuronal repair and glutamate regulation. Genes involved in lipid metabolism, including **Fabp7** ($\log_2FC = 0.39$) and **Fasn** ($\log_2FC = -0.45$), also showed significant shifts. Activation of selenoamino acid metabolism in rats corresponded with metabolomics analysis results in human subjects, indicating enhanced antioxidant capacity and potential inhibition of ferroptosis. Lipidomics in the human cohort demonstrated increased triglyceride catabolism (DG→MG, $z = 2.03$, **PNPLA2**) and a reduction in peroxidation-prone lipids (DG[40:6] →PC[40:6], $z = 6.36$, **CHPT1**). Together, these findings indicate that OMG3 fatty acids reduce NP through modulation of ferroptosis, glutamate homeostasis, lipid peroxidation, and glial function. We highlight several candidate genes with therapeutic potential for NP that warrant further investigation.

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Poster

PSTR271: Cellular Stress and Degeneration: Peripheral Degeneration and Neuropathy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR271.19/F4

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 2P20GM103432
NIH Grant P20GM121310

Title: Targeting iRhom2 to Mitigate Neuroinflammation and Sensory Impairment in Diabetic Peripheral Neuropathy

Authors: V. MATTOS PEREIRA¹, I. WASSEEN², Z. ZHANG², Q.-Q. SUN², V. HOSUR⁴, *S. NAIR³;

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Abstract: Background and Aim: Diabetic peripheral neuropathy (DPN) affects 30-50% of individuals with diabetes and is a major cause of chronic pain and disability. Current therapies are largely symptomatic and fail to address the underlying disease mechanisms. Neuroinflammation triggered by metabolic dysregulation is a key to DPN pathogenesis, with iRhom2/ADAM17 signaling pathway central to inflammation. This study investigates the role of iRhom2 in the pathogenesis of DPN and its potential therapeutic applications. **Methods:** Wild-type (WT) and iRhom2 knockout (iRhom2^{-/-}) mice were rendered diabetic by intraperitoneal injection of streptozotocin (STZ, 150 mg/kg). Sensory function was assessed using mechanical (von Frey) and thermal (hot plate) sensitivity tests. In vitro studies employed human microglial (HMC3) and neuronal (SH-SY5Y) cell lines cultured under high glucose conditions.

Inflammatory and molecular signaling pathways were evaluated via cytokine profiling, siRNA-mediated gene silencing, oxidative stress assay, and immunocytochemistry. **Results:** Diabetic WT mice exhibited marked mechanical and thermal hyposensitivity, whereas iRhom2^{-/-} mice retained normal nociceptive responses despite sustained hyperglycemia, indicating a neuroprotective effect (Figure). Nerve tissue from diabetic mice exhibited elevated expression of ADAM17, RHBDF2 (which encodes iRhom2), and TNF α . In HMC3 cells, exposure to high glucose increased the secretion of IL-6, IL-8, TNF- α , and MCP-1, all of which were suppressed by iRhom2 siRNA. iRhom2 knockdown also reduced oxidative stress, and COX-2 was upregulated in microglia but not in neurons under high glucose conditions, highlighting cell-type-specific regulatory mechanisms. **Conclusion:** iRhom2 contributes to DPN pathogenesis by promoting microglial-driven neuroinflammation and oxidative stress. Genetic ablation of iRhom2 mitigates these responses and may contribute to preserving sensory functions. These findings suggest that iRhom2 is a promising therapeutic target for the treatment of diabetic neuropathic pain.

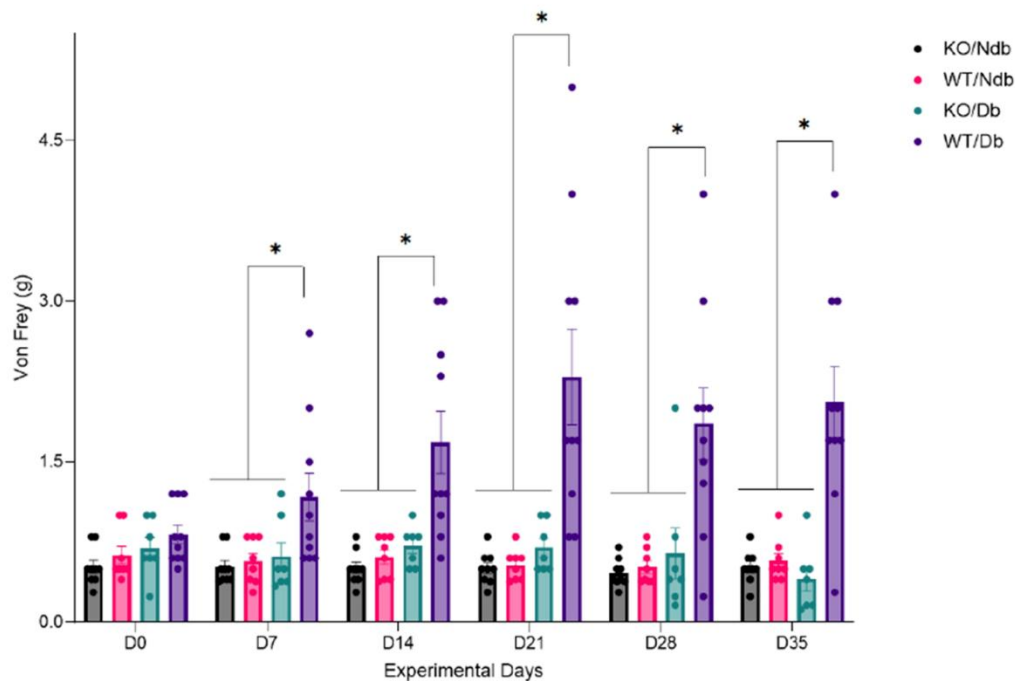


Figure. Von Frey test results for mechanical allodynia in the nondiabetic and diabetic wildtype and iRhom2 knockout mice. Data are expressed as mean $\pm$ SEM, $n = 7-10$ mice per group (* $p < 0.005$).

Disclosures: V. Mattos Pereira: None. I. Wasseen: None. Z. Zhang: None. Q. Sun: None. V. Hosur: None. S. Nair: None.

Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.01/F5

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH T32 Neuroimmunology
Pew Scholars Award
NIH R00 Pathway to Independence Award

Title: Uncovering the role of circumventricular organs during infection

Authors: *M. S. SWAROVSKI¹, J. OSTERHOUT²;
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Abstract: During an acute infection the body exhibits a set of highly stereotyped symptoms including fever, loss of appetite, fatigue, altered sleep and social interactions. A key open question concerns how the immune system communicates with the brain to generate these symptoms. We hypothesize that a major hub of communication between the periphery and the brain are circumventricular organs (CVOs). These structures are vascularized with fenestrated blood vessels that lack the usual tight junctions within the blood brain barrier and have been reported to express immune receptors. Here we aim to uncover the unique non-neuronal cell types and molecular mechanisms that facilitate the communication between the peripheral immune system and brain CVOs following different sources of systemic inflammation. We utilized single-nucleus RNA-sequencing of immune-activated CVO regions to molecularly characterize cell types and changes in immune gene expression during acute inflammation. We found that inflammation increased the expression of key immune genes in a subset of non-neuronal cell populations within several CVOs, compared to non-CVO brain regions, suggesting direct signaling mechanisms between the peripheral immune system and the brain. Moreover, we found that the gene expression changes were specific to the immune stimulus, suggesting the brain can decode unique immune programs. Together, our results reveal the immunological role of CVOs during infection and shed light on the cellular and molecular mechanisms of immune-brain communication in health and disease.

Disclosures: M.S. Swarovski: None. J. Osterhout: None.

Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.02/F6

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Glia Limitans Superficialis and Perivascularis Express Fibrinogen Chains

Authors: *E. V. GOLANOV¹, A. S. REGNIER-GOLANOV², M. CLAXTON¹, G. W. BRITZ³;
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Abstract: Cerebrospinal fluid (CSF) plays a critical role in the maintenance of normal brain function. While circulating along the subarachnoid space (SAS), the perivascular space, and through the brain parenchyma, CSF removes byproducts of brain activity. Mechanisms of CSF flow regulation are not completely understood. The SAS and the brain parenchyma are separated by glia limitans superficialis expressing myocilin (Hasel et al., 2025). Intraparenchymal paravascular space is separated from the brain parenchyma by glia limitans perivascularis. Earlier, we demonstrated that astroglia is capable of expressing fibrinogen (FBG), which is in line with earlier observations of extrahepatic constitutive expression of FBG and FBG gamma chain (FGG). Recently Stokum et al., 2021 reported constitutive expression of FGG in astrocytic endfeet forming glia limitans. We hypothesize that inflammation associated increase in FBG/FBG chains expression by glia limitans superficialis and perivascularis can hinder the CSF flow to minimize the spread of harmful elements, including pathogens, along the SAS and the perivascular space. In normal mice C57Bl/J we used immunohistochemistry (IHC) and fluorescent in situ hybridization to explore expression of fibrinogen chains by glia limitans superficialis and perivascularis identified by GFAP and myocilin expression. Intracisternal injection of kaolin has been used to trigger neuroinflammation. Fluorescent microspheres were used to trace movement of CSF along the brain ventral cisterns. In naïve mice, using IHC, we observed perivascular and brain surface FGG deposition associated with AQP-4, GFAP and myocilin suggesting a colocalization of FGG and glia limitans. Intra cisterna magna injection of kaolin, known to induce neuroinflammation, significantly increased by 43% ($P=0.038$) deposition of FBG over the ventral brain surface and thwarted CSF flow along the ventral brain surface. To ascertain the intrinsic origin of FGG in the astrocytic endfeet, we used RNAscope technology to detect FGG mRNA in-situ. Following kaolin injection, the number of FGG mRNA positive puncta increased at the ventral brain surface by 56% ($P=0.024$) compared to control. Additionally, IL-6 mRNA puncta increased by 63% ($P=0.122$) in parallel. Based on these data we conclude that astrocytic glia limitans superficialis and perivascularis are capable of FGG expression, which can be amplified by proinflammatory signals and hinder CSF flow.

Disclosures: E.V. Golanov: None. A.S. Regnier-Golanov: None. M. Claxton: None. G.W. Britz: None.

Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.03/F7

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 5R01AG078758-03

Title: Diurnal differences in immune cell trafficking through the central nervous system in aging

Authors: *A. HYNES^{1,2}, B. ROUTH^{3,2}, L. M. INCE², J. CHANEY^{3,2}, J. GOEWEY RUIZ², L. K. FONKEN²;

¹Univ. of Texas at Austin Dept. of Pharmacol. and Toxicology, Austin, TX; ²Div. of Pharmacol. and Toxicology, ³Interdisciplinary Neurosciences Program, Univ. of Texas at Austin, Austin, TX

Abstract: Immune function is coordinated by an endogenous ~24h circadian clock. The circadian clock regulates daily rhythms in peripheral immune cell migration through the blood and into tissues. This rhythmic regulation of the peripheral immune system is disrupted in inflammatory disorders, such as irritable bowel disease, asthma, and arthritis.

Neuroinflammation is a critical feature of age-associated neurodegenerative disorders, and aging results in disruptions to circadian/sleep-wake cycles. Therefore, we hypothesize disruption of immune cell migration rhythms through the brain and meninges as a potential mediator of neuropathology in aging. To test this, immune cell subpopulations were assessed in the meninges and brains of male and female adult (6 mos) and aged (18 mos) C57BL/6J mice at 4 times of day using flow cytometry. Adult female mice displayed diurnal differences in T cell migration through the meninges, with peak T cell counts occurring at the onset of the dark phase. Aging was associated with elevated T cell populations, primarily driven by an increase in CD8⁺ T cell populations in the brain and meninges during the light phase. Next, we evaluated whether altered rhythms in migratory factor gene expression in the meninges could be driving changes in immune cell migration with aging using 3' Tag sequencing. Our results demonstrate that migratory factor (*Vcam1*) expression in the meninges of aged mice was primarily elevated in the light phase. Taken together, these results show a time-of-day dependent effect of aging on T cell migration through the brain. Further analysis of T cell migration using immunohistochemistry at the onset of the light and dark phase will determine if impaired drainage by meningeal lymphatic vessels contributes to diurnal changes in T cell migration.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.04/F8

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIGMS Grant R35GM142368
CDMRP PD240020

Title: Traumatic brain injury induces T cell mediated neuroinflammation contributing to dopaminergic neurodegeneration and Lewy body pathology

Authors: *C. KELLY, J. P. MILNER, B. A. LESTER, S. BRINDLEY, L. HERNANDEZ, L. B. MURDAUGH, X. WEI, M. L. OLSEN, M. W. BUCZYNSKI, A. M. PICKRELL;
Virginia Tech., Blacksburg, VA

Abstract: Traumatic brain injury (TBI) increases the risk of Parkinson's disease (PD) development later in life but much remains unknown regarding the mechanisms driving this relationship. A single, mild brain injury may trigger activation of both resident and peripheral neuroinflammatory signaling pathways that are similarly activated in PD patients to increase susceptibility to neurodegeneration. Previous studies have demonstrated that specific subtypes of T cells mediate inflammation in preclinical models of PD in response to the neurotoxic accumulation of alpha synuclein. Certain T cell populations are also known to be activated and recruited to the brain parenchyma at subacute timepoints post-brain injury, and can persist chronically, negatively impacting TBI outcome. Here, we use mouse models (n = 6-8 male mice for each experimental group in each study) of both TBI and PD to evaluate how a pre-existing neuroinflammatory event may exacerbate PD-associated pathologies. Our transcriptomic analysis of mRNA from purified dopaminergic neurons of mice 90 days post mild TBI (mTBI) revealed upregulation of genes related to neuroinflammation, peripheral immune signaling, and IFN- γ , in addition to dysregulation of genes known to play a role in PD. Additionally, quantification of TH⁺ DA neurons in the substantia nigra shows significant ~30% cell death at 90 days post-injury compared to sham controls, with associated alterations in striatal neurotransmitter levels, like leading to behavioral phenotypes. At that same time point, CD8⁺ T cells are present throughout the brain and around the substantia nigra. When mTBI is induced 30 days prior to induction of PD-associated pathologies via intrastriatal injections of alpha synuclein preformed-fibrils, a similar susceptibility of DA neurons is observed, in addition to an increased severity in alpha synuclein propagation. In contrast, when adaptive immunodeficient Rag2 KO mice are exposed to the same experimental conditions, they display significant neuroprotection of the DA neuron population compared to wildtype animals. These findings indicate the possibility of a sustained peripheral immune cell infiltration and could support a complex, persistent, and detrimental crosstalk between both resident and peripheral immune cells which negatively affects DA neurons. Future directions of this study will determine whether T cell specific dependent mechanisms of action are directly responsible for the observed sensitivity of DA neurons after injury.

Disclosures: C. Kelly: None. J.P. Milner: None. B.A. Lester: None. S. Brindley: None. L. Hernandez: None. L.B. Murdaugh: None. X. Wei: None. M.L. Olsen: None. M.W. Buczynski: None. A.M. Pickrell: None.

Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.05/F9

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01MH112714
R01MH114858
1R01NS08916
1R21HD079249

The Nancy Lurie Marks Family Foundation
Landreth Foundation
Autism Speaks/National Alliance for Autism Research

Title: Cd8 cytotoxic t-cell infiltrates and cell damage in the hypothalamus in human obesity with causality testing in mouse

Authors: M. P. ANDERSON¹, J. T. AHRENDSEN², *Y. NONG³, Y. HUO⁴, J. STEELE⁵;
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Abstract: Rare cases of paraneoplastic obesity in children suggested sporadic obesity might arise from an adaptive immune cell-mediated mechanism. We quantified lymphocytic inflammation in hypothalamus, a central regulator of feeding behavior and energy expenditure, and other brain regions in a cohort of obese and non-obese human post-mortem brains. Here we report that CD3 and CD8-positive (cytotoxic) T-cells are increased selectively in hypothalamic median eminence/arcuate nucleus (ME/Arc) and bed nucleus of the stria terminalis (BNST) in 40% of obese (>30 BMI, body mass index) compared to non-obese patients. T-cells were not increased in other brain regions. T-cells were most abundant in individuals with concurrent obesity and diabetes. Markers of cytotoxic T-cell induced cellular damage, activated caspase 3 and poly-ADP ribose, were also elevated in the ME/Arc of obese patients. To test if the induction of T-cell infiltration into ME/Arc could trigger body weight gain in mice we performed stereotaxic injections of immunogenic green fluorescent protein (GFP) expressed by adeno-associated virus (AAV-CMV-GFP) or saline. AAV, but not saline, injections caused continuous weight gain until they were sacrificed 6 weeks after injection. CD8 T-cells were also increased in ME/Arc in mice with AAV-CMV-GFP injection. These results suggest T-cell infiltration of hypothalamus may be sufficient to cause obesity. Extrapolating the findings from these studies of the human postmortem brain and injected mice, the results suggest as much as ~40% of human obesity may be driven by dysregulated T-cells acting on feeding circuits of the hypothalamus.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.06/F10

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant NS072143
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Title: Adaptive Autoimmunity Drives Long-Term Nociceptive Sensitization in a Murine Model of Chronic Low Back Pain

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Abstract: The underlying cause of chronic low back pain (LBP) is often elusive, and it is believed that most LBP is nonspecific. Emerging evidence, including findings from our laboratory, suggests that LBP may involve autoimmune processes. Using a murine lumbar spinal disc puncture (DP) model, we previously demonstrated that pain behaviors depend on adaptive immunity, including the production of nociceptive IgM and IgG antibodies following injury. However, the temporal dynamics of nociceptive sensitization, immune activation, germinal center (GC) formation, the production of immunoglobulins (Ig) and Ig targeting of neural cell types have not been fully characterized. In this study, we employed nociceptive behavioral testing, antibody passive transfer experiments, western blotting, and immunohistochemistry to characterize the development of pronociceptive adaptive immunity after spinal disc injury. Our key findings include: (1) Wild-type (WT) DP mice exhibited mechanical allodynia, hyperalgesia, grip weakness, and conditioned place preference (CPP), while B cell-deficient muMT mice did not. (2) IgM levels increased in spinal cord and disc tissue from 1 to 33 weeks post-DP, peaking at 10 and 20 weeks, respectively. IgG levels rose more slowly, starting at 3-10 weeks and peaking at 20 weeks. (3) Passive transfer of purified IgM from WT DP mice at 3, 10, and 20 weeks—but not later—induced nociceptive behaviors in muMT DP recipients. (4) Passive transfer of IgG from WT DP mice at 10 and 20 weeks—but not earlier—induced nociceptive sensitization in muMT DP mice. (5) GC formation in lumbar lymph nodes began at 3 weeks and expanded markedly by 10-20 weeks, accompanied by increases in T follicular helper (Tfh) cells and follicular B cells. (6) Depletion of B cells with anti-CD20 attenuated nociceptive sensitization and improved grip strength. (7) Immunohistochemistry revealed increased IgG binding to satellite glial cells—but not neurons—in the dorsal root ganglia (DRG) of DP mice. Together, these data support the conclusion that lumbar disc injury induces a sustained adaptive autoimmune response. This involves GC formation and Tfh-driven B cell activation in lymph nodes, production of autoantibodies targeting DRG satellite glia, and persistent nociceptive sensitization over at least 24 weeks post-injury.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.07/F11

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Border-associated macrophages exhibit distinct signatures in response to microbial metabolites

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Abstract: The brain contains a diverse repertoire of innate immune cells, including border-associated macrophages (BAMs) and parenchymal microglia. While microglia have been extensively studied, the functional responses of BAMs under inflammatory conditions remain less well characterized. In this study, we explored how these two populations react to microbial-derived metabolites. By treating experimental autoimmune encephalomyelitis (EAE) mice with microbial-derived metabolites, we observed that BAMs exhibit distinct transcriptional signatures that are clearly separable from microglial responses. While microglia primarily responded with pathways related to metabolic remodeling, BAMs demonstrated unique expression patterns suggesting a divergent functional role in immune surveillance at the brain borders. Our findings highlight cell-type-specific immune adaptations in the central nervous system (CNS), underscoring the importance of BAMs as independent immune players during systemic immune activation. This work provides a framework for understanding how different brain-resident myeloid populations contribute to neuroinflammatory processes.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

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Program #/Poster #: PSTR272.08/F12

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: General Research Fund 17121622

Title: Investigating the role of interleukin-1 β in mediating macrophage Infiltration into the CNS after systemic challenge in a zebrafish model of neurodegenerative tauopathy

Authors: *T. YANG, S. S. YEUNG, R. CHANG;

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Abstract: Neuroinflammation is a key factor in neurodegenerative diseases, disrupting brain tissue homeostasis and affecting immune responses and cytokine production. Research using zebrafish models has shown that systemic inflammation triggers monocyte/macrophage invasion into the brain, leading to increased pro-inflammatory cytokines like interleukin-1 β (IL-1 β) and interactions with endothelial cells. This study investigates the role of IL-1 β in systemic macrophage infiltration and neurodegeneration using a macrophage-specific IL-1 β knockout zebrafish line generated via CRISPR-Cas9. To trigger neuroinflammation, we induced systemic

inflammation through i.p. LPS injection (non-sterile inflammation) and tail amputation (sterile inflammation). We utilized double transgenic lines Tg(mpeg1:egfp) and Tg(kdrl:mCherry) to visualize blood vessels and macrophages, performing 3D reconstruction with IMARIS to elucidate the chelation behavior of blood vessel-associated macrophages (BVMac). Our findings reveal that IL-1 β expressed specifically in macrophages is crucial for the neuroimmune response. In both the i.p. LPS injection and fin amputation models, macrophage-specific IL-1 β knockout downregulated microglial and astrocyte responses within the posterior tuberculum of the zebrafish brain. However, no significant changes in tau burden were observed in phosphorylation site Thr181 (AT270). Most importantly, changes in neuroinflammation were associated with impaired MPEG1+ macrophage infiltration into the CNS via interactions with KDRL+ endothelial cells of the blood vasculature. Interestingly, macrophage-specific IL-1 β knockout did not affect the populational BVMac in the fin amputation model. Instead, our results indicate that macrophage-specific IL-1 β knockout induced significant downregulation of macrophage interaction with LYVE1+ endothelial cells of the meningeal lymphatic vessels. Our results suggest significant differences in the infiltration behaviour of macrophages between sterile (fin amputation) and non-sterile (i.p. LPS) challenge, and that IL-1 β performs a significant role in modulating this preference. In conclusion, IL-1 β -dependent signaling is essential for systemic inflammation-induced macrophage invasion in the brain, highlighting its role in neuroinflammation and tauopathy in zebrafish.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

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KAKENHI 19K08025
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AMED 21wm04250XXs0101
AMED 21uk1024002h0002
AMED 22bm0804023h0003

Title: Macrophage Pathobiology in Individuals with ASD

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Med. Univ., Kashihara, Japan; ⁵Dept. of physiology, Dept. Physiol., Keio Univ. Sch. Med., Aichi, Japan; ⁶Keio Univ. Sch. of Med., Kanagawa, Japan

Abstract: Background: A growing body of evidence suggests that immune dysfunction and inflammation in the peripheral tissues as well as the central nervous system are associated with the neurodevelopmental deficits observed in autism spectrum disorder (ASD). Elevated expression of pro-inflammatory cytokines in the plasma, serum, and peripheral blood mononuclear cells of ASD has been reported. In addition, it has been discussed that microglia-dependent synapse phagocytosis is impaired in individuals with ASD compared to typically developed (TD) individuals. Methods: To elucidate the effect of immune cells on human neurons, we used two methods; a co-culture system of human-induced pluripotent stem cell-derived neurons and differentiated macrophages instead of microglia, and synaptosome phagocytosis measurement system, in which macrophages were also used. Results: Co-culture experiments showed that GM-CSF macrophages affect the dendritic outgrowth of neurons through the secretion of pro-inflammatory cytokines, interleukin-1 α and TNF- α . Macrophages derived from individuals with ASD exerted more severe effects than those derived from TD individuals. Synaptosome phagocytosis measurement revealed impaired synapse phagocytosis of M-CSF macrophages derived from individuals with ASD compared to those from TD individuals. Conclusions: Macrophages from individuals with ASD induced neuronal dendritic alterations and exhibited impaired synaptic phagocytosis functions more than those from TD individuals.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

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Title: Infiltrating monocytes contribute to an expanded population of MHCII⁺ perivascular retinal macrophages in a mouse model of glaucoma

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Abstract: Glaucoma, the leading cause of irreversible blindness worldwide, is characterized by progressive degeneration of retinal ganglion cells. While current therapeutic options for glaucoma focus on lowering the intraocular pressure (IOP), many patients continue to lose vision despite all IOP-lowering interventions, highlighting the need for better understanding of the underlying pathophysiology of this disease. Past studies have suggested significant immune cell involvement in glaucoma; however, their gene expression profiles, spatial distribution, and ontogeny remain poorly understood. To address these unknowns, we used a combination of single-cell RNA sequencing, immunohistochemistry (IHC), and lineage tracing in an experimental mouse model of glaucoma. We induced IOP elevation by injecting microbeads (MB) into the anterior chamber of 3-month-old C57BL/6J mice, while control mice received sham injections. 30 days after injection, we harvested the retinas and optic nerves, isolated CD45+ immune cells using flow cytometry, and performed sequencing using 10X Genomics, successfully identifying 28,277 immune cells. In glaucomatous retinas, we found a three-fold increase in the macrophage population compared to control retinas; interestingly, these cells expressed high levels of the antigen-presenting molecule MHCII. We confirmed the presence of this expanded MHCII+ macrophage population using IHC, and found that these cells were predominantly located in the vicinity of retinal blood vessels. We next sought to determine the ontogeny of perivascular MHCII+ macrophages in glaucoma, with one potential source being differentiation from peripheral monocytes. To test this hypothesis, we used the fate-mapping *Ms4a3^{Cre}-Rosa26^{TdTomato}* mouse line, which labels monocyte progenitors. After initial validation of the mouse line using flow cytometry, we induced experimental glaucoma in adult *Ms4a3^{Cre}-Rosa26^{TdTomato}* mice via MB injection, with sham-injected mice acting as controls. IHC revealed numerous perivascular Iba1+/MHCII+/RFP+ triple-positive cells in MB-injected retinas, while these cells were scarce in sham retinas. Interestingly, in retinas with many Iba1+/MHCII+ perivascular cells, approximately half were RFP+, suggesting that some, but not all, MHCII+ macrophages in disease originated from peripheral monocytes. Overall, our single-cell RNA sequencing and lineage tracing in a mouse model of glaucoma identify an expanded MHCII+ macrophage population that is in part derived from peripheral monocytes. Future studies will seek to determine the functional role of these immune cells in glaucoma pathogenesis.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

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Program #/Poster #: PSTR272.11/F15

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Neurodegenerative Disorder-associated Proteins Amyloid Beta, Alpha-Synuclein, Bacterial and Viral Toxins Activate Human Mast Cells and Release Inflammatory Mediators

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Abstract: Alzheimer's disease (AD) and Parkinson's disease (PD) are progressive neurodegenerative disorders. Lipopolysaccharide (LPS), Epstein-Barr virus (EBV) bacterial and viral components are neurotoxic and play a role in neurodegenerative disorders. Amyloid beta (A β) deposition contributes to the formation of amyloid plaques (APs) in AD. Alpha-synuclein (α -synuclein) is a neuronal protein, and the abnormal form of α -synuclein contributes to neuronal death in PD and dementia with Lewy bodies (DLB). Mast cells mediate allergic diseases and are also implicated in neuroinflammatory and neurodegenerative diseases, traumatic brain injury and stroke. Excessive activation of mast cells leads to the release of several multifunctional proinflammatory mediators, such as cytokines, chemokines and neurotoxic molecules that can exacerbate neuroinflammatory responses and activate glial cells to release additional inflammatory mediators. We studied the effect of A β 1-42, α -synuclein, LPS, and EBV on human mast cell activation-associated inflammatory mediator release. Human mast cells were grown by culturing human umbilical cord blood-derived CD34⁺ hematopoietic stem and progenitor cells with stem cell factor (SCF, 100 ng/ml) and interleukin-6 (IL-6, 50 ng/ml) in Iscove's Modified Dulbecco's Medium (IMDM) for 12-15 weeks in culture. These cells were then incubated with human A β (1-42) peptide, recombinant human α -synuclein, LPS, and recombinant EBV protein for 24 hours and inflammatory mediators' interleukin-8 (IL-8), IL-6 and, and matrix metalloproteinase-9 (MMP-9) were quantified in the culture supernatant media by using enzyme-linked immunosorbent assay (ELISA) kits. Results were analyzed by one-way analysis of variance (ANOVA) and Tukey's multiple comparison tests. Results show that A β , α -synuclein, LPS and EBV induced significantly high release of IL-8, IL-6 or MMP-9 as compared to the release from control human mast cells. In conclusion, A β , α -synuclein, bacterial and viral components such as LPS and EBV can activate human mast cells and release neuroinflammatory cytokines/chemokines and neurotoxic mediators that are implicated in neuroinflammatory and neurodegenerative disorders.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

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Program #/Poster #: PSTR272.12/F16

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: 1r21 ag087039

Title: Role of CNS LHCGR in the mediation of post-sepsis peripheral infiltration and neuroinflammation

Authors: ***I. ALONSO DIAZ**¹, **L. WAGNER**², **D. NACIONALES**³, **J. K. HARRISON**⁷, **P. CHAKRABARTY**⁴, **P. EFRON**⁵, **G. CASADESUS**⁶;

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Abstract: Sepsis-induced neuroinflammation, primary linked to peripheral infiltration, is associated with cognitive decline and dementia risk. The Luteinizing hormone receptor (LHCGR) has immune modulatory properties in the periphery, is expressed in the brain and the blood brain barrier, and ICV delivery of one of its ligands, hCG, improves CNS function, and regulates peripherally and centrally derived immune genes in AD mice. Given these findings, we sought to test its therapeutic potential in the post-sepsis brain by evaluating peripheral infiltration and neuroinflammatory changes in septic mice after central LHCGR activation. Specifically, ICV hCG, was delivered in a CX3CR1^{GFP/+}/CCR2^{RFP/+} double reporter mice undergoing cecum ligation and puncture (CLP) + stress, a translationally valid model of sepsis. We determined peripheral infiltration changes by quantifying RFP+ cells in the brain parenchyma and evaluated neuroinflammation by determining microglial activation using 3D IMARIS-based imaging and Sholl analysis and changes in neuroinflammatory markers. Our preliminary data suggests treatment dependent changes on these endpoints, supporting a functional immunomodulatory role of the LHCGR within the brain.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.13/G1

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: 1R21 AG087039

Title: Evaluation of peripheral immune infiltration and BBB integrity in the CLP + daily stress model of sepsis

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Abstract: Sepsis is associated with cognitive deficits and neurodegenerative disease development long term. Blood-brain-barrier (BBB) damage and peripheral immune cell infiltration into the CNS are hypothesized as mediators of these changes. To address whether such changes occur in a clinically & translationally relevant model of sepsis, cecum ligation and puncture (CLP) + restrain stress, we used a double reporter mouse line (CX3CR1^{GFP/+}/CCR2^{RFP/+}) and measured the levels of CCR2/RFP+ peripheral immune cells at 7-days, 14-days, 1-month, and 4-months post sepsis to address infiltration of peripheral immune cells. Imaging was and cell fate were further validated by flow cytometry. BBB integrity was directly evaluated via Evan's Blue injection and BBB tight-junction proteins changes at all timepoints. We observed increased peripheral immune cell presence at 7-days and 14-days post sepsis when compared to non-septic navies. This finding was further validated by changes in Evan's Blue penetrance into the CNS as well as tight junction protein alterations. Together, these data provide evidence of peripheral infiltration in this model. Given the established link between this mechanism, increased neuroinflammatory changes within the brain, and cognitive dysfunction, future studies need to directly evaluate whether peripheral infiltration in this model is a causal driver of neuroinflammatory changes that drive long-term cognitive loss.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

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Title: Early SERPINA3N signaling from hepatocytes potentiates brain dysfunction in sepsis-associated encephalopathy in mice

Authors: *J. PARK¹, M. ZHU¹, H. KIM¹, H. CHEN¹, Y. WANG¹, F. GUO²;
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Abstract: The mechanisms by which systemic inflammation breaches the brain parenchyma to trigger sepsis-associated encephalopathy (SAE) remain largely undefined. Serine protease inhibitor clade A member 3n (SERPINA3N), predominantly secreted by hepatocytes, plays a critical role in SAE pathogenesis in the context of the liver-to-brain axis. Here, we show that during the acute phase of peripheral lipopolysaccharide (LPS)-evoked systemic inflammation in mice, levels of SERPINA3N are markedly elevated in both the periphery and the brain, compromising blood-brain barrier (BBB) integrity. This breach drives glial activation towards a pro-inflammatory state and impairs neuronal function. Targeted deletion of hepatocytic *Serpina3n* using Cre-lox system abrogates early glial activation, suppresses the production of inflammatory cytokines and chemokines (IL-1 β , IL-6, TNF- α , CD86, CXCL1, CCL2, and Lcn2), and preserves BBB and neuronal integrity. Our findings identify hepatocyte-derived SERPINA3N as a key early driver of SAE and propose neuroinflammatory modulation as a promising therapeutic avenue.

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

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Program #/Poster #: PSTR272.15/G3

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Istanbul University Scientific Research Fund (Project ID:TOA-2021-37862)

Title: Peripheral Immune Trends Across Autoimmune Encephalitis Subtypes Identified by Single-Cell Protein Profiling

Authors: *Z. KARAASLAN¹, E. SANLI², S. UGUR ISERI³, E. TUZUN², V. YILMAZ²;
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Abstract: Motivation: Autoimmune encephalitis (AE) associated with antibodies against neuronal surface antigens such as N-methyl-D-aspartate receptor (NMDAR), Leucine-rich, glioma inactivated 1 (LGI1), and Contactin Associated Protein 2 (CASPR2) presents with overlapping yet distinct clinical phenotypes. Despite increasing interest in peripheral immune involvement in AE, the specific alterations in circulating immune cell subsets across AE subtypes remain poorly defined. This study aimed to characterize the peripheral immune landscape across AE subtypes using single-cell antibody-derived tag (ADT) profiling. **Methods:**

Peripheral blood mononuclear cells (PBMCs) were obtained from 12 human samples categorized into four groups: anti-NMDAR (n=4), anti-LGI1 (n=3), anti-CASPR2 (n=2), and healthy controls (n=3). Single-cell surface proteomic profiling was performed using the BD Rhapsody AbSeq platform, followed by computational analysis using Seurat and scCustomize packages in R. Cell types were manually annotated based on marker expression, and cluster-wise proportions were statistically compared using Kruskal-Wallis and Dunn post-hoc tests. **Results:** We identified 16 distinct immune cell types across all samples. Kruskal-Wallis tests revealed nominal differences in cell-type abundance across autoimmune encephalitis subtypes. The most notable trends were observed in cytotoxic natural killer (NK) cells ($p = 0.021$) and CD4⁺ central memory T cells (TCM) ($p = 0.04$). However, none of these differences remained statistically significant after Benjamini-Hochberg correction for multiple testing. Substantial inter-individual variability was observed, especially within control and anti-NMDAR groups. **Conclusion:** Although no immune cell subset differences reached statistical significance after correction for multiple testing, notable trends were observed in cytotoxic NK cells and TCM cells among autoimmune encephalitis subgroups. These trends suggest potential subtype-specific alterations in peripheral immune composition, particularly in anti-LGI1E and anti-CASPR2E patients. The lack of statistical significance may be attributable to the limited sample size ($n = 3$ per group on average), which reduces the power to detect subtle but biologically meaningful differences. Our findings underscore the importance of validating these preliminary observations in larger cohorts and support the utility of single-cell surface protein profiling for identifying disease-relevant immune shifts in autoimmune neurological syndromes.

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Poster

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R01 MH133070

Title: Modulation of fear behavior and neuroimmune alterations in a model of allergic asthma in female mice

Authors: *T. M. PEACH¹, R. SAH²;

¹Pharmacology, Physiology, and Neurobio., Univ. of Cincinnati, Cincinnati, OH; ²Pharmacol. & Systems Physiol., Univ. of Cincinnati, Cincinnati, OH

Abstract: Fear-associated conditions such as posttraumatic stress disorder (PTSD) are highly prevalent, yet contributory risk and vulnerability factors are not well understood. Accumulating evidence suggests that chronically elevated inflammatory load may be a potential risk factor for

PTSD. Allergic asthma is a common inflammatory condition known to cause airway inflammation (AI) and compromised mental health. Evidence supports a strong association of severe asthma with PTSD although underlying mechanisms are unknown. To address this knowledge gap in our lab developed a model of house-dust-mite (HDM) exposure effects on fear conditioning and extinction in male mice, reporting compromised fear extinction in mice with severe neutrophilic AI. As women have a higher prevalence of PTSD as well as immune/inflammatory conditions, it is now important to determine effects of AI in females. Accordingly, in the current study female mice were exposed to HDM +/- anti-C5aR1 antibody to elicit mild versus severe AI. Consistent with our male study, fear extinction-specific effects were observed in females suggesting a vulnerability of extinction-regulatory mechanisms to severe AI. However, contrary to males severe AI females exhibited active rearing behavior and low freezing during extinction suggesting divergent defensive coping. Preliminary mapping of regional delta FosB showed increased neuronal activation in subregions of the bed nucleus of stria terminalis (BNST) in severe AI mice, an effect absent in males. Our data indicates potential engagement of discrete fear circuits in severe AI effects on extinction. Current studies are exploring immune alterations in the lung (T cells) and brain (microglia) in females, hypothesizing increased lung neutrophilic response and microglial alterations within BBB-devoid circumventricular nodes. Collectively, these data indicate sex-specific behavioral effects of severe-airway inflammation suggesting potential divergence at the cell-circuit level. Our studies provide valuable insights on the association between lung inflammation and mental health pertinent to increased susceptibility observed in females

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Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.17/G5

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01 NS5103212
R01 NS122174.

Title: Mhc class i impacts broad recruitment of immune cells to dura mater meninges during acute theiler's virus infection

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Abstract: The dura mater of meninges hosts immune cells for CNS surveillance and response to infections, serving as a critical interface between the brain and peripheral immunity. The

contribution of MHC class I restricted antigen presentation is required for effective immunity against Theiler's murine encephalomyelitis virus (TMEV) infection. We therefore assessed the importance of MHC class I on diverse immune cell populations in the dura during acute TMEV infection. Using spectral flow cytometry, we profiled the immune response in the dura mater meninges in healthy and TMEV infected MHC class I^{-/-} (H-2Kb^{-/-}, H-2Db^{-/-}) mice and wild type controls. Loss of MHC class I expression resulted in altered myeloid and lymphoid cell populations in the dura during acute TMEV infection. Of particular interest was the increased numbers of PD1 expressing CD4 T cells, and increased PDL1⁺ plasmacytoid dendritic cells in the dura of MHC class I deficient mice post-acute TMEV infection. These findings demonstrate the importance of MHC class I and CD8 T cells in modulating dura immune cell populations and implies a shift toward CD4 T cell exhaustion in the absence of MHC class I. Subsequent studies employing in vivo cell specific ablation of MHC class I molecules in discrete cell types will further define the impact of antigen presentation on immune regulation and inflammatory outcomes within the dura mater.

Disclosures: Z. Tian: None. A. Hassani: None. M. Maynes: None. M. Pedra Seady: None. C. Lewis: None. J. Thelwell: None. F. Jin: None. M. Hansen: None. A.J. Johnson: None.

Poster

PSTR272: Neuroinflammation: Brain Borders and Peripheral Immune Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR272.19/Web Only

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Correlation between symptom severity and inflammatory biomarkers in patients with psychotic disorders at a hospital in Southeastern Mexico

Authors: *G. NOLASCO-ROSALES¹, D. RUIZ¹, O. CORTES¹, J. CRUZ-CASTILLO¹, G. V. JUÁREZ³, M. VILLAR-SOTO⁴, L. ROSALES-GARCÍA¹, Y. HERNANDEZ-DIAZ², A. GENIS-MENDOZA⁵, J. J. MARTÍNEZ-MAGAÑA⁶, I. E. JUAREZ-ROJOP¹;

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Abstract: Psychotic disorders are complex multifactorial psychiatric conditions characterized by delusions, hallucinations, mood disturbances, and behavioral abnormalities, often presenting with acute episodes throughout the course of the illness. Evidence suggests that immune system activation, particularly through microglial activation, promotes an increase in peripheral inflammatory biomarkers. These biomarkers might participate to symptom severity and could serve as indicators for disease monitoring. The aim of the present study was to correlate symptom severity and inflammatory biomarkers in patients diagnosed with psychotic disorders at the Regional High Specialty Mental Health Hospital at Villahermosa, Tabasco, Mexico. We

conducted a cross-sectional, descriptive, and prospective study. Twenty-six patients diagnosed with psychotic disorders according to DSM-5 criteria were included. Clinical evaluation consisted of The Positive and Negative Syndrome Scale (PANSS). Inflammatory biomarkers were measured (NLR, neutrophil-to-lymphocyte ratio; MLR, monocyte-to-lymphocyte ratio; PLR, platelet-to-lymphocyte ratio; and SII, systemic immune-inflammation index). Finally, PANSS and inflammatory biomarkers were correlated using software R version 4.4. We grouped the sample into affective psychosis (n=18), and non-affective psychosis (n= 18). Patients with affective psychosis had a higher age, showed greater symptom severity, were more frequently associated with SSRI (selective serotonin reuptake inhibitor) medication, and had higher SII values compared to those with non-affective psychosis. The severity of psychotic symptoms was negatively correlated with levels of inflammatory biomarkers (PLR, $r = -0.07$, $p = 0.02$; SII, $r = -0.16$, $p = 0.009$). Our findings suggest that inflammatory pathways might be involved in the pathophysiology of psychotic symptoms during periods of symptom control. Patients with affective psychosis showed greater clinical variability compared to those with non-affective psychosis. These inflammatory biomarkers might be considered a potential clinical tool for assessing disease control in Mexican patients with psychotic disorders.

Disclosures: G. Nolasco-Rosales: None. D. Ruiz: None. O. Cortes: None. J. Cruz-Castillo: None. G.V. Juárez: None. M. Villar-Soto: None. L. Rosales-García: None. Y. Hernandez-Diaz: None. A. Genis-Mendoza: None. J.J. Martínez-Magaña: None. I.E. Juarez-Rojop: None.

Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.01/G6

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Cns-penetrant lpa₁ antagonist pipe-791 provides superior analgesia compared to peripherally restricted ctx-343 in a rat model of lpa-induced osteoarthritis

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Abstract: Lysophosphatidic acid (LPA) signaling through the LPA₁ receptor contributes to the development of chronic joint pain in osteoarthritis (OA), yet the relative contributions of peripheral versus central LPA₁ signaling remain poorly understood. In this study, we evaluated the efficacy of PIPE-791, a CNS-penetrant LPA₁ antagonist, versus CTX-343, a peripherally restricted analog, in a weight-bearing rat model of osteoarthritis (OA) induced by intra-articular LPA injection. Rats were dosed with either PIPE-791 or CTX-343. Next, rats received unilateral intra-articular injections of LPA to induce joint inflammation and sustained pain behavior. Weight-bearing asymmetry was assessed with an incapacitance meter. PIPE-791 treatment significantly restored weight distribution to the affected limb out to 72h, while CTX-343 had a

more modest effect, supporting that blocking LPA1 centrally and peripherally provides superior efficacy than peripheral alone. In the periphery, satellite glial cells (SGC) in the dorsal root ganglion (DRG) express high levels of LPA1 and were confirmed in-house. During neuropathic pain, it has been proposed that aberrant LPA1 signaling leads to secretion of pronociceptive factors. To better understand this, we collected lumbar dorsal root ganglion (peripheral) and spinal cord (central) tissue from rats 24h after intra-articular LPA1 injection. In the DRG, we observed induction of glial fibrillary acidic protein transcript (*Gfap*) in lumbar DRGs, indicative of SGC activation. We also observed LPA-mediated increases in *Il6* and *Enpp2* (autotaxin). Importantly, PIPE-791 treatment prevented induction in each of these markers. In the lumbar spinal cord, we saw PIPE-791 sensitive upregulation of *Il6* and autotaxin. We also measured IL-6 protein in lumbar spinal cord tissue and observed an LPA-mediated increase in IL-6 protein that was reduced with PIPE-791. These findings demonstrate that CNS penetration is a key determinant of therapeutic efficacy for LPA₁ antagonism in OA pain. We hypothesize that an OA insult activates SGCs and the synthesis of pronociceptive signals (i.e., IL-6 and LPA). These, in turn, signal to upregulate production of IL-6 and LPA in the spinal cord and may help explain the superior efficacy with a CNS-penetrant antagonist. Of note, neuropathic pain can be ameliorated by CNS administration of either an anti-IL-6 antibody or an autotaxin inhibitor. As such, PIPE-791's superior analgesic profile highlights the importance of targeting central LPA₁ signaling and supports further development of CNS-penetrant LPA1 antagonists for OA and neuropathic pain.

Disclosures: A.R. Broadhead: None.

Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.02/G7

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH R01 NS135620-01
VISN22 MIRECC

Title: Pro-cognitive effects of blood IgM anti-NMDAR1 autoantibodies in a mouse model

Authors: *J. DEWIT¹, T. TEBYANIAN², A. UNAPANTA², M. N. VAUGHN³, V. B. RISBROUGH⁴, S. B. POWELL⁵, X. ZHOU⁶;
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Abstract: Background: Synaptic NMDAR signaling is required for cognition, however over-activation of extrasynaptic NMDARs can drive glutamate excitotoxicity in the brain. Blocking extrasynaptic receptors through IgM anti-NMDAR1 autoantibodies may protect against excitotoxicity. In humans, high levels of natural anti-NMDAR1 autoantibodies (mainly IgM

isotype) are associated with better cognition in Alzheimer's patients and healthy adults. Here we tested the hypothesis that mice producing only IgM anti-NMDAR1 autoantibodies will have improved cognitive function due to neuroprotective effects of the IgM autoantibodies.

Methods: Aicda mutant mice, which only produce IgM autoantibodies, were immunized with either control (n=20, 10/sex) or P2 NMDAR1 peptide (n=20, 10/sex) and tested in sensorimotor gating, learning and memory tasks.

Results: Levels of IgM anti-NMDAR1 autoantibodies were higher in immunized female mice compared to male mice. Female mice carrying IgM anti-NMDAR1 autoantibodies have a trend for increased sensorimotor gating [$F(1,18)=2.23$, $p=0.15$] and improved fear extinction memory [$F(1,16)=5.892$, $p=0.027$]. Both male and female mice have improved fear memory retention [$F(1,35)=5.5$, $p=0.025$]. Locomotor activity and approach behavior were unchanged.

Conclusions: IgM anti-NMDAR1 autoantibodies may enhance cognitive function by specifically inhibiting extrasynaptic NMDARs to enhance neuroprotection. Whether the auto-antibodies are more neuroprotective in females than males will be explored in future studies.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.03/G8

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: TTUHSC School of Medicine

Title: Goji Berry Supplementation Improves Brain Health by Modulating Neuroinflammation and Neurotransmission in the Frontal Cortex and Hippocampus in the LPS Rodent Model of Chronic Fatigue

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Abstract: Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS) is a complex disorder involving autonomic, immune, and cognitive dysfunction. It affects multiple systems with unclear mechanisms and limited treatments. Neuroinflammation is known to disrupt neurotransmission, contributing to cognitive and emotional deficits. In this study, we investigated region-specific molecular adaptations in the left frontal cortex (FC (L)) and hippocampus (HPC) following sustained lipopolysaccharide (LPS via i.p.) exposure and assessed the neuroprotective effects of dietary Goji berry supplementation. Twenty adult male Sprague Dawley rats were assigned to the control group receiving $1 \times$ PBS, i.p., twice/week (CON group),

LPS at 250 µg/kg, i.p., twice/week (LPS group), or LPS + 1% Goji diet (GOJI group) for six weeks. Gene expression levels in FC (L) and HPC were analyzed by qRT-PCR and normalizing by β -actin, indicating microglia (*Iba1*) and astrocyte (*Gfap*) activation, inflammatory signaling (*Tnf- α* , *Il-1 β* , *Tlr2*, *Tlr4*), mitochondrial biogenesis (*Pgc1 α*), fission (*Drp1*) and fusion (*Mfn1*, *Opa1*), antioxidative stress regulators (*Nrf2*, *Sod1*), and neurotransmission pathways (*Vglut1*, *Vglut2*, *Gad67*, *Gat1*, *Gaba γ 2*, *Gababr1*). All the statistical analysis was done using one-way ANOVA followed by *post hoc* uncorrected Fisher's LSD multi-comparison. Goji attenuated LPS-induced expression of *Iba1* and *Gfap* in FC (L) and *Gfap* in HPC. Goji also decreased LPS-induced upregulation of *Tnf- α* , *Il-1 β* , *Tlr2*, and *Tlr4* genes in FC (L) and *Il-1 β* and *Tlr2* in HPC. Goji blocked the LPS-induced downregulation of *Pgc1 α* in both regions. Goji increased the expression of *Drp1* and *Mfn1* genes in HPC, but not FC (L). Goji blocked LPS-induced downregulation of *Opa1* in FC (L) and upregulated antioxidant genes *Nrf2* and *Sod1* in both regions. Goji blocked the LPS-induced decrease of *Vglut1* expression in FC (L) but not HPC. Goji blocked the LPS-induced downregulation of *Gat1* in FC(L) and of *Gaba γ 2* and *Gababr1* in HPC. These findings indicate that Goji berry supplementation is able to block several molecular markers of LPS-induced neuroinflammation and neurotransmission changes in the FC (L) and HPC. Some GOJI's effects were region specific.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.04/G9

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: MIUR Progetto Eccellenza (2023-2027)
Post-Finasteride Syndrome Foundation

Title: Exploring allopregnanolone as a possible therapeutic strategy for post-finasteride syndrome: preclinical evidence of gut and brain protection

Authors: *S. DIVICCARO¹, R. OLEARI¹, F. AMORUSO¹, F. FONTANA¹, L. CIOFFI¹, G. CHROSTEK¹, V. ABENANTE², J. TROISI², A. CARIBONI¹, S. GIATTI¹, R. C. MELCANGI¹;
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Abstract: Post-finasteride syndrome (PFS) is a complex condition that can emerge following discontinuation of finasteride, a 5 α -reductase inhibitor commonly prescribed for androgenetic alopecia. PFS is characterized by persistent neuropsychiatric, sexual, and gastrointestinal dysfunctions. Increasing clinical and preclinical evidence suggests that PFS involves sustained dysregulation of sex steroid homeostasis, alterations in the gut microbiota, and neuroinflammation, indicating dysfunction of the gut-brain axis as a central potential

pathophysiological mechanism. A defining feature of PFS patient plasma is a marked reduction in allopregnanolone (ALLO), a neuroactive steroid that modulates GABAergic signaling and exerts neuroprotective, anti-inflammatory, and homeostatic effects in both the central nervous system and the gastrointestinal tract. In line with this, a preclinical model of finasteride withdrawal (FW) shows persistent gut ALLO depletion, accompanied by intestinal inflammation and gut dysbiosis. In this study, adult male *Sprague Dawley* rats were treated with finasteride for 3 weeks and then monitored for one month of drug suspension to evaluate the consequences of FW and the therapeutic potential of ALLO (administered subcutaneously every other day for a total of 8 injections, during FW). Results obtained suggest that animals exhibited a significant increase in pro-inflammatory M1 intestinal macrophages, disrupted profiles of microbial-derived metabolites, hypothalamic neuroinflammation, and altered expression of proteins associated with blood-brain barrier (BBB) integrity (i.e., Claudin-5, ZO-1). Systemic ALLO administration mitigated these alterations, reducing inflammatory markers in both the colon and hypothalamus. Additionally, ALLO partially restored intestinal mucosal architecture and improved BBB integrity markers and restored some microbial-derived metabolites, such as acetate and butyrate. These findings provide new preclinical evidence for the protective effects of ALLO following FW and identify intestinal and hypothalamic inflammation and microbial metabolites as key targets in the pathophysiology of PFS. This study suggests that ALLO has possible therapeutic potential for PFS and contributes to further understanding of the protective molecular mechanisms underlying this important neurosteroid.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

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Program #/Poster #: PSTR273.05/Web Only

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: PEW Innovative Grant
ANID-Subdirección de Capital Humano/Doctorado Nacional/2024 N°
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Postdoctorado ANID N°3230704 (OCM)
ANID-MILENIO NCN2023_32

Title: Physical exercise modulates neuroinflammatory markers gene expression in a prenatal alcohol exposure animal model

Authors: *B. GARCÍA-ORDENES^{1,2,3}, M. OLIVARES COSTA³, G. E. TORRES⁴, P. A. HAEGER^{2,3};

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Abstract: Alcohol consumption during pregnancy has devastating consequences on fetal development, resulting in Fetal Alcohol Spectrum Disorder (FASD). Among the range of FASD features are cognitive impairments and reduced muscle strength. At the brain level, ethanol exposure induces neuroinflammatory processes and redox homeostasis alterations. Previous studies indicate that physical exercise modulates these mechanisms in other cognitive impairment conditions. We have also recently shown that exercise significantly improves strength in FASD animal models. Here, we investigated whether physical exercise improves spatial memory acquisition in animals prenatally exposed to ethanol (PAE), through modulation of neuroinflammatory and redox-related processes in the hippocampus. Adolescent male Sprague-Dawley rats prenatally exposed to ethanol or not were assigned to four groups (n=4 per group) and underwent either resistance training or no intervention for six weeks. At the end of the training period, spatial memory performance was evaluated using the Morris Water Maze (MWM). Hippocampal mRNAs expression of neuroinflammatory markers (e.g. IL-1 β , TNF- α) and redox markers (e.g., SOD1) was measured using RT-qPCR. This study was approved by the ethical committee (No. 39/2022). In the PAE animals, physical exercise did not affect spatial memory but significantly reduced IL-1 β expression in the hippocampus (p=0.0233, T-test), while TNF- α and redox markers remained unchanged, with respect to control animals. These findings suggest that physical exercise may positively modulate neuroinflammatory markers in rats prenatally exposed to alcohol. Ongoing studies are focused on exploring potential compensatory effects by analyzing earlier stages of development. These results support further investigation of non-pharmacological interventions, such as exercise, in alcohol-related developmental disorders.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.06/G10

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: National Research Council of Argentina Grant (PIP 2022–2024 N° 334) PICT 2021 No.348 from Argentine Ministry of Science and Technology and the University of Buenos Aires (Ubacyt: 20020100100089BA)

Title: Neuroprotective and Anti-inflammatory Effects of Genistein in a Rat Model of Metabolic Syndrome-associated Encephalopathy

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Abstract: Metabolic syndrome (MS) is defined by the presence of at least three metabolic risk factors: dyslipidemia, hyperglycemia, insulin resistance, hypertension, and central obesity. The spontaneously hypertensive rat (SHR) is a validated model for studying human MS, developing key features of the syndrome when exposed to a high-fat, high-carbohydrate diet (HFD). Previous studies from our group have shown increased reactivity of GFAP⁺ astrocytes and IBA1⁺ microglia in various brain regions, reduced hippocampal neurogenesis and impaired cognitive performance in rats with MS (SHR+HFD).

Genistein (GEN), a phytoestrogen found in soy, has demonstrated neuroprotective capabilities in several neurodegenerative diseases. It binds to multiple estrogen receptors (ER α , ER β and GPER) without inducing the carcinogenic or feminizing effects typically associated with estrogen treatment. In MS animals, we previously observed a predominance of activated or hypertrophic microglial phenotypes in the hippocampus, which was reversed following GEN administration (10 mg/kg, subcutaneously, for 2 weeks).

In this study, we analyzed the secretory profile of IBA1⁺ microglia via fluorescent immunohistochemistry, assessing colocalization with pro-inflammatory (TNF α) and anti-inflammatory (Arg-1) markers in the CA1 and dentate gyrus (DG) regions of the hippocampus. MS rats showed a higher proportion of TNF α ⁺ microglia and a lower proportion of Arg-1⁺ microglia compared to normotensive Wistar Kyoto (WKY) controls. These alterations were significantly reversed following GEN treatment.

We further characterized the phenotypic distribution of IBA1⁺ microglia in the prefrontal, perirhinal and entorhinal cortices. MS rats exhibited a significant shift toward an activated microglial phenotype across these regions, which was notably reversed by GEN treatment. We also extended cognitive assessment using the discontinuous Y-maze (d-YMAZE) and analyzed hippocampal expression of cytokines and transcription factors by RT-PCR.

Our results support a neuroprotective and anti-inflammatory role for GEN in MS-associated brain dysfunction, highlighting its potential as a therapeutic strategy for MS-related cognitive impairment.

Disclosures: S.R. Ronchetti: None. A. De Nicola: None. L. Pietranera: None.

Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

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Program #/Poster #: PSTR273.07/G11

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: FAPESP 2021/12906-0
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Title: Ouabain modulates BDNF expression in lung and brain regions in a murine model of mixed asthma

Authors: *M. R. RIBEIRO¹, A. SIENA¹, J. A. LEITE², G. DOS SANTOS¹, T. A. AMORIM¹, L. LIMA¹, D. Z. VIANA¹, A. ORELLANA¹, M. OLIVEIRA¹, E. M. KAWAMOTO¹, W. TAVARES-DE-LIMA¹, C. SCAVONE¹;

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Abstract: Brain-derived neurotrophic factor (BDNF), a protein highly expressed in neurons and glial cells, plays key roles in neural development, survival and plasticity. Previous studies have shown that BDNF is involved in the pathophysiology of eosinophilic asthma (Watanabe et al., AJRCMB 2015; 53:844); however, its role in mixed asthma remains unknown. Asthma is a chronic inflammatory lung disease characterized by epithelial damage, immune cell infiltration, with no known cure. In the mixed asthma phenotype, both eosinophils and neutrophils contribute to the inflammatory response, making the disease more complex and challenging to treat. Ouabain (OUA), a potent Na⁺,K⁺-ATPase inhibitor, has been investigated for its neuromodulatory and immunomodulatory properties. It has been shown to attenuate allergic airway inflammation by reducing granulocyte infiltration into the bronchoalveolar lavage fluid (Galvão et al., 2022). Methods: Female Balb/c mice were sensitized with ovalbumin (OVA) and Complete Freund's Adjuvant (subcutaneous, Day 0). OVA challenges (intranasal) were administered on days 21-23. The OVA+OUA group received ouabain (0.56 mg/kg, i.p.) one hour before each challenge. Experiments were conducted on day 24. Control mice received PBS or OUA via the same route. Lung inflammation was assessed microscopically by quantifying cells recovered from bronchoalveolar lavage (BAL). BDNF levels were measured using an ELISA kit. Data are expressed as mean±SEM and analyzed using the Student's t-test or one-way ANOVA (GraphPad10). Results: BAL fluid from OVA-challenged mice showed a significant increase in total cells (n=5) ($\times 10^4/\text{mL}$: 116 ± 12 $P < 0.0001$), neutrophils (54 ± 4 , $P < 0.0001$), and eosinophils (59 ± 8 , $P < 0.0001$). Total lung cell recruitment was also increased. In lung tissue, BDNF levels (pg/mL) were elevated in groups treated with ouabain: OUA vs PBS (312 ± 203 , $P < 0.0003$) and OVA vs OVA+OUA (274.8 ± 617.6 , $P < 0.0001$) (N=5-7). In the frontal cortex, OUA treatment decreased BDNF levels in the OVA+OUA group compared to PBS (313 ± 575 , $P < 0.02$) (N=4-6). No significant differences in BDNF levels were observed in the hippocampus. Conclusion: This murine model of mixed asthma reflects the main pathological features described in the literature. OUA treatment significantly increased BDNF levels in the lungs of asthmatic and non-asthmatic animals. Our preliminary results indicate that OUA treatment in asthmatic animals reduced BDNF expression in the frontal cortex, with no changes observed in the hippocampus. Further studies must explore the difference between peripheral and central effects of OUA in the nervous system in the presence of an inflammatory disease.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.08/Web Only

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Departamento de Ciencias de la Salud
COMECYT

Title: Effect of environmental novelty on synaptic plasticity and neuroinflammation in the dorsal hippocampus of a model of diet-induced cognitive decline

Authors: A. HERNÁNDEZ-HERNÁNDEZ¹, Z. K. PÉREZ-ALANIS¹, S. HERNÁNDEZ-RAMÍREZ¹, *K. R. GUZMAN-RAMOS²;

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Abstract: Obesity, recognized as a global epidemic by the WHO, is associated not only with metabolic and cardiovascular diseases, but also with neuroinflammation and cognitive decline. This study evaluated the effect of constant environmental novelty consisting of exposure to sensory, physical, social and cognitive stimulus, on the levels of proteins related to neuroinflammation and synaptic plasticity in the dorsal hippocampus of mice fed a high-fat diet. Tyrosine Hydroxylase (TH) protein as a modulator of dopaminergic neurotransmission; Glial Fibrillary Acidic Protein (GFAP) as an indicator of neuroinflammation; Postsynaptic Density Protein 95 (PSD95) and N-methyl-D-Aspartate receptor subunit 1 (NMDAR1) as markers associated with synaptic plasticity. The results showed a significant protective effect of constant novelty on spatial learning of mice exposed to high-fat diet, compared to mice in standard housing conditions, which present clear memory impairments. Additionally, cognitive protection correlated with catecholaminergic terminals preservation in the hippocampus, as TH maintained the same levels in stimulated animals, comparable to control animals that consumed regular diet. Environmental novelty also decreased neuroinflammation and maintained the NMDAR1 levels. Conversely, PSD95 levels were significantly higher in obese mice with environmental novelty, which suggests that this compensatory molecular change was promoted by cognitive stimulation, but was not sufficient to prevent cognitive impairment. These findings suggest that environmental enrichment could exert a neuroprotective effect, attenuating the negative effects of obesity on the brain and contributing to the preservation of cognitive function.

Disclosures: A. Hernández-Hernández: None. Z.K. Pérez-Alanis: None. S. Hernández-Ramírez: None. K.R. Guzman-Ramos: None.

Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR273.09/G12

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

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R01 AG081599 (NIA)
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U54AG054349 (NIA Model Organism Development and Evaluation for Late-onset Alzheimer's Disease [MODEL-AD])
T32NS082174 (NINDS)
T32NS121727 (NINDS)

Title: Myeloid-derived β -hexosaminidase is essential for neuronal health and lysosome function: implications for Sandhoff disease

Authors: *K. I. TSOURMAS¹, C. A. BUTLER³, N. E. KWANG³, V. SWARUP³, M. M. ACHARYA², L. A. HOHSFIELD³, K. N. GREEN³;

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Abstract: Lysosomal storage disorders (LSDs) are a large disease class involving lysosomal dysfunction, often resulting in neurodegeneration. Sandhoff disease (SD) is an LSD caused by a deficiency in the β subunit of the β -hexosaminidase enzyme (Hexb). Although Hexb expression in the brain is specific to microglia, SD primarily affects neurons. To understand how a microglial gene is involved in maintaining neuronal homeostasis, we demonstrated that β -hexosaminidase is secreted by microglia and integrated into the neuronal lysosomal compartment. To assess therapeutic relevance, we treated SD mice with bone marrow transplant and colony stimulating factor 1 receptor inhibition, which broadly replaced Hexb^{-/-} microglia with Hexb-sufficient cells. This intervention reversed apoptotic gene signatures, improved behavior, restored enzymatic activity and Hexb expression, ameliorated substrate accumulation, and normalized neuronal lysosomal phenotypes. These results underscore the critical role of myeloid-derived β -hexosaminidase in neuronal lysosomal function and establish microglial replacement as a potential LSD therapy.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

Location: SDCC Halls B-H

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Program #/Poster #: PSTR273.10/G13

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 1P01AI073693
T32 Grant Support

Title: Anti-nmdar autoantibody-mediated neuronal damage in the amygdala mediates anxiety disorders in a model of neuropsychiatric lupus

Authors: *R. WEISSMAN-TSUKAMOTO^{1,2}, B. T. VOLPE³, K. CARROLL³, Z. IBIC³, C. KOWAL³, B. DIAMOND³;

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Abstract: Background: Up to 80% of individuals with SLE experience cognitive, emotional, or behavioral symptoms, termed Neuropsychiatric Lupus (NPSLE). NPSLE cognitive impairment and mood disorders may be mediated by autoantibodies, with specific symptoms related to the varied targets of these autoantibodies and the region of blood brain barrier (BBB) injury. We use a murine model of NPSLE induced by anti-double-stranded DNA (dsDNA) antibodies (DNRAbs) which cross-react with the excitatory neuronal N-methyl D-aspartate receptor (NMDAR) and facilitate excitotoxic neuronal death. These antibodies are present in approximately 30% of patients with SLE and at a higher rate in those with NPSLE. Prior research in our lab has shown that while LPS administration allows DNRAbs to cause hippocampal damage and chronic cognitive impairment, epinephrine administration to DNRAb+ mice leads to acute neuronal excitotoxicity in the amygdala. We hypothesized that there are also chronic neuronal abnormalities in the amygdala. **Methods :** Female B6H2d mice are immunized with DWEYS peptide on a polylysine backbone to produce DNRAbs or with the polylysine backbone as a control. PET studies with ¹¹C-AIB tracer are used for BBB permeability. Unbiased and blinded stereology and immunohistochemistry are used to identify cell type morphology and quantify neuronal loss. Behavioral testing includes object place memory, elevated plus maze, and fear conditioning. **Results and Conclusions:** In this study, we characterize the chronic effects of DNRAb antibody penetration into the brain after systemic administration of epinephrine. We show that BBB permeability due to epinephrine is selective for the amygdala, resulting in antibody penetration into the amygdala. We observe significant neuronal loss in the basolateral, lateral, and medial posteroventral nuclei of the amygdala in DNRAb+ compared to DNRAb- mice, correlating with amygdala-related behavioral findings in anxiety testing. Finally, we observe an abnormal morphology in surviving neurons and lipid oxidation of axons suggestive of axonal degeneration. Interestingly, the DNRAb-mediated neuronal pathology in the amygdala, which we currently hypothesize is due to ferroptosis, differs from what we previously reported in the hippocampus, which is characterized by chronic microglial-dependent synaptic elimination and loss of dendritic arborization. Further investigation will focus on circuit disruption and molecular mechanisms of chronic neuronal damage. This work expands our understanding of the neuropathological mechanisms which can underly psychiatric symptoms in NPSLE.

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Poster

PSTR273: Neuroinflammation, Neuroprotection, and Cellular Stress: Animal Models

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 1R21NS135088 (IK)

Title: Cerebellum and plasma metabolomics in sepsis reveal metabolic disturbances and potential diagnostic biomarkers for septic encephalopathy

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Abstract: Septic encephalopathy (SE) is a severe complication of sepsis, marked by metabolic shifts, with the cerebellum being among the most affected brain regions. Sepsis-induced energy deficits play a causal role in activating neuroinflammation and apoptosis in the brain, although the full extent of the relationship between sepsis-related immunometabolic dysfunction and SE pathophysiology is not yet well understood. Questions remain in elucidating mechanisms of injury and the roles of specific biological molecules to serve as potential early SE biomarkers. In a mouse model of acute polymicrobial sepsis induced by cecal slurry injections in mice aged 8-12 weeks (n=8) and respective controls (n=8), we measured cerebellum and plasma metabolites using liquid chromatography-mass spectrometry (LC-MS-MS) for targeted and untargeted metabolomics. The bioinformatic tool MetaboAnalystR was used to detect metabolites with significantly different levels between groups and generate receiver operating characteristic (ROC) curves for the identification of metabolites with diagnostic biomarker potential. Combined targeted and untargeted metabolomics yielded 498 metabolites in cerebellar tissue and 697 in plasma samples. In the cerebellum, the top 10 altered metabolites in sepsis were involved in energy production, such as 3-hydroxy-3-methylglutaryl-CoA, nucleotide metabolism and the citrulline-nitric oxide cycle, known to be activated in inflammation. To determine which of the metabolites affected in the cerebellum could be linked to a potential plasma biomarker, metabolites with 1) common directionality of statistically significant change in both cerebellum and plasma and 2) an area under the curve (AUC) value over 0.8 based on the ROC analysis were selected. This cross-compartment analysis revealed 6 plasma metabolites with strong diagnostic potential for cerebellar injury in SE, namely 1-Methyl-L-Histidine, Putrescine, N-

Acetylglutamine, Urea, Cystathionine and N-Acetyl-L-Aspartic acid. Overall, these findings define key cerebellar metabolic mechanisms of SE and propose plasma biomarkers for SE diagnostics. This is especially important as we strive to improve diagnostic criteria, identify reliable biomarkers, and extend the clinical window for disease prediction and timely intervention in SE.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Program #/Poster #: PSTR274.01/G15

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Wellcome Leap 1kD Program (The First 1000 Days; 222076/Z/20/Z)

Title: Regional motor network functional connectivity alterations in early infancy persist at 12-18 months in the context of perinatal HIV exposure

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Abstract: Introduction

Children who are HIV-exposed uninfected (CHEU) are often exposed to HIV and/or antiretroviral therapy during perinatal brain development. Existing literature shows that CHEU often have impaired motor function in early life compared to HIV-unexposed children (CHU). Mechanisms underlying these delays include inflammatory biomarkers and altered neurometabolic signatures. Functional brain networks, especially higher-order networks like the default mode, salience, and language networks, are linked to early developmental outcomes. Building on our prior work showing functional connectivity (FC) differences in the motor resting state network (RSN) in CHEU during early infancy, this study assessed whether such alterations persist at 12-18 months of age.

Methods

As part of the Khula birth cohort in Cape Town, South Africa, resting state fMRI was acquired from 57 infants (21 CHEU, 36 CHU) aged 12-18 months during natural sleep on a 3T Siemens Skyra scanner. A 16-channel head coil and 360 volumes (TR/TE=940/35ms, voxel size=2×2×2mm³) were acquired. Data were preprocessed using AFNI, aligned to UNC-BCP infant atlases (12- or 18-month templates), smoothed, and denoised. Group ICA was conducted using MELODIC in FSL to identify motor RSNs, followed by ROI-to-ROI FC analysis using Pearson correlations. Multivariable linear regression models compared FC between CHEU and CHU, adjusting for sex, age, and prenatal alcohol exposure, with Benjamini-Hochberg correction for multiple testing.

Results

After quality control, 54 participant data were analyzed (20 CHEU, 34 CHU, 56% male, median age 478 days). ICA identified 26 ROIs and 325 motor RSN connections. Twelve ROI connections showed significantly altered FC in CHEU ($p < 0.05$), with 11 showing reduced connectivity. Three connections left precentral to right postcentral, left superior frontal to right postcentral, and left superior frontal to right precentral were also altered at 6 months. Bilateral changes between the superior frontal and postcentral gyri were also noted (left: $B = -0.17$, $p = 0.025$; right: $B = 0.15$, $p = 0.003$).

Discussion

Findings suggest persistent motor RSN differences in CHEU into the second year of life. Reduced connectivity involving the precentral (motor) and postcentral (sensory) gyri may reflect disrupted sensorimotor integration due to perinatal HIV exposure. These changes may underlie early motor delays.

Conclusion

Persistent alterations in motor RSNs at 12-18 months in CHEU highlight potential biomarkers of early neurodevelopmental risk. Future work should link FC changes to developmental outcomes to better understand the long-term effects of perinatal HIV exposure.

Disclosures: S.R. Williams: None.

Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

Location: SDCC Halls B-H

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

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Title: Elevated inflammation in people with HIV is associated with aberrant neural oscillations serving selective attention

Authors: *K. M. MCDONALD^{1,2}, J. J. SON^{1,3}, M. SCHANTELL¹, N. M. PETRO¹, R. GLESINGER¹, H. J. OKELBERRY¹, J. A. JOHN¹, M. C. ZIMMERMAN³, R. K. SPOONER^{1,2}, T. W. WILSON^{1,2};

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Abstract: Background: Medical advances have greatly improved the quality of life and extended the longevity of people with HIV (PWH). However, despite effective viral suppression, many PWH still develop cognitive deficits, which can greatly impact their ability to perform daily activities. Such deficits have been linked to aberrant oscillatory activity in primary sensory and higher-order cortical regions, with some suggesting the persistently high inflammatory levels may drive these aberrations. Tasks probing selective attention, the ability to allocate attentional resources to relevant cues and inhibit irrelevant information, commonly reveal abnormal neural responses in PWH. However, it is unclear whether elevated inflammation may impact such responses among PWH and ultimately degrade selective attention. Hypothesis: Inflammation will be greater among PWH and differentially scale with frontoparietal neural activity during selective attention compared to controls. Methods: Magnetoencephalography data from 76 healthy controls (HC) and 69 PWH were collected during a Simon task and imaged in the time-frequency domain with a beamformer. Participants were shown 3 digits and identified via button press the digit that differed from the others, responding to its numerical identity (i.e., 1, 2, 3) and not its spatial location. Plasma from whole-blood samples were examined using multiplex immunoassays, and a neuroinflammation index (NI) was computed from concentrations of key inflammatory biomarkers implicated in CNS function (i.e., TNF- α , CRP, IP-10). Functional maps were subjected to whole-brain linear mixed effect models to identify relationships between the NI and oscillatory activity among HC and PWH ($p < .005$, corrected). Results: PWH were significantly slower to respond during the task and had an elevated NI compared to HC. Significant 3-way condition-by-group-by-inflammation interactions in the theta (4-8 Hz) and alpha (8-14 Hz) ranges were identified in task-related brain regions, with greater NI scores among PWH scaling with stronger theta responses within frontoparietal cortices. Conversely, among HC, greater NI scores scaled with weaker theta in these regions. In the alpha range, weaker parietal activity was significantly correlated with a greater number of years since HIV diagnosis and longer time on antiretroviral therapy controlling for age. Conclusion: These findings support the view that elevated inflammation among PWH may drive the aberrant oscillatory neural activity and cognitive dysfunction that remains common in the era of viral suppression. Interventions targeting such elevated inflammation may mitigate cognitive decline.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

Location: SDCC Halls B-H

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Program #/Poster #: PSTR274.03/H1

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R01DA047906
NIH Grant P30MH062512

Title: Cannabis use in young adults may mitigate HIV-related effects on cortical structure and neuroinflammation

Authors: A. K. MACH¹, D. STALEY⁶, K. HALL², A. MANGAT¹, T.-C. WU³, A. BISCHOFF-GRETHER¹, A. E. LANSING^{1,7}, J. JACOBUS¹, X. TU³, S. L. NICHOLS⁴, *C. FENNEMA-NOTESTINE⁵;

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Abstract: In young adults living with HIV (YWH), cannabis use has risen and may alter immune suppression and neuroinflammation, both pathways underlying the CNS manifestations of HIV infection. Their interaction may alter brain integrity and continuing CNS development. Based on our preliminary work, we hypothesized that while high levels of cannabis use may exacerbate HIV-related smaller caudate volume and thinner prefrontal cortex, moderate use may mitigate these effects, leaving YWH and seronegative controls more similar with respect to gray matter integrity and neuroinflammation. In a cross-sectional study, we acquired structural MRI and proton MR single voxel spectroscopy (MRS) for 81 young adults aged 18-24 years ($\bar{x}$ = 21.5) with HIV infection (n = 30) or without (n = 51). Frequency of cannabis use across one year was categorized as none (n = 17), light/moderate (1-259 days/year, n = 42), or high ($\geq$ 260 days/year, n = 22). We used a semiparametric generalized linear model to model interactions of HIV status and cannabis use on MRI outcomes. Covariates included age; intracranial vault volume for volumetric analyses; and the phantom reference value for MRS analyses. We found that YWH had thinner left prefrontal cortex in the left pars opercularis (β = -0.22, p < 0.000) and the left rostral middle frontal regions (β = -0.11, p = 0.002), and smaller caudate volumes (left: β = -306.8, p = 0.040; right: β = -262.0, p = 0.043). YWH also had higher levels of frontal white matter myoinositol (MI) suggesting greater neuroinflammation (β = -0.69, p = 0.006). Interactions between cannabis use and HIV status were evident in left prefrontal cortex: YWH had thinner cortex only in the non-cannabis using category for the left pars opercularis (Moderate vs None: β = 0.21, p = 0.004; High vs None: β = 0.27, p = 0.001) and the left rostral middle frontal (Moderate vs None: β = 0.17, p < 0.000; High vs None: β = 0.17, p = 0.002). This interaction was not significant for caudate volume. YWH also demonstrated higher levels of MI in frontal white matter for non-cannabis users only, suggesting neuroinflammation; however, relative to controls, YWH had similar levels with light/moderate cannabis use (β = -.097, p = 0.006) and lower levels with high cannabis use (β = -1.46, p = 0.002). For these comparisons, seronegative controls were not different across cannabis use categories. Consistent with our hypothesis, these cross-sectional results suggest that some cannabis use may mitigate neuronal and neuroinflammatory effects associated with HIV infection. Future work will examine associations with cognitive performance and behavioral assessments in more detail to assess clinical relevance of these findings.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH grant R01 DA044552
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Title: Chronic cocaine self-administration disturbs K^+ channel activity in cortical astrocytes of rats and that is worsened by neuroHIV but diminished by chronic cART treatment

Authors: *L. CHEN, S. L. CASSODAY, D. IMERI, L. AL-HARTHI, X.-T. HU;
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Abstract: Cocaine (COC) is a highly addictive and widely abused psychostimulant. Chronic exposure to COC or HIV induces neuron hyperactivity in the medial prefrontal cortex (mPFC, a critical regulator of cognition and addiction); and this neuron dysfunction is worsened when they are merged. Although combined antiretroviral therapy (cART) suppresses active HIV replication and prolongs life of HIV⁺ individuals, the prevalence of HIV-associated neurocognitive disorders (HAND, a.k.a. neuroAIDA/HIV) still occurs in ~50% of people living with HIV. Our previous work and research of others indicate that besides cART's therapeutic effect, chronic Triumeq treatment (a first-line cART regimen) induces neurotoxicity. Previous studies also suggest that COC, HIV, and cART-induced cortical neuron dysfunction results from the alterations in their synaptic/intrinsic excitability, alone with astrocyte dysfunction that disturbs extracellular glutamate and K^+ homeostasis. However, the effects of chronic COC-SA, neuroHIV, cART on astrocytic K^+ channel activity, and their underlying mechanism, remain unknown. To fill this knowledge gap, we used whole-cell patch-clamping approach to assess the functional activity of different subtypes of K^+ channels in mPFC astrocytes in brain slices from COC-SA and HIV-1 transgenic (HIV-Tg) rats (~6-month-old) that also received Triumeq pretreatment. Age-matched F344 non-Tg rats were used as control. Rats received repetitive daily s.c. injections of Triumeq for 28 days (d). On d15, they self-administered (SA) COC for 14d followed by 2-5d of withdrawal. We found that there was no significant change in K_v , K_{ir} and K_{2P} channel activity (reflected by K^+ efflux/influx) in mPFC astrocytes in Saline (SAL)-Yoked HIV-Tg rats compared to SAL-Yoked non-Tgs. There was also no change in K_{2P} channel activity of mPFC astrocytes after COC-SA, regardless of genotype. However, COC-SA enhanced K_v and K_{ir} channel activity in non-Tg rats, but decreased the activity of astrocytic K_v and K_{ir} channels in mPFC astrocytes of HIV-Tg rats. All these COC-SA-induced K^+ channel dysfunctions were reversed in response to combined Triumeq and COC exposure. Collectively, this study demonstrates that chronic COC-SA induces K_v , K_{ir} , but not K_{2P} channel dysfunction in mPFC astrocytes in the brain. This not only evokes astrocyte reactivity, but also worsens astrocyte dysfunction in the context of neuroHIV, which could enhance neurotoxicity by elevating $[K^+]_e$ or damage astrocytes by rising intracellular osmolarity. Further study needs to define if COC-SA

plus Triumeq-induced reversal of K⁺ channel dysfunction was due to a protective mechanism or damage.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Program #/Poster #: PSTR274.05/H3

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH NIDA Grant 1U01DA053630-01

Title: Cell-type-specific transcriptomic and epigenomic responses to HIV and opioid use disorder in the human brain

Authors: *A. GREEN¹, J. BUCHANAN², K. GAULTON³, A. WANG⁴, E. A. MUKAMEL⁵; ¹UCSD Dept. of Neurosciences, San Diego, CA; ²Ctr. for Epigenomics, ⁴Ctr. of Epigenomics, ³Univ. of California San Diego, San Diego, CA; ⁵Cognitive Sci., Univ. of California San Diego, La Jolla, CA

Abstract: HIV and opioid use disorder (OUD) are intertwined epidemics. 20-50% of people with HIV are prescribed opioids for chronic pain, while one third of individuals with OUD are at higher risk of HIV infection due to injection drug use. Opioids exacerbate cognitive dysfunction and systemic inflammation in people living with HIV, yet little is known of how HIV and OUD impact gene expression and regulation across human brain cell types and regions. Understanding the cell-type and region-specific mechanisms that underlie HIV and OUD's neuropathogenesis will critically inform the development of targeted therapies for neurodegeneration. Generated as part of the Single Cell Opioid Responses in the Context of HIV (SCORCH) consortium, here we present a multi-omic single-nucleus transcriptome (snRNA-seq) and chromatin accessibility sequencing (snATAC-seq) dataset. This dataset consists of approximately 580,000 high-quality single-nuclei profiles across post-mortem prefrontal cortex (PFC), cerebellum, and amygdala tissue in 45 male and female donors. Although glial cell types across the PFC, amygdala and cerebellum exhibit high transcriptomic and epigenomic correlation, we found that cerebellar glia may be under greater metabolic load compared to the amygdala and PFC. In PFC microglia, gene co-expression network analyses of HIV donors compared to Controls show upregulation of interferon signaling, DNA damage repair, and response to viral processes in addition to downregulation of T-cell differentiation and activation pathways. In contrast, OUD predominantly altered the transcriptome of granule cells and PFC excitatory cell types. Specifically, we observed widespread downregulation of energy metabolism pathways in PFC excitatory neurons, and downregulation of genes involved in synaptic transmission in granule cells. Further, analyses suggest that HIV and OUD interact to respond to HIV and OUD-induced

neurotoxicity, with genes including TMEM164 upregulated in astrocytes. Overall, these results provide insight into cell-type specific transcriptional changes and regulatory mechanisms associated with HIV and OUD across brain regions, identifying potential targets for therapeutic intervention to address HIV and OUD-associated neuroinflammation and degeneration.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Program #/Poster #: PSTR274.06/Web Only

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: DA060085
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DA044579
DA059849
HL126559
MH128022
MH072567

Title: Insulin-like growth factor-1 mediates pericyte-neural progenitor cell communication following methamphetamine and hiv-1 exposure of neural progenitor cells

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Abstract: Methamphetamine (METH) use and HIV-1 infection are independently associated with blood-brain barrier (BBB) dysfunction and altered neurovascular interactions. Insulin-like growth factor-1 (IGF-1) is a neurotrophic factor involved in neurovascular health, but its role in mediating communication between neural progenitor cells (NPCs) and brain microvascular pericytes under METH and HIV-1 exposure remains unclear. In a mouse model, co-exposure to METH and HIV-1 increased IGF-1 mRNA expression in subventricular zone (SVZ) NPCs and the caudate putamen, while HIV-1 alone significantly elevated IGF-1 mRNA in SVZ tissue. IGF-1 protein levels remained unchanged in the frontal lobe; however, plasma IGF-1 levels were reduced in HIV- and METH+HIV-exposed mice. In vitro, ReNcells (a human NPC line) exposed to HIV-1 or METH+HIV exhibited increased Ezrin phosphorylation and F-actin polymerization—changes that were reversed by Ezrin inhibition, which restored NPC morphology. METH+HIV exposure also suppressed phosphorylation of FAK, ERK, and AKT in NPCs. Pharmacological inhibition of FAK reduced ERK and Src activity; Cdc42 inhibition decreased Src phosphorylation; and Ezrin inhibition modestly reduced ERK and Src activation, suggesting coordinated regulation of cytoskeletal and survival signaling. Conditioned media

(CM) from ReNcells treated with HIV or METH+HIV significantly decreased FAK, ERK, and AKT phosphorylation in primary brain pericytes. Notably, IGF-1 neutralization in CM restored ERK and AKT signaling, indicating that IGF-1 contributes to NPC-mediated suppression of pericyte survival pathways under pathological conditions. These findings highlight a novel role for IGF-1 in modulating pericyte responses to HIV-1 and METH via NPC-derived signals.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Contract 75N95024C00035

Title: A human iPSC-derived CNS tri-culture model of NeuroHIV for studying neurotoxic and neurocognitive effects of HIV-infection and anti-retroviral (ARV) treatment

Authors: *C. G. RINES, L.-M. OJA, A. S. SMITH, L. HARRISON, S. K. GILMORE-HALL, J. H. PRICE, P. M. MCDONOUGH, K. L. GORDON, B. TEKWANI; Vala Sciences, Inc., San Diego, CA

Abstract: There are about 39.9 million people living with HIV (PLWH) worldwide with about 1.3 million new infections per year. Left untreated, infection with HIV will lead to AIDS and ultimately death. Antiretroviral (ARV) therapy has been lifesaving for PLWH but the drugs have been linked to HIV associated neurocognitive decline (HAND), a condition that encompasses a range of cognitive impairments that can significantly impact daily functioning and quality of life for PLWH.

We have developed a hiPSC-derived CNS tri-culture model of neurons, astrocytes and microglia (BrainXell) to investigate neurotoxic effects of HIV infections and ARV treatment. To study the effects of ARVs we treated tri-cultures with ARVs either alone (Dolutegravir, Elvitegravir, Tenofovir Disoproxil Fumarate, Emtricitabine) or in clinical combinations at four concentrations, for 7 days. Tri-cultures were then loaded with the calcium indicator Cal-520 and calcium transients were measured for 2 min at 4Hz using Vala Sciences IC200 Kinetic Image Cytometer (KIC®). Following calcium imaging, cultures were fixed and immunostained for TUBB3 (neurons) and IBA 1 (microglia). Calcium activity and cell morphology analysis was performed using Vala Sciences' Image analysis software CyteSeer®. Analysis revealed Dolutegravir alone and in combinations at 3xC_{max} concentration caused significant calcium changes, and the Dolutegravir/Elvitegravir combinations caused significant microglial loss and morphological changes at 1xC_{max} and 3xC_{max} concentrations.

To establish a model for CNS HIV infection (NeuroHIV), hiPSC-derived tri-cultures and microglia monocultures were infected with a GFP-expressing HIV strain (Virongy Biosciences)

targeting microglia, the CNS's primary HIV reservoir. The IC200 KIC® was used to image GFP+ cells in culture over time. GFP+ microglia were observed as early as 24 hours after infection and increased over time. Additionally, HIV replication in microglia was blocked after treatment with dolutegravir compared to untreated control. HIV p24 levels were analyzed by ELISA to confirm infection over time. The infected CNS tri-cultures may be assessed for neurotoxicity from HIV-infection and ARVs by measuring neuronal calcium transients and cell viability.

This CNS model of HIV infection developed here can be used to measure both efficacy and toxicity of current and future anti-retroviral therapies. The human-iPSC-based NeuroHIV model offers a translatable-platform for evaluating HIV and ARV-induced cognitive side effects and reducing animal use in drug development.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

Location: SDCC Halls B-H

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Title: Dopaminergic modulation of neuroimmune responses in HIV-infected iPSC-derived cortical and midbrain organoids exposed to stimulant drugs

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Abstract: Human immunodeficiency virus (HIV) affects almost 40 million people worldwide, and nearly half will develop neurologic symptoms. HIV enters the CNS within days of infection, where data suggest it establishes a persistent reservoir in brain-resident immune cells such as microglia. Chronic viral replication and neuroinflammation in response to infection drive synaptodendritic injury, contributing to deficits in cognition, mood, and movement. Use of addictive drugs, such as methamphetamine (METH), is more prevalent among people with HIV, and stimulants have been shown to worsen neuroinflammation and increase viral load. Our

published data show that dopamine (DA) increases inflammation and HIV replication in myeloid cells in vitro, and we hypothesize that disruptions in dopaminergic neurotransmission contribute to exacerbated neuroinflammation and increased CNS infection. To test this, we developed human induced pluripotent stem cell (iPSC)-derived cortical and midbrain organoids co-cultured with iPSC-derived microglia, representing low (cortex) and high (midbrain) dopaminergic tone. Organoid identity and microglial integration were validated via immunostaining, gene expression profiling, and single-cell RNA sequencing. Multi-electrode array recordings revealed increased spontaneous firing rates and synchrony in midbrain versus cortical organoids in response to METH treatment, and METH-induced dopamine release in midbrain organoids was confirmed using a live-cell GRAB:DA2 dopamine sensor assay. Organoid-microglia co-cultures were inoculated with HIV in the presence or absence of METH or DA, then cultured for five weeks. Changes in HIV replication and inflammatory response were assessed via staining for p24⁺ cells and AlphaLISA measurement of p24, IL-6, CXCL10, IL-1 β and TNF- α secretion in supernatants. Astrocytic activation was assessed by GFAP immunostaining, and synapse loss was quantified by synaptophysin and PSD95 puncta. Results indicate that local changes in dopamine concentration in response to METH increased infection and amplified HIV-associated dysregulation of regional neuroimmune response. These data also highlight the importance of accounting for neuroimmune heterogeneity when studying neuroHIV and its interaction with neuropsychiatric comorbidities.

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Poster

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Support: RS-2024-00508420 funded By the Ministry of Trade Industry & Energy (MOTIE, Korea)

Title: High-resolution spatial dissection of zikv-host Interactions using an integrated SLACS and SELECT-seq approach in brain organoid models

Authors: *H. RHO¹, S. LEE², A. C. LEE²;

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Abstract: Zika virus (ZIKV) infection disrupts fetal brain development by targeting neural progenitor cells, but the spatial heterogeneity of virus-host interactions within the complex brain microenvironment remains poorly understood, necessitating spatial transcriptomic analysis to

elucidate these mechanisms. We present an integrated spatial transcriptomics platform that combines Spatially-resolved Laser-Activated Cell Sorting (SLACS) with full-length RNA sequencing (SELECT-seq), developed to dissect virus-host interactions within preserved tissue architecture. Using iPSC-derived human brain organoids infected with ZIKV, we performed high-resolution, image-guided microdissection of ~5-10 cells per region of interest (ROI), targeting ZIKV-infected and uninfected SOX2⁺ neural progenitor cells and TUJ1⁺ differentiated neurons identified through immunostaining and viral protein (ZIKV) expression. SLACS enabled precise isolation of these regions while maintaining spatial context and pathological features. From the isolated cells, spatially barcoded full-length cDNA libraries were generated to simultaneously profile host transcriptomes and ZIKV RNA. Viral RNA was identified via BLAST-based alignment to the reference genome, allowing quantification of viral load and detection of spatially localized sequence variants. In parallel, host transcripts were mapped to the human genome, and differential gene expression and A-to-I RNA editing were analyzed using REDItools and ANNOVAR. This integrative approach revealed region-specific transcriptomic and epitranscriptomic alterations associated with mitochondrial dysfunction and innate immune activation. Furthermore, we identified spatially clustered viral mutations, indicating local viral evolution within infected tissues. The SLACS-SELECT-seq platform enables spatially targeted, high-resolution, full-length RNA profiling of both host and pathogen within complex tissue environments, advancing neurovirology, spatial (epi-)transcriptomics, and organoid-based disease modeling. Our findings underscore the power of spatial transcriptomics to uncover localized virus-host interactions, reveal mechanisms of ZIKV-driven neurodevelopmental disruption, and support spatial analysis of pathogenesis across diverse biomedical contexts.

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Poster

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: DA059849
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Title: Investigating the Neuroimmune Basis of HIV Induced Cognitive and Neuropsychiatric Dysfunction

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Abstract: People living with HIV (PLWH) experience high levels of depression with a lifetime prevalence of 42%. They also experience a higher risk of cognitive dysfunction which often correlates with their degree of depression. Cognitive impairment in HIV infection is also associated with atrophy of the dopamine-rich caudate nucleus, suggesting that the loss of dopaminergic neurons could drive both depression and cognitive impairment in PLWH. HIV protein, Tat, can interact directly with dopamine transporters (DAT) to modulate extracellular dopamine levels and increase catecholamine induced oxidative stress and neurotoxicity.

Additionally, in PLWH receiving anti-retroviral treatment, neuroinflammation not only persists but also correlates with cognitive changes. Molecularly, HIV infection and several HIV proteins including Tat, gp120, and viral protein R (VPR) have been shown to induce reactive oxygen species (ROS) production and oxidative stress as well as neuroinflammation. Taken together, these studies suggest that neuroinflammation and toxicity, especially in dopaminergic areas of the brain associated with degeneration in HIV infection, drive cognitive changes and depressive symptoms. The present study focuses on profiling mice for depression, cognition, and brain inflammatory responses over a month of infection with ecoHIV, a murine HIV model.

A series of behavioral tests indicated cognitive impairments and increased susceptibility to depressive-like behaviors of mice infected with ecoHIV in a time-specific manner. Sociability assays trended toward less social behavior at 1 and 3 weeks post infection. Moreover, mice 1 week post infection demonstrated worse spatial reference memory and a trend toward worse object recognition. At 3 weeks post infection, mice also exhibited a trend toward worsening object recognition. Mice were then subjected to brain and serum inflammatory profiling to associate changes in systemic- and neuro- inflammation with behavioral differences over time. We also propose the repurposing of FDA approved drugs, especially those which mediate dopaminergic signaling, to mediate the inflammatory and neuropsychiatric effects of HIV infection. Overall, our studies meet a critical need for understanding the mechanism of neuropsychiatric dysfunction, neuroinflammation, and neurotoxicity in PLWH to allow for better treatment of HIV associated neuropsychiatric dysfunction and subsequently improve quality of life.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Contract 75N95023C00013
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Title: Working memory and executive functioning impairment are associated with changes in cortical structure and neuronal integrity in people with HIV

Authors: *A. BISCHOFF-GRETHER¹, A. K. MACH¹, C. FENNEMA-NOTESTINE², D. R. FRANKLIN¹, D. J. MOORE¹;

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Abstract: The National NeuroAIDS Tissue Consortium (NNTC) is a multisite resource that includes neuromedical, neurocognitive, neuroimaging, and post-mortem tissue, mainly for persons with HIV (PWH), including older individuals. This rich resource enables analyses of specific domains of neurocognitive effects of HIV infection in an aging cohort, and their relationship to brain integrity. In this preliminary analysis, using cross-sectional data acquired by the California NeuroAIDS Tissue Network (CNTN) site of the NNTC, we examined derived structural MRI measures of cortical thickness and metabolite levels from proton MR single-voxel spectroscopy (MRS), as well as processed cortical myelin density based on a T1w/T2w ratio, to assess their relationship with cognitive impairment in older PWH (n = 17, mean age = 64.4 [52, 76]) using multivariable linear regression. Covariates included age and the phantom reference value for MRS analyses. Analyses focused on working memory (WM) and executive function (EF) domains. Relative to unimpaired PWH (n=9), PWH with WM impairment (n=8) exhibited significantly thinner prefrontal cortex in the left caudal anterior cingulate ($\beta = 0.20$, $p = 0.021$) and trended towards thinner cortex in the right rostral middle frontal cortex ($\beta = 0.06$, $p = 0.070$), regions commonly associated with working memory. PWH who exhibited WM impairment also trended towards lower myelin content in the left caudal middle frontal region relative to those without impairment ($\beta = 0.004$, $p = 0.051$), suggesting altered cortical microstructure. There was also a trend towards lower N-acetyl aspartate (NAA) levels in individuals with WM impairment in frontal white matter ($\beta = 0.23$, $p = 0.094$), suggesting altered neuronal integrity. Individuals with EF impairment (n=7) trended towards thinner cortex in the right rostral middle frontal ($\beta = 0.07$, $p = 0.065$) and the right caudal middle frontal regions ($\beta = 0.08$, $p = 0.092$), relative to the unimpaired group (n=10). Myelin content also was significantly lower in the left caudal anterior cingulate cortex for those with EF impairment ($\beta = 0.02$, $p = 0.025$). Despite the small sample, these findings suggest cognitive impairment in older people with HIV is associated with thinner prefrontal cortex and altered neuronal integrity. These preliminary results are important steps towards a better understanding of the neuroimaging correlates of cognitive impairment in older people with HIV. Future work with the larger NNTC cohort will allow more detailed analyses between HIV-related cognitive performance, neuroimaging profiles, and important factors such as antiretroviral treatment, substance use, and depression.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR274.12/H9

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Swedish Medical Research Council
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Title: Sustained Activation of the Kynurenine Pathway in CSF Is Linked to Cognitive Deficits in Patients 1.5 Years After Acute Tick-Borne Encephalitis

Authors: *S. K. ERHARDT, X. LI, J. AHLBERG, A. ATLAS, E. FRANZEN-RÖHL, G. C. ENGBERG;
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Abstract: Emerging evidence highlights cognitive impairment as a significant sequela of viral infections. Since the onset of the COVID-19 pandemic, a significant proportion of patients have exhibited neurological and psychiatric symptoms, especially cognitive decline, with an increased incidence persisting post-recovery. Similarly, tick-borne encephalitis (TBE) frequently leads to psychoneurological impairments, reducing patients' quality of life and resulting in substantial labor force losses for society. Viral infections are recognized as early environmental risk factors for schizophrenia, yet the mechanisms by which viruses affect the central nervous system (CNS) remain insufficiently understood.

The dysregulation of the kynurenine pathway in the brain appears to be long-term. CSF and serum samples from a cohort of Swedish TBE patients have been analyzed to discover the dynamics of kynurenine pathway metabolites over a longer period, for demographics. Preliminary results from this observational study reveal persistently elevated CSF levels of tryptophan, kynurenine, KYNA, and QUIN 1.5 years post-infection, suggesting prolonged metabolic disturbances following an acute TBE infection. Consistent with the findings presented in Lithuanian patients (Brain, Behavior, and Immunity 125 (2025): 452-465), Swedish patients with acute TBE infection display elevated CSF levels of kynurenine and its downstream metabolites, including KYNA, QUIN and nicotinamide. Notably, longitudinal data shows that CSF tryptophan levels remain persistently elevated, and CSF kynurenine levels stay high for up to 1.5 years after infection. Although CSF KYNA and CSF QUIN concentrations gradually decrease over time, they are still significantly higher than those observed in control subjects 1.5 years post-infection. Together, with our other studies, these results support the hypothesis that elevated levels of KYNA is a common pathophysiology in neuroimmune disorders.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR274.13/Web Only

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Title: Glial fibrillary acid protein and neurofilament light chain concentrations on post COVID-19 condition subjects and its association with cognitive impairment, depression, and anxiety symptoms

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Abstract: There are multiple hypotheses for Post COVID-19 condition in the nervous system, the most accepted is neuroinflammation. Elevated concentrations of Neurofilament Light Chain and Glial Fibrillary Acid Protein have been related to cognitive impairment and other neuropsychiatric symptoms such as depression, anxiety, and PTSD. Therefore, this research had the objective to determine the relationship between plasmatic concentrations of NfL and GFAP and the presence of cognitive impairment, depression, anxiety, and stress. The present study was a quantitative study with a cross-sectional, analytic design. 41 subjects were divided between two groups: mild and asymptomatic SARS-CoV-2 infection and moderate and severe asymptomatic SARS-CoV-2 infection. Sociodemographic data, SARS-CoV-2 infection data, DASS-21 tests, MoCA test, and quantitative ELISA tests for NfL and GFAP were evaluated. 41 subjects were studied with a mean age of 26 ± 9.85 years. 65.85% were students, 29.26% employees and 4.87% self-employees. 100% had bachelor's degree or superior. Mean of months since recovery from infection was 22.73 ± 11.95 . 11 subjects had Post COVID-19 condition, 90.1% were from the group with moderate or severe infection. 75.61% (n=31) of samples were cases. The severity of SARS-CoV-2 infection was related to cognitive impairment with 64.51%, $p=0.003$, OR= 16.36 (1.82-146.66), depression with 74.19%, $p<0.001$, OR=3.87 (2.13-7.03), both anxiety and stress with 87.09%, $p<0.001$, OR=27 (4.15-175.49). There were no differences between protein concentrations among groups. Moderate correlation was established among NfL and GFAP concentrations ($p=0.006$), Post COVID-19 condition and female sex ($p=0.001$), severe anxiety and severe depression ($p=0.001$), severe stress and severe depression ($p=0.002$), severe anxiety and severe stress ($p<0.001$) and a mild correlation among Post COVID-19 condition and NfL concentrations ($p=0.05$) and severity of infection and female sex ($p=0.036$). Altogether, this study suggests that cognitive impairment, depression, anxiety and stress are related to SARS-CoV-2 infection severity, and finally, that higher concentrations of NfL are related Post COVID-19 condition.

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Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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Program #/Poster #: PSTR274.14/H10

Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

Support: Long-COVID Registry / Universidad Simon Bolivar

Title: Major Depressive Disorder in Long COVID and Exposure to Pro-inflammatory Cytokines During Infection by SARS-CoV-2

Authors: *H. GUILLEN-BURGOS^{1,2};

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Abstract: Objective: Major Depressive Disorder (MDD) is common in long COVID syndrome; however, the neurobiological mechanisms are unclear. An immune activation response has been associated with COVID-19 severity as well as in MDD. We hypothesize that high levels of pro-inflammatory cytokines during SARS-CoV-2 infection may be associated with new-onset MDD and severe outcomes such as treatment-resistant depression (TRD) and risk of suicide ideation and behavior (SI/SB).

Methods: A case-control nested to a cohort study was carried out on a total of 678 COVID-19 survivors (MDD = 184 vs. non-MDD = 494). A pro-inflammatory panel of serum levels of cytokines (IL-1 β , IL-4, IL-6, IL-8, IL-13, IL-17 α , TNF- α) was evaluated during COVID-19 hospitalization and severe outcomes such as TRD and SI/SB were assessed in individuals with new onset of MDD after hospital discharge compared to non-MDD COVID-19 survivors.

Results: High levels of pro-inflammatory cytokines during SARS-CoV-2 infection were identified in MDD participants compared to non-MDD subgroups during long COVID. These differences were sustained also for TRD and SI/SB severity outcomes. There is a mild association of high levels of pro-inflammatory cytokines and MDD, TRD, and SI/SB.

Conclusion: High levels of pro-inflammatory cytokines during severe or critical COVID-19 exposure may explain long COVID associated MDD and thus severe outcomes such as TRD and SI/SB.

Disclosures: H. Guillen-Burgos: None.

Poster

PSTR274: Viral Infection: Impact on Neural Circuit Function

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CAPES #88887.508608/2020-00

Title: Sars-cov-2 infection disturbs neural function, compromising synaptogenesis and glutamate release, inducing neuroinflammation

Authors: *N. CARRIJO OLIVEIRA^{1,2}, F. B. RUSSO¹, F. R. TOCANTINS^{1,2}, A. CASTANHA^{1,2}, E. SEGABINAZI², P. C. B. BELTRÃO-BRAGA^{1,2};

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Abstract: SARS-CoV-2 rapidly spread worldwide, infecting millions. Although primarily a respiratory virus, it has been linked to neurological and psychiatric-related symptoms, such as loss of smell/taste, headache, cognitive and memory issues ("brain fog"), and dizziness. Using 2D neural progenitor cells (NPC) and neurons cell cultures and 3D brain organoids derived from human induced pluripotent stem cells (hiPSC), we investigated the neurobiological mechanisms of SARS-CoV-2 infection with low viral load (MOI=0,5). We used a Brazilian SARS-CoV-2 B lineage (GenBank: MT350282.1) and three hiPSC lineages (Russo et al., 2018), in experimental triplicate. We applied RT-qPCR for cell and virus molecular targets, a spectrophotometric assay for glutamate release quantification, Luminex xMAP for cytokine detection, and immunofluorescence with quantification for cellular and viral protein targets. We found that NPC (t=6.736, df=4, p=0.0025) and neurons (t=14.33, df=4, p=0.0048) are susceptible to SARS-CoV-2, and upon infection, the cells underwent activation of the apoptotic pathway (NPC: t=4.6788, df=6, p=0.0034; neuron: t=2.923, df=6, p=0.0265). Additionally, the infection induced a neuroinflammatory response, marked by increased release of pro-inflammatory cytokines, especially interleukin 6 (IL-6) (NPC: t=28.83, df=5, p<0.0001; neuron: t=29.809, df=4, p<0.0001; organoid: t= 4.9982, df=4, p=0.0075). We also observed an increase in glutamate production in both cortical neurons (F (1, 6) = 17.30, p=0.0059) and brain organoids (F (1, 4) = 239.7, P=0.0041), accompanied by a decrease in the expression of Excitatory Amino Acid Transporter 1 (EAAT-1) (t=20.07, df=4, p=0.0025) and a tendency increase in EAAT-2 expression (t=3.044, df=4, p=0.0931), suggesting a compensatory mechanism for clearing excess glutamate. Moreover, we identified an imbalance in synaptic components, characterized by elevated postsynaptic density protein 95 (PSD95) (t=4.751, df=7, p=0.0021) and reduced presynaptic Synapsin 1 (Syn1) (t=4.682, df=7, p=0.0023), reflecting synaptic function impairment. These findings highlight the disruption of neuronal connectivity and cell death as part of the disease, shedding light on the neurological consequences of SARS-CoV-2 infection. Russo, FB, et al. (2018). Modeling the Interplay Between Neurons and Astrocytes in Autism Using Human Induced Pluripotent Stem Cells. *Biological psychiatry*, 83(7), 569-578.

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Poster

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Topic: D.01. Neurotoxicity, Inflammation, and Neuroprotection

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Title: Targeting lung endothelium-derived tau to mitigate pneumonia-induced cognitive deficits and tau pathology

Authors: C.-S. CHOI¹, M. ALEXEYEV¹, C. ZHOU¹, S. L. SAYNER¹, R. STEVENS¹, R. BALCZON¹, A. BAUMAN¹, S. CHANEY², A. R. NELSON³, B. WAGENER⁴, T. STEVENS¹, *M. T. LIN¹;

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Abstract: The lung endothelium is a key site for the expression of peripheral tau, a protein commonly associated with neurodegenerative diseases. In previous studies, we have investigated the role of lung endothelium-derived tau in the context of pneumonia-induced cognitive deficits. Specifically, we explored how infection with *Pseudomonas aeruginosa* (*P. aeruginosa*) influences tau phosphorylation, release, and its subsequent effects on neuronal function. Our findings revealed that *P. aeruginosa* infection induces tau hyperphosphorylation and its release from lung endothelial cells, producing cytopathic tau species that contribute to neurotoxic effects. These released cytopathic tau species serve as seeds for tau aggregation in neurons and inhibit long-term potentiation (LTP), a surrogate *in vitro* measurement for learning and memory. *In vivo* experiments in wild-type mice demonstrated that lung infection with *P. aeruginosa* led to a significant reduction in LTP, a result not observed in tau knockout mice, suggesting that tau is integral to the lung infection-dependent brain damage observed in pneumonia. Given that tau mutations, hyperphosphorylation, and aggregation are hallmarks of several tauopathies, our study aimed to determine whether targeting tau expression in the lung endothelium could mitigate pneumonia-associated cognitive impairments.

Preliminary results indicated that an antisense oligonucleotide targeting the 3' untranslated region (UTR) of tau effectively reduced tau expression in lung endothelial cells *in vitro* in a dose- and time-dependent manner. *In vivo*, systemic administration of this antisense oligonucleotide via tail vein injection led to a reduction in lung endothelial tau expression, with no observed effect on brain tau levels. Importantly, this treatment did not impair LTP in uninfected control mice, suggesting that the antisense oligonucleotide selectively targets lung tau without disrupting normal brain tau homeostasis.

We are currently investigating whether antisense oligonucleotide treatment reduces the release of cytopathic tau species from the lung and alleviates LTP impairment in mice following

pneumonia. These findings could provide critical insights into the therapeutic potential of targeting lung-derived tau to mitigate the cognitive deficits associated with pneumonia and other pulmonary infections, which could have broader implications for tau- or pneumonia-related neurodegenerative diseases.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

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Topic: D.03. Stroke

Support: JSPS KAKENHI Grant Number JP23K10906

Title: Kynurenine 3-monooxygenase mediates stress vulnerability following intracerebral hemorrhage in mice

Authors: *M. OHNISHI, A. MACHIDA, K. BANSHOYA;
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Abstract: The prognosis of intracerebral hemorrhage (ICH) remains poor. Decreased motivation, often accompanying the sequelae of ICH, hinders rehabilitation and delays recovery. Our previous research demonstrated that alterations in the kynurenine pathway following ICH lead to an increased ratio of quinolinic acid (QUIN) to kynurenic acid (KYNA), thereby contributing to neuronal damage. QUIN functions as an N-methyl-D-aspartate (NMDA) receptor agonist, whereas KYNA serves as an NMDA receptor antagonist. Therefore, the QUIN/KYNA ratio can be considered an indicator of NMDA receptor activation. Ketamine, an NMDA receptor antagonist, is known to exert antidepressant effects. In this study, we focused on the role of kynurenine 3-monooxygenase (KMO), an enzyme situated at the metabolic branch point directing kynurenine toward either QUIN or KYNA. An *in vivo* ICH model was employed by microinjecting collagenase into the striatum of male mice. Depression-like behavior was assessed using the sucrose preference test, which was selected to minimize the confounding effects of motor dysfunction due to ICH. After 14 days of repeated tail suspension stress, ICH mice exhibited more pronounced depression-like behavior than controls which were injected with saline. This behavioral impairment was ameliorated by Ro61-8048, a KMO inhibitor. Previous studies have shown that the antidepressant effect of ketamine is abolished by NBQX, an AMPA-type glutamate receptor antagonist. Similarly, NBQX diminished the antidepressant effect of Ro61-8048. QUIN levels were notably elevated in the prefrontal cortex (PFC) following ICH, but this increase was suppressed by Ro61-8048, indicating that QUIN levels reflect KMO enzyme activity. The PFC also undergoes phosphorylation of mTOR, which has been identified

as a key component of the antidepressant mechanism of (*S*)-ketamine, an enantiomer with more potent NMDA receptor antagonistic properties. In the present study, Ro61-8048 reversed the ICH-induced reduction in mTOR phosphorylation within the PFC, but this effect was canceled by NBQX. Additionally, results from the conditioned place preference test showed that Ro61-8048 did not produce the addictive side effects associated with ketamine. This finding may be attributed to the distinct mechanism of Ro61-8048, which regulates elevated QUIN levels via KMO inhibition, rather than directly blocking NMDA receptors as ketamine does. In conclusion, KMO inhibition alleviates post-ICH motivational deficits without producing dependency, suggesting its therapeutic promise.

Disclosures: M. Ohnishi: None. A. Machida: None. K. Banshoya: None.

Poster

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Swedish Brain Foundation
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Title: Neuronal hyperconnectivity after stroke is maladaptive and is preventable by intranasal complement C3a

Authors: *M. PEKNA¹, A. STOKOWSKA¹, M. ASWENDT², R. KLASSEN³, P. ABÁFFY³, M. MODE¹, M. KUBISTA³, L. VALIHRACH³, M. HOEHN⁴, M. PEKNY¹;

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Abstract: Despite substantial progress in acute treatment and care, stroke remains a major cause of long-term disability. To reduce neurological impairment after stroke, better understanding of the mechanisms that drive stroke recovery and novel interventions that would interact with these mechanisms are needed. Neural plasticity responses in the ipsilesional hemisphere are involved in recovery of function after stroke. The stroke-induced responses in the contralesional cortex, how these responses change over time, their role in functional recovery and the effects of recovery-enhancing interventions in the contralesional cortex are incompletely understood. Using photothrombotic stroke model in mice, we show that ischemic stroke triggers pronounced changes in gene expression in the contralesional cortex that persist for at least 2 months. The

most upregulated genes are those involved in inflammation, glial reactivity and synaptic transmission. Intranasal treatment with C3a starting on post-stroke day (P) 7, previously shown to accelerate recovery of motor function, increased the expression of *Kcnj6*, *Grik2*, *Sv2c*, *Nptx2*, *Prkcd* and *Ctss* and reduced the expression of *Lgals3*, *C1qa*, *Serpina3n*, *Lyz2*, *Gpnmb*, *Kcnj10*, *Cpeb2*, *Lmtk3*, *Trpc3* and *Manf*. To investigate the functional correlates of these findings, we assessed stroke-induced changes in functional connectivity using resting-state functional MRI. We found prominent functional hypoconnectivity in the acute phase (P7) that was most pronounced in brain regions directly affected by stroke. In contrast, in the subacute phase (P28-56), we found prominent brain-wide functional hyperconnectivity that negatively correlated with recovery of motor function. This increase in brain connectivity was absent in C3a-treated mice. We conclude that stroke-induced gene dysregulation in the contralesional cortex comprises genes involved in inflammation and regulation of the balance between excitation and inhibition and coincides with brain-wide hyperconnectivity. Recovery-enhancing C3a treatment modulates transcriptomic responses in the contralesional cortex and prevents post-stroke hyperconnectivity, suggesting maladaptive nature of hyperconnectivity in the subacute phase after stroke.

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Poster

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Topic: D.03. Stroke

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Title: Complement C3a receptor (C3aR) Regulates Inflammatory Signaling in Murine Stroke Pathophysiology

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Abstract: Background: Stroke is the third leading cause of death and disability worldwide. Previously, we have demonstrated that endothelial C3a receptor (C3aR) is expressed in murine stroke brain as well as in brain endothelial cells subjected to oxygen-glucose

deprivation (OGD). A growing body of work suggests that activation of the C3a/C3aR axis in endothelial cells plays a prominent role in post-stroke neurovascular dysfunction. Hence, we hypothesized that endothelial C3aR signaling has prominent role in post-stroke neurovascular dysfunction and it is critical to investigate the signaling pathways. **Hypothesis:** Endothelial C3aR deletion is neuroprotective in stroke. **Methods:** We used a *loxP-cre* approach (C3aR^{Flox/Flox} x Cdh5^{Cre}) to generate conditional C3aR endothelial sufficient or deficient mice (C3aR^{Endo+/+} or C3aR^{Endo-/-}) of both sexes (3-4 months old) which were subjected to photothrombotic stroke. WT littermates were used as controls. We assessed cerebral blood flow (CBF), Infarct volume, BBB integrity, and myeloid cell infiltration. Neurobehavioral functions were tested at 3 and 21 days. P<0.05 was considered statistically significant. **Results:** Photothrombotic (PT) MCA occlusion resulted in decreased CBF in C3aR^{Endo+/+} mice compared with sham animals. Infarct volume was decreased and CBF was increased 72 h post PT in C3aR^{Endo-/-} vs C3aR^{Endo+/+} mice. C3aR^{Endo-/-} mice showed improved behavior and cognitive function compared to C3aR^{Endo+/+}. Finally, we found reduced myeloid cell/neutrophil infiltration that resulted improved BBB integrity in C3aR^{Endo-/-} vs C3aR^{Endo+/+} mice.

Conclusion: Overall, our data suggest that endothelial deletion of C3aR is neuroprotective. Further work is warranted to explore underlying mechanisms of C3aR mediated brain injury in stroke.

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Poster

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Program #/Poster #: PSTR275.04/H16

Topic: D.03. Stroke

Support: TOURO/NYMC Bridge Support

Title: Ferroptosis and Neuroinflammation during Postnatal Hippocampus formation after intraventricular hemorrhage

Authors: *G. VINUKONDA¹, M. SUNSHINE⁶, V. CASTRO DIAZ⁷, F. HU², R. SACHIDANANDAM³, J. VELISKOVA⁴, S. SUBBIAN⁸, L. VELISEK⁵, E. F. LA GAMMA⁹; ¹Pediatrics-Newborn Med. and Cell biology&Anatomy, ²Pediatrics, ³Genomic Core- Dept. of Pathology, ⁴Dept Cell biology and Anat., ⁵Cell biology and Anat., New York Med. Col., Valhalla, NY; ⁶Neonatology, Maria Fareri Children's Hospital, Westchester Med. Ctr., Valhalla, NY; ⁷Neonatology, Maria Fareri Children's Hospital, Westchester Med. Ctr., Yonkers, NY; ⁸Dept. of Med., New Jersey Med. School, Rutgers Univ., Newark, NJ; ⁹Pediatrics - Div. of Newborn Medicine, Biochem & Molec Bio, Westchester Med. Center, New York Med. Col., Valhalla, NY

Abstract: Intraventricular hemorrhage (IVH) is a common complication of prematurity birth. IVH results in reduced neurogenesis, cerebral palsy, learning and memory disabilities. After IVH, hematoma and debris can concurrently occur in the hippocampus due to i) its position in the inferior horn of the lateral ventricle and ii) its immature blood vessels. Emerging research suggests that ferroptosis, a novel form of cell death contributes to the secondary brain injury following IVH. We hypothesize that IVH results in iron accumulation from lysed red blood cells (RBCs) that will induce ferroptosis, neuroinflammation, and interrupt hippocampal maturation. Sulforaphane (SFN; Nrf2, transcription factor activator) and deferoxamine (DFN; iron chelator) treatments may reduce ferroptosis and inflammation by mitigating dysregulated ferroptosis-associated network gene transcription. We used our rabbit model of glycerol-induced IVH with ultrasound confirmation and grading. After IVH, the pups in the experimental groups were assessed for hippocampal hematoma, iron accumulation histologically using coronal serial sections by H&E, immunofluorescence imaging evaluation and then quantified iron and free hemoglobin in CSF and tissue lysates. Treated IVH groups received SFN-DFN (SFN 25mg/kg IP; DFN 50mg/kg SQ), compared with IVH saline or no IVH controls. Hippocampal tissue iron accumulation increased two-fold compared to controls ($p < 0.01$); whereas combined drug treatment decreased $Fe^{2+/3+}$ to control levels ($p < 0.05$, $n=5/grp$ for both). After IVH, iron accumulation was found in the dentate gyrus (DG), hilus and CA3 subregions ($n=5/grp$). Cell density studies for inflammation/apoptosis showed 60% reduction of Iba-1+ microglia and 53% reduction of TUNEL-positive apoptotic cells after SFN-DFN treatment ($p < 0.05$ for both, $n=5/grp$). Together, after IVH, pups showed increased hippocampal free hemoglobin, $Fe^{2+/3+}$, microglia, and apoptosis, where effects were attenuated after SFN-DFN treatment. To assess iron mediated gene expression changes associated with ferroptosis & inflammation, we performed RNA-seq analysis using dissected hippocampus tissue. We observed significantly increased expression of ferritin light & heavy chain (FTL, FTH), and hemoxygenase-1 (HO1) as well as other transcriptome products linked to ferroptosis in IVH. After SFN-DFN treatment, pups show decreased levels of FTL, FTH and HO1 compared with IVH ($p < 0.05$, $n=4/grp$). This study is the first to observe ferroptosis in the hippocampus/DG region after IVH and significant reduction of IVH induced pathology following SFN-DFN treatment.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

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Topic: D.03. Stroke

Support: NIH R01NS115759
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Title: Examining Synaptic Connections after Stroke and anti-Nogo-A Antibody Treatment

Authors: *S.-Y. TSAI¹, D. NGUYEN^{1,3}, B. E. POWERS^{4,2}, S. T. TON¹, H. HIOKI⁵, R. P. NOCKELS⁶, G. L. KARTJE^{1,4};

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Abstract: Neuronal plasticity plays important roles in functional improvement after stroke. We have reported that enhancement of axonal plasticity and dendritic complexity/spine density in layer V pyramidal neurons was associated with better sensorimotor function after stroke and anti-Nogo-A antibody (Ab) treatment. It is also well known that both inhibitory and excitatory synapses influence functional recovery after brain lesions including stroke. However, there are no studies examining how these synaptic inputs are changed by anti-Nogo-A Ab treatment post-stroke. Therefore, we proposed this study to examine both excitatory and inhibitory synaptic connections to layer V pyramidal neurons in the contralesional sensorimotor cortex after stroke and anti-Nogo-A Ab treatment. Adult male Long-Evans rats were first trained to perform the skilled ladder rung walking test followed by middle cerebral artery occlusion (MCAO) surgery. At one week after stroke, rats were randomly assigned to receive no Ab, control Ab or anti-Nogo-A Ab treatments. Behavioral testing was performed before stroke (baseline), one week after stroke (before treatment) and nine weeks after stroke. At the end of behavioral testing, adeno-associated virus (AAV) encoding EGFP directed to plasma membrane was injected into the contralesional cortical forelimb area to label the dendrites of layer V pyramidal neurons. Rats were later perfused and brains processed using immunofluorescent histochemistry. VGlut1 or VGlut2 paired with PSD95 (excitatory synapses) and VGAT paired with gephyrin (inhibitory synapses) within the EGFP labeled dendrites of layer V pyramidal neurons were analyzed by confocal microscopy. Behavioral data showed that rats receiving anti-Nogo-A Ab treatment significantly improved in the skilled ladder walking test as compared to control rats. Analysis is still undergoing to determine whether anti-Nogo-A Ab treatment altered either excitatory or inhibitory synaptic connections to layer V pyramidal neurons in the forelimb cortex opposite the stroke side. Our study will provide further understanding on the effect of Nogo-A blockade on neuronal connectivity after stroke and may lead to the development of new approaches for post-stroke care.

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Poster

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Topic: D.03. Stroke

Support: AHA 24SCEFIA1255866
AHA 24TPA1300751

Title: The role of fatty acid synthetases in modulating functional outcomes in sickle cell disease

Authors: T. YUAN, Y. ZHOU, R. H.-C. LEE, *C. Y.-C. WU;
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Abstract: Sickle cell disease (SCD) is the most common severe monogenic disorder affecting 300,000 individuals worldwide annually. The main challenges in the treatment of SCD are the sickle-shaped red blood cells that tend to aggregate along the vessel walls leading to inflammation and vascular occlusions. Therefore, patients with SCD suffer from ischemic stroke, transient ischemic attacks, and multiple organ dysfunction. One of the major hallmarks of SCD is the low levels of omega-3 fatty acids (ω 3-FA) such as docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) in the blood. Thus, identifying the regulatory elements that control ω 3-FA metabolism holds high therapeutic potential. As such, biosynthesis of ω 3-FA through long-chain acyl-CoA synthetases (ACSLs) is essential for cell member function and survival. In fact, it has been reported that downregulation of ACSL can cause ω 3-FA deficiency, as a result of cardiovascular diseases. The impact of ACSL on the central nervous system in pathological conditions (e.g., SCD), however, remains largely unknown. The goal of the study is to determine the role of ACSLs in brain circulation and neurological outcomes in SCD. We discovered in our pilot study that only ACSL6 expression is reduced in brain regions that are susceptible to ischemia (e.g., hippocampus and cortex). We hypothesize that ACSL6 in the brain is critical to combat SCD-induced hypoperfusion, mitochondrial dysfunction, neuroinflammation, and neurological deficits. To answer our hypothesis, we will upregulate ACSL6 and use pharmacological and cell type-specific genetic approaches in mouse model of SCD. Completion of the proposed study will reveal the fundamental roles of ACSL6 in neuronal survival/death in cerebral ischemia.

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Poster

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Topic: D.03. Stroke

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Title: Developing Small Molecule Therapeutics for the Treatment of Global Cerebral Ischemia through TREM1 Inhibition

Authors: *P. L. TIWARI¹, A. DHULER², H. KIM¹, R. URQUHART¹, V. DUNCAN³, P. ABEL¹, G. JADHAV¹, J.-Y. HWANG¹;

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Abstract: Global cerebral ischemia is a severe form of stroke often caused by cardiac arrest, which affects 200,000 Americans each year, leading to cognitive deficits or death. While emergency treatments of cardiac arrest focus on restoring cardiac function and blood flow, an effective treatment for neurodegeneration and cognitive deficits remains an unmet medical need. TREM1 is a membrane immune receptor expressed on the surface of myeloid-derived cells where it magnifies the innate pro-inflammatory immune response. Our recent studies showed that global ischemia activates TREM1 and neuroinflammation in the hippocampal CA1 in rats. Additionally, TREM1 inhibition by the peptide inhibitor LR12 demonstrated neuroprotection against ischemic insult. Therefore, we hypothesized that TREM1 inhibition could be a new therapeutic strategy. Current inhibitors, however, have limitations as potential drug candidates due to their poor cell permeation and short half-life. To address these limitations and develop small molecule TREM1 inhibitors, we first identified an *N*⁴-(amino-substituted)-*N*-substituted-benzene sulfonamide pharmacophore as a hit molecule from molecular docking of 80K molecules to the hTREM1 (PDB: 1SMO) crystal structure. Surface Plasmon resonance (SPR) analysis confirmed hit molecule's affinity to TREM1 with a K_d of 14.3 μM, but displayed poor solubility. To address this, we synthesized para-fluoro analog of hit molecule (GJ079), which showed enhanced solubility and improved affinity to TREM1 (K_d = 4.8 nM). Thus, we further modified hit molecule's structure and synthesized 44 additional analogs to enhance their pharmacological and pharmacokinetic properties through five series: Series 1 required an additional aromatic-acetamido substitution on a thiazolyl group containing nitrogen of 4-amino-N-(thiazol-2-yl) benzenesulfonamide; Series 2 involved an aromatic-acetamido substitution on the *N*⁴- aromatic amino group; Series 3 featured ortho, meta and para fluoro substitution on the end phenyl ring of hit molecule; Series 4 involved the replacement of the 2-thiazolyl ring with another hetero aromatic ring in the hit molecule; and Series 5 with aromatic-amido substitution on the *N*⁴- aromatic amino group. To evaluate therapeutic potential of these compounds, we first tested GJ079 under inflammatory and ischemic conditions *in vitro* and in a rat model of global ischemia *in vivo*. GJ079 reduced pro-inflammatory cytokines, attenuated neuroinflammation and prevented neuronal death, indicating its neuroprotective effect. We are currently validating 44 additional analogs *in vitro*, with lead molecules to be tested *in vivo*.

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Poster

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Support: NSTC 113AO15B (National Science and Technology Council, Technology Management Bureau (TMB), Taiwan)
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Title: Erinacine A treatment in the post-stroke cognitive impairment — a preclinical study

Authors: *T.-L. TSAI¹, P.-C. HSU², L.-Y. LEE³, C.-C. CHEN³, Y.-C. KAO⁴, Y.-H. Y. LEE⁵;
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Abstract: Post-stroke cognitive impairment results from neurodegeneration in stroke patients, causing significant physical and mental burdens due to severe brain damage. Current neuroprotective strategies for stroke primarily focus on reducing brain damage during the acute phase. However, approaches to improve functional outcomes and address neurodegeneration during the chronic phase remain limited. Erinacine A (EA), an active component of *Hericium erinaceus* (HE), has been shown to preserve astrocytic function and maintain glutamate homeostasis, mitigating excitotoxicity during the acute phase of ischemic brain injury. In this study, we further investigated the effect of EA and HE ethanol extract (HEet) effects on survival, neurological deficits, brain structure (MRI), cognition, and histology after transient hypoxia-ischemia (tHI)-induced stroke. Mice received daily intranasal EA (nEA) or HEet (nHEet) for 14 days. MRI, cognitive tests, and histology were conducted on day 28. Our results show that nEA significantly reduced acute-phase mortality and deficits. MRI showed day 3 brain swelling correlated with day 28 infarct atrophy. Although spontaneous motor activity recovered in the chronic phase as assessed by the open field test (OFT), persistent neurocognitive deficits were observed by the novel object recognition (NOR) test. Daily nEA improved NOR performance by day 28, regardless of day 3 deficits. Daily administration of nEA significantly improved performance in the NOR test on day 28, independent of the severity of neurological deficits at day 3 post-tHI. Noteworthy, histological examination showed that EA did not prevent stroke-induced neuronal damage but significantly enhanced functional astrogliosis. In conclusion, daily intranasal administration of EA reduces post-stroke mortality, alleviates neurological deficits, and improves cognitive outcomes. These effects may involve the chronic regulation of astrocytic function. Our findings highlight EA's therapeutic potential for enhancing survival in the acute phase and supporting cognitive recovery in the chronic phase, ultimately improving stroke patients' quality of life.

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Poster

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Topic: D.03. Stroke

Support: NIH Grant NS090904
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Title: Bivalent GPCR-sequence-based peptides reduce neuronal apoptosis and ischemic stroke injury in mouse models

Authors: *B. XIANG¹, A. P. SAGARE², Y. WANG², Y. KIM², M. ZHANG², R. RUST², J. A. FERNÁNDEZ³, B. V. ZLOKOVIC², W. MACK², J. GRIFFIN³, **K. KISLER**²;
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Abstract: Plasma-derived activated protein C (APC), a serine protease, demonstrated anti-inflammatory, antithrombotic and neuroprotective effects in murine ischemic stroke models. Studies showed that APC's cell signaling requires protease-activated receptor (PAR) PAR1 and PAR3, and APC's PAR cleavages generate peptides that are capable of acting as a biased agonist for G-protein coupled receptor (GPCR) cytoprotective signaling resulting in anti-inflammatory and anti-apoptotic activities and endothelial barrier stabilization. Peptides derived from the extracellular N-terminal tails of human and murine PAR1 or PAR3 can also initiate cell signaling that mimics APC's cell signaling and cytoprotective effects. Recently, we showed that the most potent human PAR-derived peptide, 34-residue "G10 peptide" comprising PAR1 residues 47-55 covalently attached by a 10-glycine linker to PAR3 residues 51-65 is an orthosteric/allosteric bivalent GPCR agonist that potently mimics APC's anti-inflammatory activity and endothelial barrier stabilization activity.

We hypothesized that the G10 peptide could mimic APC's ability to reduce N-methyl-D-aspartate (NMDA)-induced neuronal apoptosis and reduce brain injury due to ischemic stroke. Our data showed that the human G10 peptide inhibited N-methyl-D-aspartate (NMDA)-induced neuronal apoptosis in neuronal cell cultures. Compared with the control group, the G10 peptide reduced the number of TUNEL-positive and caspase-3-positive cells by 47% and 53%, respectively. Furthermore, in vivo studies using a transient proximal middle cerebral artery occlusion (tMCAO) murine stroke model showed that the murine G10 peptide, administered intravenously (1 mg/kg) 10 minutes and 4 hours after tMCAO in wild-type C57BL6 mice, reduced the mean values of damage volume, edema volume, and infarct volume by approximately 45-50% compared to vehicle.

In summary, bivalent PAR-derived 34-residue G10 peptides function as cytoprotective agonists that mimic APC's neuronal anti-apoptotic protective activity in neuronal cultures and neuroprotective activity in a murine tMCAO stroke model. These data support the rationale for pursuing further studies of G10 peptides and G10 peptide analogs for potential therapeutic value for multiple neuropathologies.

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Poster

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Program #/Poster #: PSTR275.10/I6

Topic: D.03. Stroke

Title: Pharmacological Inhibition of Drp1 Protects Against Cerebral Ischemia/Reperfusion-Induced Neuronal Damage in Zebrafish

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Abstract: Cerebral ischemia remains a leading cause of mortality and long-term disability worldwide. It results from an arterial blockage that restricts the supply of oxygenated blood to the brain, leading to hypoxia and neuronal damage. Current therapeutic interventions, such as recombinant tissue plasminogen activator (rTPA) and mechanical thrombectomy, aim to rapidly restore cerebral blood flow. However, this reoxygenation process paradoxically increases the risk of cerebral ischemia/reperfusion (I/R) injury due to the generation of reactive oxygen species and secondary cellular damage. In this study, we evaluated the neuroprotective potential of a Dynamin-related protein 1 (Drp1) inhibitor in a zebrafish model of cerebral I/R injury using 6-day post-fertilization (dpf) larvae. Drp1 is a key mediator of mitochondrial fission and has been implicated in mitochondrial dysfunction and neuronal apoptosis following ischemic injury. Our results demonstrated that Drp1 inhibition significantly mitigates neuronal damage induced by I/R in zebrafish larvae. The treatment reduced oxidative stress levels, preserved mitochondrial integrity, and suppressed apoptotic cell death. A marked downregulation in Drp1 expression was also observed, confirming the compound's target-specific effect. These findings suggest that Drp1 inhibition may be a promising therapeutic intervention for reducing neuronal injury and improving outcomes following cerebral I/R. Further studies in mammalian models are warranted to validate its translational potential.

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Poster

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Topic: D.03. Stroke

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Department of Neurology, HFH

Title: Anti-inflammatory role of perfluorocarbon in a mouse model of subarachnoid hemorrhage

Authors: M. AHMED, M. ABDULLAH, S. RAMESH SHARMA, Z. KHAN, S. GIRI, *A. S. AHMAD;
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Abstract: Inflammation and oxidative stress are key drivers of subarachnoid hemorrhage (SAH) pathology, leading to brain injury and vasospasm. This study aimed to evaluate the effects of a lipid emulsion-based oxygen carrier, perfluorocarbon-Oxygent (PFC-Oxygent), as a promising therapeutic agent to limit inflammation and oxidative stress following SAH. PFC-Oxygent consists of lipid emulsion microparticles with high flexibility. Unlike red blood cells, the ultra-small size of these particles allows them to flow even with minimal residual plasma trickle and efficiently deliver oxygen, thereby aiding in the prevention of tissue damage. Adult C57BL/6 wild-type mice were subjected to the endoperforation model of SAH, followed by intravenous injection of PFC-Oxygent or saline (9ml/kg) at 5 hours post-SAH. At 48 hours post-SAH, the mice were evaluated for functional outcomes. Subsequently, the mice were transcardially perfused, and the brains were harvested for immunohistochemistry and biochemical analyses. SAH induced significant behavioral impairments and triggered NLRP3 inflammasome activation and upregulation of oxidative stress markers at 48h post-SAH. Treatment with PFC-Oxygent, however, attenuated these functional impairments. Notably, the SAH group treated with PFC-Oxygent, showed a significant reduction in NLRP3 inflammasome components and oxidative stress markers. These findings demonstrate that PFC-Oxygent treatment improves functional outcomes and reduces NLRP3 inflammasome-mediated inflammation and oxidative stress following SAH. Overall, these findings underscore the therapeutic potential of PFC-Oxygent treatment in mitigating SAH severity.

Disclosures: M. Ahmed: None. M. Abdullah: None. S. Ramesh Sharma: None. Z. Khan: None. S. Giri: None. A.S. Ahmad: None.

Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.12/I8

Topic: D.03. Stroke

Title: Metabolic reprogramming of neutrophils in CKD impairs stroke recovery

Authors: *H. KAUR¹, N. PANDEY¹, L. CHANDALURI¹, N. SHAABAN¹, E. KIDDER¹, A. MARTINEZ¹, V. SINGH⁴, J. HARKRIDER¹, S. ANAND¹, R. PANDIT², A. ORR¹, A. YURDAGUL², O. ROM^{1,2}, R. MOHANDAS⁵, B. SACHDEVA³, N. DHANESHA¹;

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Abstract: Chronic kidney disease (CKD) is a significant risk factor for stroke and is associated with poorer post-stroke outcomes, including increased neurological deterioration, higher in-hospital mortality, and impaired functional recovery. However, the precise mechanisms by which CKD exacerbates stroke severity remain largely elusive. Emerging evidence indicates that CKD induces systemic inflammation and immune dysregulation, particularly altering neutrophil functions, yet the metabolic underpinnings of this neutrophil dysfunction and its contribution to stroke outcome are unclear. Importantly, neutrophils fuel effective inflammatory responses and oxidative stress through glycolysis. We first investigated transcriptomics, glycolysis status, neutrophil activation markers in individuals with CKD. Unbiased RNA sequencing of circulating neutrophils from CKD patients and age-matched healthy controls revealed a distinct shift toward glycolytic metabolism, highlighted by the upregulation of phosphoglycerate kinase 1 (PGK1), a rate-limiting glycolytic enzyme. Functional profiling using Seahorse extracellular flux analysis confirmed increased glycolytic activity. Furthermore, flow cytometry demonstrated elevated mitochondrial reactive oxygen species (ROS) and glucose uptake (2-NBDG) in CKD neutrophils, accompanied by increased PGK1 expression at both mRNA and protein levels. These metabolic alterations were associated with enhanced pro-inflammatory responses, including augmented neutrophil extracellular trap (NET) formation and cytokine release. To determine the *in vivo* relevance of neutrophil glycolysis, we utilized neutrophil-specific PGK1-deficient (*PGK1^{fl/fl}Mrp8cre^{+/-}*) mice and littermate controls (*PGK1^{fl/fl}Mrp8cre^{-/-}*) in a model of CKD (28-day adenine administration) followed by 30-minute middle cerebral artery occlusion (MCAo). Sensorimotor recovery, assessed over 4 weeks, was significantly improved in neutrophil-specific PGK1-deficient mice with CKD compared to their control counterparts. These findings identify PGK1-driven glycolysis as a critical mediator of neutrophil activation in CKD and a novel contributor to worsened stroke outcomes. Targeting PGK1-dependent metabolic reprogramming in neutrophils may therefore represent a promising therapeutic approach to mitigate inflammation and improve cerebrovascular recovery in CKD patients.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.13/19

Topic: D.03. Stroke

Title: Reperfusion injury is exacerbated by the loss of mitoNEET in mice

Authors: *G. N. WILSON¹, W. J. GELDENHUYS², K. SMITH¹, P. BHANUSHALI¹;

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Abstract: Mitochondria play a central role in neuronal survival and death following brain injury. They act as key regulators of energy metabolism, calcium homeostasis, and apoptotic signaling. Mitochondrial dysfunction and oxidative stress are critical contributors to ischemic brain injury, yet the role of specific mitochondrial targets in stroke pathology and reperfusion injury remains underexplored. Previous work has demonstrated that the outer mitochondrial membrane protein, mitoNEET, plays a crucial role in mitochondrial bioenergetics and iron homeostasis. MitoNEET (CISD1) is an iron-sulfur cluster-containing protein that acts as a redox sensor; when stabilized it can limit mitochondrial iron uptake thereby maintaining redox balance, and reducing oxidative stress, lipid peroxidation, and cell death. We have also shown that genetic ablation of mitoNEET is also associated with impaired cognitive outcomes and increased neuroinflammation in the absence of injury. In the present study, we investigated the role of mitoNEET (CISD1) in ischemic stroke and reperfusion injury using a murine model of transient middle cerebral artery occlusion (tMCAO) combined with genetic knockout of CISD1. Wild-type (CISD1^{+/+}) and mitoNEET knockout (CISD1^{-/-}) mice underwent 60 minutes of MCAO followed by 24 hours of reperfusion prior to euthanasia and tissue collection. Using immunohistological and immunofluorescent techniques, we assessed infarct volume, glial and astrocytic response, oxidative stress, and blood-brain barrier permeability. Compared to wild-type controls, mitoNEET knockout mice exhibited significantly larger infarct volumes and increased expression of Iba1 and GFAP, suggesting heightened neuroinflammation and impaired neuroprotection. Additionally, knockout animals exhibited higher immunoglobulin (IgG) staining throughout the brain, suggesting a role for mitoNEET in maintenance of the blood brain barrier. Our findings highlight the role of mitoNEET in regulating glial reactivity, blood brain barrier permeability, and infarct progression following stroke. Further, they underscore the therapeutic potential of targeting mitoNEET to improve post-stroke outcomes.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.15/Web Only

Topic: D.03. Stroke

Support: Programa de Concurrencias Financieras para la Investigación y Atención de la Vinculación

Title: Ketamine Promotes Plasticity-Related Protein Changes in Rats with Permanent Middle Cerebral Artery Occlusion

Authors: *N. I. MARTINEZ TORRES;

Univ. de Guadalajara/Centro de Investigación Biomédica de Occidente, Guadalajara, Mexico

Abstract: INTRODUCTION

Ketamine (KET) is a drug that has been used as an anesthetic for many years. However, in the last two decades, new properties have been discovered that make it useful in treating various psychiatric and neurological disorders. Ketamine has shown effectiveness in preclinical models and patients with conditions such as depression, Parkinson's disease, and obsessive-compulsive disorder, among others.

MATERIAL AND METHODSIn the present study, we evaluated the effect of ketamine administration following permanent middle cerebral artery occlusion (pMCAO) in rats, a model of stroke. We used 27 male Sprague-Dawley rats, each weighing ~220 g. The rats were randomly assigned to one of three groups (Sham, pMCAO-Saline, and pMCAO-KET), with nine animals in each group. pMCAO was performed, and BDNF and AMPA protein presence were characterized through immunohistochemistry, and the infarct area was measured.

CONCLUSIONSOur results suggest that ketamine promotes the reduction of the infarct area and alleviates neuropathological features associated with astrocyte reactivity and microglial infiltration. Also, KET increased the presence of AMPA and BDNF proteins, key components of neural plasticity that may help recovery after a stroke. These findings suggest that ketamine may be a promising strategy for stroke treatment, with neurobiological changes related to plasticity underlying its effects.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.16/I10

Topic: D.03. Stroke

Support: NIH 1R01NS138259
NIH R01NS142187

Title: Elovonoids are homeostatic lipid mediators that protect the brain after experimental ischemic stroke in rats

Authors: *L. S. BELAYEV¹, C. METON DE ALENCAR GADELHA VIEIRA², A. JACOTTE-SIMANCAS³, A. GENC³, G. GULSEN⁴, A. OBENAU⁵, J. X. JI⁶, P. MUKHERJEE³, N. G. BAZAN⁷;

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Abstract: Recently, we demonstrated that the intranasal (IN) delivery of very long-chain polyunsaturated fatty acid Elovonoid (ELV) precursors (FFA 32:6n-3 or 34:6n-3) protects the brain after an experimental stroke. ELVs improved neurological deficit, decreased T2WI lesion volume, and protected the blood-brain barrier. The purpose of this study was to determine the dose-response study. Sixty-six male (3 months old) SD rats underwent 2 hours of MCAo, treated with ELV precursors (34:6n-3 or 32:6n-3) (5, 10, or 15 µg per nostril; totaling 10, 20, or 30 µg per animal) or vehicle, delivered IN at 1, 24, and 48 hours after the onset of MCAo. A composite neurological battery (0 = normal and 12 = maximal deficit) was assessed on days 1, 2, 3, 7, and 14; then rats were perfused with 4% paraformaldehyde, their brains were removed, and *ex vivo* MRI analysis of lesion volumes was performed. Rectal and cranial temperatures, plasma glucose, blood gases, hematocrit, and blood pressure showed no significant differences among all groups. There were no adverse behavioral side effects observed after ELV administration in the present study. Treatments with FFA-34:6 (20 and 30µg) improved total neurological scores on days 7 (25, 38%) and 14 (36, 49%), FFA-32:6 (10, 20 and 30µg) on day 7 (44, 49, 53%) and day 14 (44, 61, 61%, respectively), compared to vehicles. T2WI revealed that rats treated with vehicle had large cortical and subcortical infarctions. In contrast, both treatments reduced T2WI lesion volumes compared to vehicles, which were primarily located in the subcortical areas. In conclusion, these data suggest that IN-administered FFA 32:6 and 34:6 provide high-grade neurobehavioral recovery and decrease lesion volumes when administered 3 hours after MCAo. The neuroprotection observed here for the post-ischemic brain offers the basis to explore therapeutic translational avenues for ischemic stroke.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.17/I11

Topic: D.03. Stroke

Support: Italian Ministry of Health (RF-2021-12373484; CUP: C53C22001570001)
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Ricerca Corrente 2025 from Fondazione Policlinico Universitario “A. Gemelli” IRCCS

Title: Extracellular vesicles and transcranial direct current stimulation facilitate recovery after stroke by modulating inflammation and brain connectivity

Authors: C. FEROLETO¹, C. D'AMELIO², M. MOROTTI¹, S. BARBATI², I. PAOLETTI¹, F. D'ALELIO¹, C. CALIGIURI², C. CODAZZI², I. NIFO SARRAPOCHIELLO², F. NATALE², D. D. LI PUMA², F. FRASCA³, C. PAPPALETTERA³, L. NUCCI³, F. MIRAGLIA³, F. VECCHIO³, L. LEONE^{2,1}, S. FUSCO^{2,1}, *M. V. PODDA^{2,1}, C. GRASSI^{2,1};

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Abstract: Stroke occurs when the brain's blood supply is interrupted. This interruption triggers biological responses, including activation of resident immune cells like microglia and recruitment of astrocytes, which form a glial scar around the affected area. While these processes are essential for tissue repair, excessive neuroinflammation can lead to neuronal death and disrupt synaptic connections, impairing neuroplasticity in the peri-infarct region. Therefore, understanding and modulating these processes is essential to develop effective post-stroke therapies. In our study, we explored non-invasive strategies by independently assessing the effects of extracellular vesicles (EVs) derived from bone marrow mesenchymal stem cells and transcranial direct current stimulation (tDCS) in mice subjected to focal ischemia of the motor cortex (MC) induced by photothrombosis. EV-treated mice received a daily dose of 0.1×10^9 EVs, starting 48 h after stroke and continuing twice weekly for 4 weeks. The tDCS group underwent a 20-min anodal tDCS over the injured MC and a cathodal tDCS over the contralateral side once daily for 3 days, beginning 72 h post-stroke. Forelimb coordination and strength were assessed through behavioral tasks conducted the day before stroke, 48 or 72 h post-stroke prior to EV or tDCS treatment, respectively, and weekly for 4 weeks. Our findings revealed that both treatments significantly improved forelimb deficits, as supported by histological analyses showing reduced infarct volume and enhanced neuronal survival in the peri-infarct area. Notably, EVs decreased the glial scar area and thickness, as well as microglia density. The anti-inflammatory effects of EVs were confirmed using an *in vitro* stroke model, where microglia cell cultures were subjected to oxygen-glucose deprivation. Specifically, EVs reduced the pro-inflammatory marker CD86 expression and increased the expression of the anti-inflammatory marker CD206. Similarly, tDCS reduced microglial activation in the peri-infarct area and modulated neuroplasticity by enhancing long-term potentiation at layer II-III synapses. Analysis of EEG-like recordings indicated increased cortical connectivity, which positively correlated with motor function improvement in tDCS-treated stroke mice. In conclusion, our results demonstrate that both EV and tDCS treatments ameliorate motor deficits, with reduced inflammation, increased neuronal survival, and enhanced brain plasticity potentially contributing to these outcomes. These findings suggest the potential for further investigation into the combined effects of tDCS and EV-based therapy to improve stroke recovery.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: D.03. Stroke

Support: NIH Grant AA031465
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NIH Grant AA025744
NIH Grant AA026708
NIH Grant AA025744-01S1

Title: Targeting the Endothelial Autotaxin-LPA-LPP3 Axis Mitigates Blood-Brain Barrier Disruption and Mitochondrial Dysfunction After Ischemic Stroke

Authors: *M. PANCHATCHARAM¹, V. MANIKANDAN², S. MANIKANDAN², H. SUN³, S. MIRIYALA²;

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Abstract: Disruption of the blood-brain barrier (BBB) and mitochondrial dysfunction are major contributors to ischemic stroke pathology, particularly during reperfusion. Autotaxin (ATX), an enzyme that generates lysophosphatidic acid (LPA), and lipid phosphate phosphatase 3 (LPP3), which degrades LPA and terminates its signaling, play opposing roles in regulating cerebrovascular integrity. However, the precise contribution of the endothelial ATX-LPA-LPP3 axis to stroke outcomes remains poorly understood. Using a mouse model of transient middle cerebral artery occlusion (tMCAO) with 24-hour reperfusion, we observed a significant upregulation of ATX mRNA and enzymatic activity in the ischemia-reperfusion (I/R) group. This was accompanied by elevated LPA levels in plasma and brain tissue, increased BBB permeability, loss of junctional proteins in brain microvascular endothelial cells (BMECs), and impaired mitochondrial bioenergetics, including reductions in basal oxygen consumption, ATP production, and spare respiratory capacity. Inhibition of ATX (HA130, PF8380) or LPA receptors (BrP-LPA, Ki16425, AM095) significantly reversed these effects, restoring BBB integrity and mitochondrial function. To directly assess the endothelial role of ATX, we generated inducible endothelial-specific ATX knockout mice (ERT2 ATX^{-/-}). Compared to control I/R mice, ERT2 ATX^{-/-} mice exhibited reduced infarct volumes, preserved cerebral blood flow (laser speckle imaging), and significantly decreased BBB permeability (Evans blue assay). These improvements correlated with lower ATX activity and LPA concentrations in the brain vasculature. These findings underscore the pathologic role of ATX-driven LPA signaling and highlight the modulatory role of LPP3 in limiting LPA activity at the vascular interface. Collectively, our results identify the endothelial ATX-LPA-LPP3 axis as a central regulator of vascular permeability and mitochondrial integrity after stroke. Targeting this pathway may offer a novel therapeutic strategy to protect the BBB and improve functional outcomes during reperfusion therapy.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.19/J1

Topic: D.03. Stroke

Support: AHA

Title: Immune checkpoint B7-H3 (CD276) mechanisms drive stroke pathogenesis in young and old age

Authors: *B. CHELLUBOINA¹, K.-K. MAK¹, A. BODA²;

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Abstract: Age-associated immunosenescence is a key determinant of age-related disease susceptibility. After stroke, restoring immunosurveillance in the aging brain requires precision strategies that mitigate chronic neuroinflammation and resolve immune dysregulation. CD276 (B7-H3) is an Immune checkpoint that contributes to inflammaging. CD276 levels are high in stroke patients. CD276-mediated mechanisms in stroke pathology and their involvement in adult and aged stroke prognosis have not yet been investigated. We investigated the effect of CD276 inhibition on post-stroke brain damage in young (3 months) and aged (18 months) mice. Adult and aged C57BL/6 mice were subjected to 60 min (adult)/35 min (aged) transient middle cerebral artery occlusion (MCAO) and injected (retroorbital) with either CD276 siRNA or a negative (non-targeting) siRNA at 5 min after reperfusion. Temporal levels of CD276 were measured up to 7 days of reperfusion. We assessed the brain damage (MRI; T2, diffusion, ADC mapping) at 1 day of reperfusion. Brain tissues were collected at 1 day of reperfusion, assessed for acute post-stroke changes in immune cell types and immune activation markers, involved in stroke pathology using the nCounter immune-focused panel, and the effect of CD276 inhibition was determined. CD276 was expressed in the brain as early as 6h in males and 12h in females (n=4/group) after 60 mins of transient MCAO. CD276 inhibition showed significantly less post-stroke brain damage than the age-matched negative siRNA (n=10/group; adults and n=5/group; aged). Immune-focused panel (n=3) showed CD276-mediated modulation in the brain after stroke. CD276 siRNA-treated mice showed improved motor functions compared to negative siRNA-treated MCAO-operated groups.

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Poster

PSTR275: Stroke Recovery: Pharmacological Approaches to Therapy

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR275.20/J2

Topic: D.03. Stroke

Title: Probiotic BM109 attenuate ischemic brain injury in high-choline diet rats via trimethylamine N-oxide (TMAO) modulation and neuroprotective effects

Authors: C. YOON¹, J. YOU², Y. KIM¹, B. KIM¹, J. SONG¹, H. JOO¹, J. JUNG¹, I. KWON¹, Y. KIM¹, J. HEO¹, S. YOON², *H. NAM¹;

¹Neurol., ²Microbiology and Immunol., Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Trimethylamine N-oxide (TMAO), a gut microbiota-derived metabolite, is increasingly recognized as a contributor to vascular and inflammatory diseases. While its cardiovascular risks are well reported, its impact on ischemic brain injury and the potential for microbiota-targeted interventions remain underexplored. We evaluated the dual protective effects of BM109, a probiotic strain designed to suppress TMAO production, in a rat model of transient middle cerebral artery occlusion (tMCAO) under high-choline dietary conditions. A total of 138 adult male Wistar rats (8 weeks old) were randomized into eight groups based on diet (standard chow or 1.2% choline-enriched), treatment (BM109 or vehicle) and surgical condition (tMCAO or sham). Rats were maintained on their assigned diet for 3 weeks. BM109 (5×10^{10} CFU/mL/day) or vehicle was administered orally during the final week. After 3 weeks, rats underwent 90-min tMCAO followed by 24-h reperfusion. Blood was collected via tail vein, and plasma TMAO levels were measured by liquid chromatograph-mass spectrometry (LC-MS). Infarct size was assessed by 2,3,5-triphenyltetrazolium chloride (TTC) staining, and neurological function was evaluated using Garcia, Longa, and modified neurological severity scores (mNSS). Immunofluorescence analyses and Western blotting examined microglial activation, oxidative stress and blood-brain barrier (BBB) integrity. In addition, we evaluated inflammatory receptor signaling involved in TMAO-induced injury. All procedures were performed in a blinded and randomized manner. Rats on a high-choline diet showed significantly elevated TMAO levels, and subjected to ischemia, exhibited greater infarct size compared to chow-fed controls ($30.01 \pm 4.67\%$ vs. $20.35 \pm 4.42\%$; $p = 0.0001$), and worsened mNSS scores (13.13 ± 1.09 vs. 10.31 ± 2.73 ; $p = 0.0025$). BM109 supplementation significantly reduced plasma TMAO and infarct size ($12.7 \pm 3.3\%$; $p < 0.0001$), and improved neurological outcomes (Garcia score: $p = 0.0003$; mNSS: $p = 0.0025$) compared to choline-fed controls. Interestingly, infarct size remained lower in BM109-treated rats despite comparable TMAO levels, suggesting the possibility of additional neuroprotective mechanisms beyond TMAO reduction. To explore this, we examined markers of blood—brain barrier (BBB) integrity, oxidative stress, and microglial activation. These findings support a dual mechanism by which BM109 mitigates ischemic brain injury—through TMAO-lowering and via TMAO-independent neuroprotective effects—highlighting the therapeutic potential of microbiota-based interventions in stroke.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR276.01/J3

Topic: D.04. Brain Injury and Trauma

Support: DOD Grant HT9425-23-1-1003

Title: Moderate Prefrontal Cortex Injury Remodels the Corticostriatal Circuit: A potential role for Microglia and Inflammation in Impulsivity

Authors: *E. CHU, S. JAMI, C. MAHONEY, J. MCCLOSKEY, M. MCLEOD, F. ZHAO, C. ASKWITH, J. PACKER, J. GODBOUT, T. SMITH, K. MARTENS, C. VONDER HAAR; Neurosci., Ohio State Univ., Columbus, OH

Abstract: Background: Traumatic brain injury (TBI) leads to a variety of chronic psychiatric symptoms, including increased impulsivity and attention deficits. The specific mechanisms are unclear. Prior research suggested abnormal neuroinflammation may cause circuit remodeling between the cortex and regions responsible for decision-making and motivation, such as the nucleus accumbens. Microglia are one of the crucial glial cells that regulate neuroinflammatory responses after TBI, therefore we hypothesized abnormal microglial responses and neuroinflammation would impact circuit remodeling and psychiatric symptoms after TBI.

Method: Young adult male and female Long-Evans rats underwent a model of moderate to severe prefrontal cortex brain injury. Acutely post-injury, nucleus accumbens nuclei were isolated for unbiased sequencing. Patch-clamp electrophysiology was performed to examine neuronal functioning, and optogenetics was used to determine the connectivity between frontal cortex and nucleus accumbens. To delineate microglia's role, we pharmacologically eliminated them post-injury and allowed for repopulation, then behavioral outcomes were assessed chronically. **Results:** Single nucleus sequencing revealed changes in the percentage of glutamergic and dopaminergic neurons post-injury, and reductions in numerous neurotransmitters and synaptogenesis pathways, except for increased GABAergic signaling. Furthermore, frontal TBI increased excitatory post-synaptic currents in the nucleus accumbens and a loss of excitatory input from the frontal cortex. TBI significantly increased impulsivity and decreased motivation during five-choice serial-reaction time task chronically post-injury, however, microglia turnover did not ameliorate these deficits. Interestingly, microglial turnover worsened their accuracy during the task. TBI rats also spent more time in the open arm during Elevated Plus Maze testing, but microglial turnover had no effects. Lastly, combination of TBI and microglial turnover worsened Morris Water Maze performances. **Conclusion:** Moderate, focal injury to the prefrontal cortex induced neuroplasticity changes amongst the region responsible for impulsivity and decision-making. The subacute microglial response may be critical in impulsive behavior development. Further work will be conducted to understand the effects of microglial turnover and neuroplasticity changes that may explain the chronic psychiatric symptoms observed in human patients.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

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Topic: D.04. Brain Injury and Trauma

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Research funds of the University of Missouri (Z.G.).

Title: The interplay of non-inertial primary traumatic brain injury and tau: alterations in phosphoproteome networks and cognitive function following low-intensity blast exposure

Authors: *T. MONY^{1,2}, W. YU¹, M. JACKSON^{1,2}, A. ZUCKERMAN^{1,2}, S. CHEN^{1,2}, H. R. SIEDHOFF^{1,2}, A. BALDERRAMA^{1,2}, R. LI^{1,2}, G. Y. SUN¹, J. CUI^{1,2}, Z. GU^{1,2};

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Abstract: Low-intensity blast-induced neurotrauma (BINT) exposure is a known risk factor for cognitive dysfunction and may interact with tau leading to tau-related pathology to influence neurodegeneration manifested in tauopathies including frontotemporal dementia (FTD). To investigate behavioral heterogeneity and underlying molecular mechanisms, we combined cognitive assessment with phosphoproteomic profiling in a FTD mouse model overexpressing human tau (hTau). Two-month-old male hTau and non-carrier mice anesthetized with ketamine/xylazine mixture, were exposed to a single low-intensity blast (LIB) using open-field paradigm in the upright position placed on a platform at 3 meters away from detonation with a 350 g C4 at peak overpressure of 46.6 kPa. Sham animals conducted identical anesthesia and handling except blast exposure. After one-month post-injury, learning and cognitive flexibility were examined using a home cage monitoring system for high-resolution and sensitive assessment. The reversal learning data underwent the Boltzmann fitting curves. A flow chart was developed that enable categorizing individual mouse to different levels of learning deficits and patterns. Mice were classified into cognitive subgroups—normal learners, mild deficits, and

learning deficits—based on deviation from modeled curves, revealing pronounced inter-individual variability in reversal learning even among genetically identical animals. This study revealed a considerable degree of complexity and heterogeneity in learning flexibility across different groups, notably in relation to BINT. To determine molecular correlates of this variability, cortical and hippocampal tissues were analyzed for quantitative global and phosphoproteomics by timsTOP PRO platform. Results showed LIB exposure induced persistent phosphorylation changes in proteins involved in synaptic plasticity, cytoskeletal structure, and survival signaling. Several phosphoproteomic profiles aligned with behavioral classifications, suggesting mechanistic links between tau-associated signaling and individual cognitive outcomes. This study unveils novel interplays between genetic susceptibility of tauopathy and BINT on cognitive dysfunction, supporting the use of precision phenotyping and phosphoproteomics to uncover therapeutic targets.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR276.03/J5

Topic: D.04. Brain Injury and Trauma

Support: NIH Grant 5R01NS120099-04
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Title: Measurement of brain injury-induced sleep disruptions using a custom-built activity monitoring set-up

Authors: *I. A. DÍAZ NIEVES¹, A. FARRUGIA², A. S. COHEN²;

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Abstract: Sleep disruptions like excessive daytime sleepiness and sleep fragmentation are common after traumatic brain injury (TBI). These disruptions are thought to arise from damage to brain regions responsible for regulating arousal and wakefulness, such as the lateral hypothalamus. Our lab focuses on utilizing electrophysiological recordings in brain slices to assess alterations in neuronal activity after injury. To relate changes in *ex vivo* recordings to changes in behavior, it is important to know exactly how an animal's sleep varies before and after TBI. Our approach takes advantage of the fact that sleep-wake cycling in mice is highly correlated with their locomotor activity. By analyzing this activity, we can reliably estimate the amount of time spent sleeping and assess sleep architecture. To this end, we designed a custom actigraphy set-up consisting of (1) an open-topped box, (2) a cage lid equipped with infrared (IR)-emitting LEDs, (3) a USB camera sensitive to IR light, and (4) a food hopper and water

bottle. We used a segmentation algorithm to measure activity as a proxy for sleep in mice before and after undergoing lateral fluid percussion injury. We found that injured and sham mice do not differ in the amount of time spent asleep during the light phase. However, we did observe an increase in the average length of a sleep bout in our injured mice during the dark phase. These results show that mild TBI causes alterations in sleep architecture, a finding consistent with previous literature using other sleep assessment strategies like electroencephalography (EEG) and supports the notion that our strategy is effective for studying injury-induced sleep disruptions.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

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Program #/Poster #: PSTR276.04/J6

Topic: D.04. Brain Injury and Trauma

Support: VA IK2BX006125

Title: Chronic deficits in probabilistic reversal learning resulting from TBI are associated with the structural integrity of cortico-striatal white matter tracts.

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Abstract: Patients with a TBI are 3 times more likely to develop a neuropsychiatric disorder and display changes in impulsivity, reward valuation, and decision-making, but current treatment strategies do not address chronic cognitive impairments contributing to this increased risk. Controlled cortical impact (CCI) in rodents replicates contusion, concussion and hemorrhage observed in humans. CCI damage to the prefrontal cortex reliably results in reward learning deficits, functions commonly attributed to the cortico-striatal network. We hypothesize that chronic behavioral interruptions in reward learning are related to structural integrity of the cortico-striatal network. In 46 rats (22 males; 24 females), we measured behavior on a probabilistic reversal learning task (PRL), requiring reward discrimination, valuation, and behavioral flexibility, up to 12 weeks after injury (20 sham; 26 severe TBI). During the PRL task, rats choose between a high probability reward port (80% chance of reward) or low probability reward port (20% chance of reward). Reversals, where high and low probability ports switch, occurred if eight of the ten previous trials were from a high probability port. For each rat, behavior on the PRL task is compared to structural integrity of white matter tracts measured from ex-vivo diffusion tensor imaging scans. Bifrontal TBI chronically impairs behavior on the PRL task. Rats with TBI perform significantly fewer reversals (linear mixed model, $p < 0.001$), show disrupted Win-Stay behavior ($p < 0.001$) and have trouble discriminating between reward contingencies ($p = 0.002$). We found that white matter integrity was also significantly impacted at

the focal injury site (mPFC) (t-test, fractional anisotropy, $p=.006$) and its subcortical afferents (striatum; fractional anisotropy, $p=0.023$). Moreover, diffusion metrics were correlated with PRL behavior (fractional anisotropy, $R^2=0.03$). Rats with chronic structural cortico-striatal damage performed fewer reversals. Overall, females showed a greater behavioral and structural impact of TBI than males. Our results confirm that chronic reward learning deficits following TBI are associated with damage to cortico-striatal structures.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

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Topic: D.04. Brain Injury and Trauma

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Northwest Network Geriatric Research, Education and Clinical Center
Northwest Network Mental Illness Research, Education and Clinical
Center

Title: Seasonal and circadian variation in acute outcomes of blast trauma in female and male mice

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Abstract: All animals have internal rhythms that can be influenced by extrinsic annual and daily variance. Circannual (seasonal) and circadian (daily) rhythms help entrain the body to the demands of the external environment. While circadian rhythm biology has received increased focus within the stress research community, the influence of season on experimental results has not been well studied. This is of particular interest to stress neuroscientists given the complex interplay between stress, hormones, and behavior. The present study investigates how acute outcomes and survival of blast-induced trauma are impacted by season and time of day in order to increase rigor and reproducibility in stress and trauma research. Blast polytrauma is a full-body injury that results in complex pathophysiological and behavioral outcomes including mild

TBI, PTSD, chronic pain, and risky substance use. We conducted a meta-analysis of historical data from our lab, and here we detail results from over 370 female and male adult C57Bl6 mice that were exposed to either single or repetitive (3x) blast polytrauma (or sham control). Acute outcome variables examined include loss of righting reflex time (LORR), weight change after blast exposure, and mortality. Covariates include sex, time of year (season), time of day (hour), initial body weight, and blast pressure (PSI). We used a multivariate linear regression model to determine the significance of the interactions of our variables. Our results show that LORR and weight change after blast exposure vary significantly by season ($p < 0.01$). Weight loss after multiple blast exposures in female mice is significantly higher in summer and autumn compared to spring ($p = 0.005$; $p = 0.024$). Weight loss after three blasts in females was significantly higher in late morning compared to early afternoon ($p = 0.001$). The percentage of female mice that survived the first blast is significantly higher for those that received a blast exposure in early afternoon compared to late morning ($p = 0.036$). The percentage of male and female mice that survived the first blast exposure is significantly higher in winter than summer ($p = 0.038$; $p = 0.013$). The percentage of male mice that survived all three blast exposures is significantly higher in early evening than in late morning ($p = 0.045$). These results highlight a complex interaction between sex, time of year, and time of day. Ongoing work now focuses on using a random forest machine learning approach to better how these factors are working to contribute to adverse blast outcomes. The results of this study highlight the importance of considering seasonal and circadian rhythms as covariates in stress-related research.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

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Program #/Poster #: PSTR276.06/J8

Topic: D.04. Brain Injury and Trauma

Support: U.S. Department of Veterans Affairs (I01 BX005127 and I01 BX006062)
U.S. Department of Veterans Affairs (5IK6BX005690-05)

Title: Influence of traumatic brain injury timing on neurobehavioral outcomes

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Abstract: Circadian rhythms are intrinsic biological cycles synchronized with the 24-hour solar day, enabling organisms to adapt to regular environmental changes. In mammals, these near-24-hour rhythms are driven by molecular clocks in the brain, which are reset each day by exposure to light-dark cycles to maintain precise timing. These internal clocks regulate the transcription of

more than half of all protein-coding genes, coordinating a wide range of physiological and behavioral functions. The circadian clock influences the progression of several neurological diseases and dictates the effectiveness of neuroprotective therapies. Here, we investigated whether the time of day at which traumatic brain injury (TBI) occurs influences behavioral and neuropathological outcomes in male and female mice. TBI was induced by controlled cortical impact (CCI) injury during either Zeitgeber Time (ZT) 1-4 or ZT13-16, corresponding to early light and early dark phases, respectively. Behavioral assessments revealed no significant differences in post-TBI motor function between the ZT1-4 and ZT13-16 groups in either sex, when compared to sex-matched naïve controls. However, female mice exposed to CCI at ZT13-16 exhibited increased anxiety-like behavior relative to naïve controls, though no significant differences were found between injured groups in either sex. Similarly, spatial learning and memory performance did not differ significantly between time-of-injury groups in both males and females. Histological analysis showed that post-TBI cortical lesion volumes were also not significantly different between ZT1-4 and ZT13-16 groups in either sex. Overall, this study suggests that the time of day at which TBI occurs does not significantly influence the extent of brain injury or behavioral impairments in male and female mice.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR276.07/J9

Topic: D.04. Brain Injury and Trauma

Title: Foot Shock Stress paired with CHIMERA-induced TBI suppresses exploratory behavior

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Abstract: Traumatic brain injury (TBI) is a prevalent and serious consequence of domestic violence, especially in cases involving physical abuse, such as head trauma caused by punches, kicks, or blunt objects. Survivors of intimate partner violence can experience both mild and severe TBIs, leading to long-term cognitive, emotional, and physical impairments. The symptoms of TBI such as memory loss, confusion, headaches, mood swings, and difficulty concentrating—often overlap with the effects of emotional and psychological abuse, complicating diagnosis and treatment. To date, there are no animal models specifically representing TBI in the context of domestic violence. The present preclinical study aims to develop a novel animal model to explore the effects of pairing stress paradigms with CHIMERA-induced TBI. Two cohorts of female C57BL6 mice (N=48) were acclimatized to investigator handling over a series of four days. On the study day, animals were either exposed to foot shock

stress (FSS) or placed in a fear conditioning chamber without foot shock stress (NFSS) for five minutes. Within thirty minutes, mice either received SHAM-TBI or TBI using the Closed-Head Impact Model of Engineered Rotational Acceleration (CHIMERA). Mice that experienced TBI, whether paired with FSS or NFSS, showed increased time to righting and walking compared to SHAM-FSS and -NFSS mice. The mice were divided into two subsets: Cohort 1 (N=24) performed in the Elevated Plus Maze (EPM) and Cohort 2 (N=24) performed in the Open Field Test (OFT) three hours post-injury. Mice performing in the EPM demonstrated no difference in total distance explored, however, the velocity of TBI-FSS compared to TBI-NFSS was significantly reduced ($p<0.05$). SHAM and TBI, FSS and NFSS mice did not differ in time spent or frequency of entering the open arms. However, TBI FSS and NFSS had significantly increased latency to enter the center and open arms ($p<0.0001$). Cohort 2 mice performing in the OFT demonstrated no difference in the total distance travelled in the apparatus. However, TBI mice rested significantly more than SHAM mice ($p<0.5$). TBI mice spent less time being active and had reduced horizontal counts ($p<0.05$). These findings suggest that prior exposure to FSS subsequent to CHIMERA-induced TBI led to distinct behavioral outcomes in the EPM and OFT. Future work will measure gene expression of relevant stress and TBI markers and corticosterone levels. These results imply that heightened stress prior to TBI may significantly influence the progression of TBI outcomes. Further research is needed to better understand this relationship.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

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Topic: D.04. Brain Injury and Trauma

Support: NINDS R01-NS110905-01
ICRG

Title: Frontal brain injury alters Pavlovian responding and impairs decision-making, potentially through nucleus accumbens dysfunction: Evidence from in vivo calcium activity

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Abstract: Traumatic brain injury (TBI) is a leading cause of disability with chronic cognitive and behavioral impairments. Decision-making deficits are common, which could increase vulnerability to addictive disorders such as gambling disorder (GD). Gambling-like decisions can be evaluated using the Rodent Gambling Task (RGT), which recapitulates impairments seen after TBI. A version with win-paired cues (cued RGT; cRGT) further promotes risky choices, particularly following TBI. A key mechanism implicated in addictive disorders is the excessive

attribution of motivational value to reward-paired cues—an influence that may similarly shape post-TBI decision-making deficits. This work investigates how frontal TBI affects sensitivity to probabilistic outcomes and reward-paired cues, as well as real-time neural activity in the nucleus accumbens (NAc) during GD-related behaviors. Adult rats (N=80) received moderate-severe bilateral frontal controlled cortical impact or sham surgery. In Experiment 1, rats were assessed for cue sensitivity by pairing a light (CS+) and lever with sucrose (US+) over 10 sessions, followed by a conditioned reinforcement probe measuring lever presses for the CS+. Subsequently, rats completed the cRGT, a four-choice task varying in reward and punishment probabilities and magnitudes, where win-paired audiovisual cues scale in salience with reinforcer size. While TBI reduced sign-tracking and conditioned reinforcement, indicating weaker cue influence, TBI rats paradoxically made cue-driven risky choices on the cRGT. In Experiment 2, Pavlovian conditioning was modified to use auditory rather than visual cues, confirming that TBI effects were not due to spatial cue proximity. Results replicated Experiment 1, with TBI reducing cue-driven conditioned behaviors. In Experiment 3, we incorporated fiber photometry with GCaMP6f to monitor NAc activity in TBI and sham rats (N=28), prioritizing the NAc based on Δ FosB evidence of reduced activity post-TBI. Rats completed Pavlovian conditioning and the cRGT to explore real-time neural correlates of decision-making deficits. Initial results indicate variable NAc signals during Pavlovian conditioning and outcome-tied signaling during the RGT. These findings can inform future targeted cellular and molecular approaches. The integrated behavioral and neural data reveal complex, dissociable TBI effects on cue-driven decision-making and offer a foundation for probing how maladaptive decision-making emerges after TBI.

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Poster

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Topic: D.04. Brain Injury and Trauma

Support: NIH R01NS109585
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Title: Sleep fragmentation intensifies fatigue-like behavior and sleep architecture disruption after traumatic brain injury

Authors: C. J. COTTER¹, Z. TAPP¹, S. HOULE², J. F. SHERIDAN³, J. P. GODBOUT⁴, A. M. INGIOSI⁵, *O. N. KOKIKO-COCHRAN¹;

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Abstract: Sleep wake disturbances (SWDs) affect many survivors following traumatic brain injury (TBI); however, the expression of non-rapid eye movement sleep (NREMs) and rapid eye movement sleep (REMs) is a complex homeostatic process that is influenced by both biological and environmental factors. We hypothesize that extrinsic stressors such as environmental stimuli amplify SWDs after brain injury. Here, we examine the acute, sub-acute, and chronic effects of TBI on sleep architecture. Additionally, we evaluate the combined effects of mechanical sleep fragmentation (SF) on sleep architecture following injury. Male and female C57BL/6/J mice received a moderate lateral fluid percussion TBI or sham injury at 10 weeks of age. Each mouse was implanted with a wireless telemetry sensor to monitor sleep expression and activity. Half of the mice in each injury group were undisturbed, and half were exposed to 4 hours of daily SF (ZT 0 -3) for 30 days post-injury (DPI). SF after TBI advanced activity deficits by one week compared to TBI control mice. Cosinor analysis of activity showed that TBI and SF independently induce fatigue-like behavior, but when combined, this response is exacerbated and associated with dysfunctional control of rest-activity circadian profiles. Overall, TBI impaired REMs expression acutely with only modest effects on wake and NREMs expression chronically. As expected, SF interrupted long bouts of NREMs with chronic ablation of REMs. Sham SF mice compensated for decreased REMs expression, but this was not the case for TBI SF mice. Sleep bout analysis revealed altered NREMs quality and consolidation during the inactive phase (ZT 12-23) after TBI and TBI SF. Considerable changes in sleep bout structure for REMs was only apparent in TBI SF mice several weeks after injury and during the active phase (ZT 4-11). Using spectral analysis for assessment of sleep microarchitecture, we observed a blunted NREM delta power phenotype in TBI SF mice only. Additionally, REMs power spectra were altered temporally and revealed exacerbated blunting due to the combination of TBI and SF including decreased REMs beta (8-20 Hz) and gamma (20-30 Hz) power. Together, these data highlight the vulnerability in NREMs and REMs following TBI as well as the critical role of environmental stimuli in exaggerating activity impairments and SWDs over time.

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Poster

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Topic: D.04. Brain Injury and Trauma

Support: NIH R01 NS1030517 to KML
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Title: Early life stress and pediatric traumatic brain injury cause sex-specific effects on behavior, the stress response, and microglia-synaptic interactions in rats

Authors: M. A. SMAIL, M. MCDONALD, R. BOLAND, K. G. LEE, C. N. DYE, M. R. BREACH, K. M. MARTENS, J. P. GODBOUT, C. VONDER HAAR, O. N. KOKIKO-COCHRAN, *K. M. LENZ;

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Abstract: Children are vulnerable to the adverse effects of traumatic brain injury (TBI) because their brains are still developing. Pediatric TBI has been linked to social, cognitive, and mood-related impairments later in life. A history of early life stress (ELS) (i.e., poverty, parental absence) can increase the risk for poor outcomes following injury. Furthermore, sex differences have been observed in the symptomology of both insults. In these studies, we combined rat models of ELS and pediatric TBI to investigate the relationship between early life TBI, ELS, and sex. **Methods:** Studies used a 2 (ELS) x 2 (TBI) x 2 (Sex) design. ELS consisted of maternal separation in an incubator for 4 hours/day from postnatal day (P) 1-14. TBI consisted of a mild-moderate lateral fluid percussion injury on P15. Control pups were undisturbed. Starting at P45, two cohorts of rats underwent behavioral testing. Cohort 1 (n=8-12) underwent open field/novel object recognition, social interaction, and forced swim tests, and blood was collected after forced swim to assess corticosterone response to stress. Cohort 2 (n=8) were trained and run daily on the rodent gambling task (RGT) for 8 weeks until stable performance was reached. Cohort 3 (n=4-8) was euthanized 7 days post injury (DPI) and brains processed for histological analyses. ELS and TBI impacted multiple behaviors, with the direction of these effects varying by test and sex. In the open field, ELS and TBI together decreased center time in males, while females exhibited a decrease with TBI alone that was blocked by ELS. Novel object recognition was impaired by both ELS and TBI in both sexes, with no additive effect. Active social interaction was decreased by TBI in both sexes. ELS increased passive coping in the forced swim test in both sexes, while TBI decreased passive coping in females alone. The corticosterone response to acute swim stress was increased by both MS and TBI in males but decreased by TBI alone in females. On the RGT, MS+TBI rats displayed increased risky choice and decreased optimal choice relative to controls, MS or TBI groups. Ongoing high-resolution analyses of immunohistochemically labelled tissue are aimed at uncovering MS, TBI, and MS+TBI-induced alterations in microglia interactions with pre- and post-synaptic excitatory markers, vglut1 and PSD95, as well as synapse stabilizing perineuronal nets. Analyses are focused on the hippocampus and prefrontal cortex, two regions that are important modulators of mood, social and cognitive behaviors. Future studies will be aimed at better understanding the molecular substrates underlying these complex interactions between early life stress, TBI, and sex.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

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Topic: D.04. Brain Injury and Trauma

Support: 1IK2BX006567

Title: Latent Sensitization after Traumatic Brain Injury is Driven by the Rostral Ventromedial Medulla

Authors: *Q. CHEN, D. J. CLARK;
Stanford Univ., Stanford, CA

Abstract: Introduction: Patients with traumatic brain injury (TBI) frequently experience chronic headaches and prolonged recovery post-injury. Dysregulated central pain-modulating circuits may underlie these symptoms. We hypothesized that TBI leads to enhanced nociceptive sensitization after stress or minor injury, driven by mu-opioid receptor-positive (OPRM+) nociception-facilitating neurons (ON-cells) in the rostral ventromedial medulla (RVM).

Methods: We employed a recently developed mouse model of closed-head TBI, combined with models of incisional pain and bright light stress (BLS)-induced headache. Nociceptive sensitization was quantified using mechanical thresholds in the hindpaw after incision or in the periorbital area after BLS. Latent sensitization was tested by administering naloxone after nociceptive thresholds returned to baseline. In separate cohorts, RVM OPRM+ neurons were lesioned using dermorphin-saporin (Derm-sap) and sensitization was assessed following incision or BLS.

Results: Post-incision, mice without TBI (control) recovered within 7 days, whereas TBI mice showed extended hindpaw allodynia up to 38 days. A 15-min exposure to BLS triggered periorbital allodynia in TBI but not control mice. Central, not peripheral, OPRM blockade induced hindpaw allodynia in TBI mice. Control mice without TBI did not display hindpaw allodynia after OPRM blockade. RVM OPRM+ neuron lesioning (Derm-sap) prevented nociceptive sensitization after incision and BLS in TBI mice.

Conclusion: TBI leads to dysfunctional descending pain modulation. The “recovery” after TBI is not a simple return to a pre-injury state but rather a progression to a state of latent sensitization of nociception-facilitating neurons in the RVM, which can be unmasked by stress, injury, or opioid antagonism. Lesioning RVM nociception-facilitating neurons mitigates this sensitization. These findings suggest that targeting endogenous pain-facilitating mechanisms could benefit individuals with post-TBI pain.

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Poster

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Title: Sex-specific Alterations in Cortical Dopamine Metabolism in a Mouse Model of Alzheimer's Disease

Authors: *J. LANGMAN, M. UDO, D. J. SMITH, J. ZACCARELLI MAGALHÃES, L. H. MATUGUMA, C. T. CITADIN, H. LIN;
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Abstract: Background and Methods: Dopaminergic dysfunction is becoming recognized as a potential contributor to Alzheimer's Disease (AD) physiology, yet the enzymatic and metabolic mechanisms involved have been unexplored. In this study, we investigated sex-specific alterations in dopamine metabolism in the 3xTg-AD mouse model using ultra-performance liquid chromatography-mass spectrometry (UPLC-MS) and protein quantification with ProteinSimple automated capillary-based Simple Western (JESS) assays. We quantified dopamine (DA), and associated metabolites: 3-Methoxytyramine (3-MT), 3,4-Dihydroxyphenylacetic acid (DOPAC), Homovanillic acid (HVA), and protein levels of Catechol-O- methyltransferase (COMT), Monoamine oxidase (MAO). **Hypothesis:** In Alzheimer's disease, sex-specific differences in cortical dopamine metabolism are driven by altered expression or activity of dopamine-degrading enzymes, contributing to imbalances in dopamine signaling. **Results:** Male 3xTg-AD mice presented with significantly lower dopamine levels (-1.999 ± 0.9103 nmol/g) in the cortex, along with decreased 3-MT (-0.6676 ± 0.3306 nmol/g) and increased metabolite-to-dopamine ratios (3-MT/dopamine and DOPAC/dopamine) (0.7399 ± 0.2339 nmol/g, 1.293 ± 0.4120 nmol/g, respectively), as well as elevated HVA/DOPAC (0.8816 ± 0.3761 nmol/g) and HVA/3-MT (1.859 ± 1.102 nmol/g) ratios, suggesting an increased in catabolic activity. In line with increased COMT-mediated turnover, 3-MT levels were significantly decreased (-0.6676 ± 0.3306 nmol/g) while COMT protein expression was significantly elevated (0.01057 ± 0.003965 AU). On the other hand, female 3xTg-AD mice showed elevated levels of DOPAC (0.9084 ± 0.4269 nmol/g) and HVA (0.6503 ± 0.2630 nmol/g), but no significant changes in their COMT expression and their metabolite ratios, indicating a more stable metabolic profile. **Conclusion:** These results suggest that whereas female 3xTg-AD mice presented with more moderate changes, males show increased cortical dopamine degradation, possibly due to increased COMT activity. This sex-specific dysregulation of cortical dopamine metabolism may contribute to male-specific vulnerability to AD pathology.

Disclosures: J. Langman: None. M. Udo: None. D.J. Smith: None. J. Zaccarelli Magalhães: None. L.H. Matuguma: None. C.T. Citadin: None. H. Lin: None.

Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR276.13/J15

Topic: D.04. Brain Injury and Trauma

Support: CIHR FRN 175042

Title: Adolescent repeated mild traumatic brain injury impairs long term potentiation in a sex dependent manner

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Abstract: Repeated mild traumatic brain injury (r-mTBI) can be associated with long-lasting cognitive and emotional impairments, often linked to disrupted synaptic function. The hippocampus, a critical regulator of these behaviours, is highly vulnerable to injury, often exhibiting structural atrophy and impaired long-term potentiation (LTP). However, the dentate gyrus (DG) remains underrepresented in r-mTBI research, and this gap is further exacerbated when considering sex-specific effects during adolescence. This study aimed to address these omissions by investigating how r-mTBI alters synaptic plasticity in the adolescent DG between males and females. On postnatal day 35, r-mTBIs were induced in adolescent male and female Sprague-Dawley rats using a lateral impact model. To assess hippocampal synaptic plasticity, field electrophysiology recordings were conducted in acute brain slices on post-injury day 7, targeting the medial perforant pathway of the DG. Sample sizes were determined based on power analyses and prior literature to ensure statistical validity; additionally, sham controls were used to account for the effects of anesthesia, handling, and experimental procedures. Field recordings revealed that r-mTBI significantly reduced LTP in the DG compared to sham controls ($p < 0.05$, two-way ANOVA, $n = 6$ animals per group, $n = 18$ slices per group). This reduction was primarily driven by female rats who exhibited significantly greater impairment in LTP following r-mTBI than males. Overall, mean fEPSP slope potentiation was reduced by $26.08\% \pm 4.81$ SEM following r-mTBI compared to sham injuries. Baseline fEPSP properties (paired-pulse ratio, input/output curves) were unaffected by injury or sex, indicating that LTP deficits reflect synaptic plasticity impairments rather than changes in slice health. These findings suggest that the dentate gyrus is a site of heightened synaptic vulnerability, with sex-specific impairments in plasticity emerging during adolescent development following r-mTBI. However, the precise underlying mechanisms driving these differences remain unresolved. Future experiments will employ whole-cell patch-clamp electrophysiology to examine AMPA and NMDA receptor-mediated synaptic changes.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

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Program #/Poster #: PSTR276.14/J16

Topic: D.04. Brain Injury and Trauma

Support: AHA 24POST1196128
NIH 1R01AG081258-01A1
NIH 1R01AG081874-01
NIH 1RF1NS132291-01
AHA 22TPA970253

Title: Alzheimer's disease hypoperfusion is driven by an imbalance between A2A and A1 adenosine receptors

Authors: *M. S. B. UDO¹, J. ZACCARELLI MAGALHÃES², C. T. CITADIN², L. H. MATUGUMA², D. J. SMITH², J. LANGMAN², H. LIN²;
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Abstract: Background: Alzheimer's disease (AD) affects nearly seven million older adults in the United States, with women at greater risk than men. In addition to hallmark features such as β -amyloid plaques and neurofibrillary tangles, chronic cerebral hypoperfusion has been identified as a contributing factor to neuronal loss in AD. Elevated levels of adenosine receptor A2A (A2AR) and reduced levels of A1 (A1R) have been observed in the AD brain. These receptors play a key role in regulating cerebral blood flow and neurotransmission, acting as a functional "switch." Disruption in their balance may contribute to impaired perfusion. The goal of this study is to modulate A2AR and A1R expression to restore proper blood flow in the brain of aged AD mice. Method: Aged (15-month-old) female 3xTg-AD and C57BL/6J mice were treated with istradefylline (A2AR antagonist - 2 mg/kg/day, IP) or tecadenoson (A1R agonist - 15 μ g/kg, IP) for 7 days. Vascular reactivity, brain perfusion, blood-brain barrier (BBB) integrity, A2AR and A1R expression were evaluated via two-photon laser scan microscopy, laser speckle, and western blotting. Results: Aged 3xTg female mice showed decreased blood perfusion (198 ± 7.9) compared to age-match C57 mice (313 ± 18.5) that was restored by istradefylline (293 ± 18.3) (both $p < 0.01$). Vascular reactivity was reduced in aged 3xTg ($-5.9 \% \pm 1.5 \%$) vs C57 ($12.4 \% \pm 1.9 \%$, $p < 0.001$) and restored by istradefylline ($7.4 \% \pm 3.1 \%$, $p < 0.01$). Aged 3xTg showed disrupted BBB integrity observed by leakage ($80.3 \% \pm 12.2 \%$ vs $5.4 \% \pm 2.4$ (C57), $p < 0.0001$), and decreased ZO-1 ($p < 0.5$) and occluding ($p < 0.05$) protein expression, that was restored by istradefylline (all $p < 0.01$). Aged 3xTg expressed higher A2AR and lower A1R protein levels vs C57 mice (both $p < 0.05$). Although istradefylline did not decrease the levels of A2AR, it increased the levels of A1R ($p < 0.05$). Although tecadenoson showed improvement in BBB integrity parameters (ZO-1 and occluding), the changes observed in brain perfusion and BBB leakage were not statistically different from 3xTg not treated. Conclusion: Although agonism of A1R were not sufficient to restore A2AR/A1R balance, blockage of A2AR were. We are considering a different pharmacological approach to improve A1R, to mimic the improvements observed with A2AR blockage.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

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Topic: D.04. Brain Injury and Trauma

Support: US Department of Defense under the Congressionally Directed Medical Research Program (CDMRP)
National Hockey League Players Association Challenge Fund

Title: Synergistic effects of repetitive mild traumatic brain injury and reduced cholinergic tone on attentional deficits and neurodegenerative pathology with aging in humanized A β and Tau mice

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Abstract: Mild traumatic brain injury (mTBI) increases the risk of developing Alzheimer's disease. Both early cholinergic decline in Alzheimer's disease and cholinergic damage following TBI suggest a distinct role of the cholinergic system in vulnerability to Alzheimer's disease after brain injury. This study investigated the combined effects of repetitive mTBI and cholinergic dysfunction on cognitive deficits and Alzheimer's-related neurodegenerative pathology in transgenic mice that carry humanized beta-amyloid and tau genes. Repetitive mTBI was induced at 6 months old using a closed head impact injury delivered three times at 24-hr intervals to produce diffuse axonal injury. Mild cholinergic dysfunction was achieved by using mice heterozygous for a hypomorphic vesicular acetylcholine transporter allele (VACHTKD). Cognitive function was assessed using the rodent continuous performance task. Repetitive mTBI in the presence of an already compromised cholinergic system resulted in chronic, sex-specific attentional impairments under increased attentional demand (shorter stimulus durations). At 6 months post-injury, females exhibited reduced discrimination sensitivity (d') and hit rates, while males had lower d' and increased false alarm rates. These attentional deficits (lower d' and hit rate) persisted at 12 months in injured VACHTKD females but not males. Neither repetitive mTBI nor cholinergic dysfunction alone induced significant deficits. At 12 months post-injury, levels of pathological phosphorylated tau (pThr231) were elevated exclusively in females with higher cortical tau burden correlating with lower hit rates. No differences were present in insoluble A β 42 levels at this time point. At 6 months post-injury, repetitive mTBI exacerbated cholinergic dysfunction in females with injured VACHTKD females exhibiting lower levels of

ChAT-positive cells in the basal forebrain. Injured cholinergic deficient mice also exhibited an increase in autophagy dysregulated astrocytes at 6 months post-injury. The observed cholinergic neuron loss in the basal forebrain of females may have rendered them more susceptible to downstream neurodegenerative processes (i.e., tau phosphorylation) and attentional deficits. These findings indicate the synergistic role of cholinergic dysfunction and mTBI in driving behavioural deficits and neurodegenerative pathology with aging. Further, they indicate a particular vulnerability of females with compromised cholinergic systems to the chronic effects of repetitive mTBI.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

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Topic: D.04. Brain Injury and Trauma

Support: NIH Grant NS110609 (COB)
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Title: Adult hypertension as a clinically-relevant comorbidity augmenting cognitive deficits, anxiety, and pathological outcomes after traumatic brain injury in pediatric and adult rats

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Abstract: In the United States, approximately 2.8 million people sustain a traumatic brain injury (TBI) yearly, with 500,000 ER visits attributable to childhood TBI. Patients experience cognitive and psychopathological symptoms, thus assessing complex attention deficits post-TBI, especially

with comorbidities, is critical. As a prevalent condition, we investigated how hypertension affects cognitive and psychological impairments in adult rats after pediatric or adult TBI. We hypothesized that hypertensive rats will perform worse post-TBI than normotensive rats. Male Spontaneously Hypertensive (SHR) and normotensive Wistar rats received a moderate, right-parietal controlled cortical impact or a sham injury at postnatal day 17 or at four months of age. At approximately five months of age, four tasks measured neurobehaviors in both pediatric-TBI and adult-TBI groups: the 3-Choice serial reaction time test (3-CSRT, sustained attention), T-maze attentional set-shifting (behavioral flexibility), open field testing (OFT, anxiety), and the shock-probe defensive burying test (SPDB, active and passive coping). In the pediatric-TBI cohort, TBI rats required longer training on 3-CSRT, with normotensive TBI rats displaying lower accuracy and higher omissions. Pediatric-TBI SHR rats displayed enhanced distractibility ($p<0.05$). In the adult-TBI cohort, all TBI rats displayed lower accuracy and higher omissions on 3-CSRT, with SHR-TBI rats demonstrating the greatest attentional impairments and more premature responses, indicating impulsivity ($p<0.05$). Pediatric-TBI SHR rats showed impaired rule learning on T-maze and set-shifting deficits when the rule required a left turn, indicating hemispatial neglect ($p<0.05$), and had longer active avoidance times in SPDB ($p<0.05$), indicating increased anxiety. Adult-TBI SHR rats spent less time approaching and actively burying the probe in SPDB ($p<0.05$). In the pediatric-TBI cohort, hypertension enlarged lateral ventricles, while TBI enlarged ipsilateral ventricles regardless of tensive status ($p<0.05$). TBI-SHR rats also exhibited disrupted colocalization of glial fibrillary acidic protein (GFAP, astrocytes) and aquaporin 4 (AQP4, tubules) in the dorsal hippocampus. In the adult-TBI cohort, receiving a TBI while exhibiting hypertension resulted in worse performance on a complex attentional task compared to either TBI- or hypertension-alone groups, with specific effects on percent accuracy and impulsivity measures. Pre-clinical investigations into the effects of underlying conditions like hypertension on TBI are critical to enhance bench to bedside translatability and develop therapies.

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Poster

PSTR276: Traumatic Brain Injury: Behavioral and Cognitive Outcomes

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR276.17/K3

Topic: D.04. Brain Injury and Trauma

Title: Integrative Behavioral Assessment of Mild Traumatic Brain Injury in Mice: Enhancing Translational Relevance with PhenoTyper Home Cage Monitoring

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Abstract: Mild traumatic brain injury (mTBI) represents a significant challenge in neuroscientific research due to its subtle yet persistent cognitive, emotional, and motor disruptions. mTBI is difficult to assess accurately using conventional short-duration behavioral assays. This project demonstrated the advantages of utilizing the PhenoTyper home cage monitoring system (HCM) in detecting delicate behavioral alterations in a rodent model of mTBI. Adult male ICR mice underwent closed-head mTBI via a weight-drop method, with control groups receiving sham treatment. Behavior was either sampled once a day using gold standard methods or continuously assessed over several days post-injury using automated video tracking within the HCM. HCM methods provided information regarding habituation and circadian activity that could not be assessed using standard methods, while complementing information related to anxiety and cognitive flexibility in mTBI mice. Our results indicate that although each method has specific advantages, integrating both is necessary to achieve a comprehensive phenotypic characterization. Importantly, combined results from both methods closely resemble clinical presentations, highlighting the translational significance of incorporating HCM methods into behavioral assessments.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.01/K4

Topic: E.01. Somatosensation – Pain and Itch

Support: Shenzhen-Hong Kong-Macau Technology Research Program (Category C) (SGDX20210823103534005)
CUHK Research Matching Grant (8601370)
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Title: Bu alleviates neuropathic pain by suppressing spinal microglial activation via P2RY12-p38MAPK signaling

Authors: *J. YANJUN;
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Abstract: Neuropathic pain (NP) is a chronic disorder with limited therapeutic options. Emerging evidence suggests that gut microbiota can influence nociceptive processing. Here, we identify BU, a human gut commensal, as a microbiota-derived modulator that alleviates NP through both central and peripheral mechanisms. BU was orally administered in four mouse models of NP: spared nerve injury (SNI), chronic compression of the dorsal root ganglion (CCD), oxaliplatin-induced neuropathy (OXA), and streptozotocin-induced painful diabetic neuropathy (PDN). In all models, BU significantly reduced mechanical allodynia and thermal hyperalgesia. Behavioral improvements were reproducible across independent cohorts. Mechanistic studies revealed that BU suppressed microglial activation and proliferation in the spinal dorsal horn. Immunohistochemistry showed reduced Iba1+ microglia and attenuated morphological reactivity. In addition, BU downregulated phosphorylated p38 MAPK and pain-associated genes including Bdnf, IL6, and TNF α . Pharmacological manipulation confirmed that BU's analgesic effects required inhibition of the purinergic receptor P2RY12, a key regulator of microglia-neuron crosstalk in NP. To assess whether BU's efficacy involved microbial community changes, we performed shotgun metagenomic sequencing across all four models. The PDN model showed the greatest species richness and a distinct microbial composition, likely reflecting hyperglycemia-induced systemic alterations. BU supplementation led to a shift in gut microbiota away from the pain-associated state in all models. Importantly, fecal microbiota transplantation (FMT) from BU-treated mice conferred analgesic effects to antibiotic-treated recipients, providing causal evidence for gut microbiota involvement in pain modulation. Additionally, BU reduced depressive-like behaviors in SNI mice, suggesting that it may influence shared neural circuits mediating pain and emotion. These findings indicate that BU exerts broad-spectrum analgesic and mood-modulating effects by targeting both spinal immune signaling and gut microbial ecology. Together, this study highlights BU as a promising microbiome-based candidate for managing neuropathic pain and its affective comorbidities.

Disclosures: J. yanjun: None.

Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.02/K5

Topic: E.01. Somatosensation – Pain and Itch

Support: KAKENHI 23K24221
KAKENHI 19K07856
AMED 23ek0610027h0003

Title: Spinal dorsal horn-specific protein BEGAIN modulates pathological pain transmission

Authors: *T. KATANO¹, A. YOSHIKAWA¹, M. ABE², M. KAWAMURA², K. SAKIMURA², T. KOBAYASHI¹;

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Abstract: In neuropathic pain, Brain-enriched guanylate kinase-associated protein (BEGAIN) has been reported to be upregulated in the postsynaptic density (PSD) of the spinal dorsal horn, and mechanical allodynia was significantly reduced in BEGAIN knockout (KO) mice (*eNeuro*, 2016). Although BEGAIN is broadly expressed throughout the cerebrum, no protein expression was detected in the ventral horn of the spinal cord by western blotting. In contrast, *BEGAIN mRNA* is extensively expressed across the entire spinal cord. To clarify this discrepancy, we investigated the distribution of BEGAIN using BEGAIN-iCre driver and Ai9 reporter mice. tdTomato fluorescence, representing BEGAIN expression, was specifically observed in the superficial laminae of the spinal cord. Furthermore, proteomic analysis of PSD fractions confirmed BEGAIN protein expression in the dorsal horn but not in the ventral horn. To examine the functional role of BEGAIN in the spinal dorsal horn, we generated conditional knockout (cKO) mice using a floxed BEGAIN mouse line (targeting exons 3-5) and the Lbx1-Cre driver, which induces Cre recombinase expression specifically in the spinal dorsal horn. Successful recombination and BEGAIN protein deletion were verified by PCR and western blotting. Importantly, BEGAIN protein levels in the hippocampus were unchanged between wild-type and BEGAIN-cKO mice, confirming region-specific knockout. Using the cKO mice, behavioral analyses by von Frey test were performed after spared nerve injury (SNI). Mechanical hypersensitivity was observed in both wild-type and BEGAIN-cKO mice following SNI. However, the mechanical allodynia was significantly suppressed in BEGAIN-cKO mice compared to wild-type controls. These findings suggest that BEGAIN in the spinal dorsal horn plays a critical role in modulating pathological pain transmission following peripheral nerve injury.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.03/K6

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH Grant NS101880
NIH Grant NS132398
Pamela and Wayne Garrison Distinguished Chair Endowment

Title: Tonic GABAergic and Glycinergic Inputs Suppress Nociceptive Hypersensitivity by Limiting $\alpha 2\delta$ -1- and mGluR5-Dependent NMDA Receptor Activity at Primary Afferent-Excitatory Neuron Synapses

Authors: *Y. HUANG, H. CHEN, S.-R. CHEN, H.-L. PAN;
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Abstract: Impaired GABAergic and glycinergic synaptic inhibition contributes to the imbalance between excitatory and inhibitory inputs in the spinal cord in chronic neuropathic pain. In this study, we investigated how GABAergic and glycinergic inputs regulate synaptic NMDA receptor (NMDAR) activity in spinal dorsal horn neurons of mice. We found that blockade of GABA_A receptors with gabazine or glycine receptors with strychnine potentiated NMDAR-mediated miniature EPSC (mEPSC) frequency, the amplitude of EPSCs monosynaptically evoked from the dorsal root, and puff NMDA currents in VGluT2, but not VGAT, neurons. These effects were abolished by silencing neuronal activity with tetrodotoxin or in *Cacna2d1* knockout mice. In mice with conditional *Grin1* knockout in primary sensory neurons (*Grin1*-cKO), gabazine and strychnine did not affect mEPSC frequency but still enhanced puff NMDA currents in dorsal horn neurons. Furthermore, intrathecal gabazine- or strychnine-induced nociceptive hypersensitivity was diminished by *Grin1*-cKO, *Cacna2d1* knockout, or $\alpha 2\delta$ -1 C-terminus peptide. Additionally, blocking metabotropic glutamate receptor 5 prevents gabazine- and strychnine-induced increases in NMDAR-mediated mEPSC frequency, evoked EPSCs, and puff NMDA currents in VGluT2 neurons as well as nociceptive hypersensitivity. Our findings indicate that normal GABAergic and glycinergic inhibition tonically suppresses both presynaptic and postsynaptic NMDAR activity at primary afferent-excitatory neuron synapses. $\alpha 2\delta$ -1 and metabotropic glutamate receptor 5 are essential for disinhibition-induced nociceptive hypersensitivity and synaptic NMDAR hyperactivity in the spinal cord.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.04/K7

Topic: E.01. Somatosensation – Pain and Itch

Title: Characterization of 5-HT receptor expression in dorsal horn pain processing circuits of male and female rodents and humans

Authors: *C. M. MURRAY-LAWSON^{1,2}, E. SIMPSON¹, L. DAVID³, E. MARSHALL¹, K. WALKER¹, N. MARTIN¹, K. GRIFFITHS^{1,2}, J. PARNELL^{1,2}, S. TEMI¹, A. DEDEK^{4,2,1}, E. C. TSAI², M. E. HILDEBRAND^{1,2};

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Abstract: Serotonin, or 5-HT, is an important descending modulator of spinal pain processing. Pharmacological studies in rodent preclinical pain models, as well as preliminary clinical findings with psychedelics, suggest that specific 5-HT receptor subtypes may be promising potential analgesic targets. The superficial dorsal horn of the spinal cord is a crucial site for nociception, and yet the molecular and cellular determinants of serotonergic signalling in these circuits are poorly understood, especially across sex and in humans. We therefore investigated

the relative expression and distribution of specific 5-HT receptor subtypes in the dorsal horn of viable spinal cord tissue from male and female rodents, as well as human organ donors. Using complementary single-cell/nuclei sequencing, quantitative reverse transcription PCR, and immunohistochemistry approaches, we systematically compared the relative expression of all 5-HT receptor subtypes (5-HT1-7) across sex and species, as well as how expression changed in the hind paw incision model of postsurgical pain. Surprisingly, we found that the 5-HT2C receptor subtype is robustly expressed in the rodent and human dorsal horn at the mRNA and protein level, which was conserved across sexes. In contrast, the relative expression of 5-HT2A mRNA increased in humans when compared to rodents. Both 5-HT2A and 5-HT2C receptors were preferentially localized to the superficial laminae of the rodent dorsal horn. Finally, we identified that the expression profile for 5-HT receptor subtypes was conserved between baseline and postsurgical pain states. Our findings suggest that both the 5-HT2A and 5-HT2C receptor subtypes may be important modulators of spinal nociception across sex and species, representing emerging molecular targets for the treatment of pain in humans.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

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Topic: E.01. Somatosensation – Pain and Itch

Support: RS-2022-NR067566
IITP-2025-RS-2024-00437359
RS-2024-00361688
2020M3H2A1078045

Title: Spinal Cord Neural Signals as Digital Biomarkers for Chronic Pain Detection in Contusive Spinal Cord Injury of Rats

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Abstract: Traumatic spinal cord injury (SCI) disrupts neuronal homeostasis, leading to chronic pain and spasticity that severely compromises patient quality of life. While neuromodulation has emerged as a therapeutic approach, the mechanism underlying spinal neuronal hyperexcitability remains poorly understood, limiting spinal cord-based closed-loop systems. This study aimed to characterize spinal cord neural signals as digital biomarkers for chronic pain states, examining both evoked and spontaneous pain-related neuronal signals in SCI rat models. Adult male

Sprague-Dawley rats underwent T11 spinal segment using Infinite Horizons (IH) impactor (150 kdyn). Motor function was assessed using the Basso, Beattie, and Bresnahan (BBB) locomotor scale and combined behavioral score (CBS). Spasticity was evaluated with Modified Ashworth Score (MAS), and mechanical sensitivity was measured via paw withdrawal threshold (PWT) testing. At 28 days post-SCI, after behavioral validation of chronic pain, we recorded spinal neural activity from L4-L6 using 16-channel probes in three groups: Normal (n = 5), Sham (n = 5), and SCI (n = 5). Evoked activity was recorded during mechanical hind-paw stimulation (0.4 g - 300 g, 10 steps) using von Frey filaments. Spontaneous neural activity was continuously recorded for analysis of ongoing pain-related signals. Neural data were processed using spike sorting algorithms and analyzed for multi-unit activity patterns. SCI rats showed significantly enhanced responses to ≥ 15 g vs. Normal and ≥ 26 g vs. Sham rats (group $\times$ stimulus interaction, $p < 0.05$). Sham rats showed hypersensitivity only to the highest stimulus intensity (300g) compared to Normal rats. These neural responses aligned with behavioral hypersensitivity. Hypersensitivity-validated datasets were used to train an end-to-end deep learning model that accurately classified low, mid, and high stimulation levels. Additionally, spontaneous activity was segmented into 6-second chunks, enabling a classifier that distinguished chronic pain rats with $>98\%$ accuracy, highlighting its potential for identifying abnormal pain-related neural dynamics. These findings support spinal neural signals as reliable digital biomarkers for evoked hypersensitivity and spontaneous pain. Our work demonstrates that spinal cord-centered measurements provide a foundation for the development of closed-loop neuromodulation systems, advancing adaptive pain management strategies.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.06/K9

Topic: E.01. Somatosensation – Pain and Itch

Support: R01NS128403
R01NS117484.

Title: Stim1 functionally couples to AMPA receptors contributing to the development of neuropathic pain

Authors: Y. ZHONG¹, J. XIA², Y. MEI¹, D. WEI², R. JEAN-TOUSSAINT², Y. TAO¹, *H. HU¹;

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Abstract: Calcium-permeable ion channels have emerged as targets for chronic pain. Increasing evidence indicates that store-operated calcium channels (SOCs) in the nervous system contribute

to neuronal Ca^{2+} signaling. Stromal interaction molecule 1 (STIM1) is a key component of SOC_s and is implicated in various functions of many cell types. However, its expression and functional significance in spinal dorsal horn neurons remain elusive. Here we report that STIM1 is highly expressed in the dorsal horn and specifically in excitatory neurons. To explore the role of neuronal STIM1 in pain plasticity, we generated vglut2 (an excitatory neuronal marker)-specific STIM1 conditional knockout (KO) mice. A deficiency of STIM1 in excitatory neurons attenuates the mechanical and thermal hypersensitivity caused by spinal nerve injury (SNI) in earlier time points. Our results also show that the STIM1 protein level is significantly increased, and the SOC function is enhanced in dorsal horn neurons from SNI mice. Surprisingly, depleting calcium stores from the endoplasmic reticulum (ER) induces a large calcium entry, which is not altered in dorsal horn neurons from vglut2-STIM1 KO mice, suggesting that the involvement of STIM1 in neuropathic pain is not store-dependent. To investigate how STIM1 contributes to the development of SNI-induced neuropathic pain, we recorded field excitatory postsynaptic potentials (fEPSPs) evoked by dorsal root high-frequency stimulation in the superficial dorsal horn from wild type (WT) and vglut2-STIM1 KO mice. Long-term potentiation (LTP) is dramatically attenuated in the dorsal horn from STIM1 KO mice, indicating STIM1 is implicated in synaptic transmission. Furthermore, we found that AMPA-induced Ca^{2+} influx and currents are reduced in vglut2-STIM1 KO neurons, highlighting a regulatory role of STIM1 in AMPAR function in dorsal horn neurons. While Western blot analysis indicates that STIM1 deletion does not alter the expression levels of GluA1 and GluA3, immunoprecipitation results show that STIM1 physically interacts with GluA1. Together, our findings demonstrate that STIM1 plays an important role in the development of neuropathic pain, at least in part through the regulation of AMPAR function. Our study opens a new area in pain research and may suggest potential novel avenues for neuropathic pain.

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Poster

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Program #/Poster #: PSTR277.07/K10

Topic: E.01. Somatosensation – Pain and Itch

Support: KQTD20200820113040070
JCYJ20220818100615033
NSFC82471250

Title: Neuronal and glial YAP/TAZ in the spinal dorsal horn contributes to development of bone cancer pain in mice

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Abstract: The treatment of bone cancer pain (BCP) remains a major clinical challenge and the mechanisms underlying BCP still remain poorly understood. The neural hyperexcitability and glia cell-mediated inflammatory responses in the spinal cord are considered to play a critical role in the spinal central sensitization in BCP. The transcriptional coactivators YAP and TAZ have been shown to be involved in neuronal functions and immune responses in various neurological disorders, including neuropathic pain. Whether and how the YAP and TAZ impact neuronal and neuroimmune alterations in BCP remains elusive. BCP was produced by tumor cell implantation (TCI) in the tibial bone cavity in mice. Behavior tests, biochemical experiments, and electrophysiology studies in the spinal cord were performed on conditional YAP/TAZ knockout and the wild type of control mice. The results showed that TCI treatment led to nuclear accumulation of YAP and TAZ and an upregulation of their mRNA expression in Vglut2⁺ excitatory neurons, microglia, and astrocytes in the spinal dorsal horn. Conditional knockout of YAP/TAZ specifically in Vglut2⁺ excitatory neurons, microglia, or astrocytes significantly alleviates TCI-induced thermal hyperalgesia, mechanical allodynia, and spontaneous pain-like behaviors. Mechanistically, ablation of YAP and TAZ in Vglut2⁺ excitatory neurons suppressed TCI-induced hyperexcitability of the spinal dorsal horn neurons via β -catenin signaling modulation; concurrently, deletion of YAP/TAZ in microglia and astrocytes prevented TCI-induced activation of these glial cells and increased level of proinflammatory cytokines, tumor necrosis factor- α (TNF- α), interleukin-1 β (IL-1 β), and interleukin-6 (IL-6), through regulation of nuclear factor kappa-B (NF- κ B) signaling pathway. These findings reveal a novel role of YAP and TAZ in the pathogenesis of BCP through β -catenin and NF- κ B signaling pathways, respectively, and they contribute synergistically to neural hyperexcitability and inflammatory responses in the spinal cord after TCI treatment. This study suggests a potential molecular target for therapeutic intervention in BCP.

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Poster

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Program #/Poster #: PSTR277.08/K11

Topic: E.01. Somatosensation – Pain and Itch

Support: DoD Grant W81XWH2211100

Title: Paired peripheral nerve and intraspinal stimulation to promote plasticity in spinal cutaneous reflex pathways

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Abstract: Spinal cord injury-related neuropathic pain (SCI-NP) affects ~70% of people living with spinal cord injury (SCI). Generally, treatments for SCI-NP lack mechanistic specificity, which often causes undesired side effects (e.g., opioids to alleviate SCI-NP may result in substance use disorders) and may lead to discontinuation of therapy. Indeed, most currently used treatments do not directly address neural mechanisms responsible for the development and persistence of SCI-NP, for instance, pathologically increased neural transmission in spinal nociceptive pathways below the lesion. Previous works suggest that spinal nociceptive transmission can be reduced via increased transmission in non-nociceptive spinal sensory pathways (e.g., the gate theory). Thus, therapies that seek to increase non-nociceptive spinal transmission could provide a mechanism-based alternative to treating SCI-NP. Here, we explore the capacity of paired peripheral and spinal stimulation to up-regulate neural transmission in non-nociceptive cutaneous reflex pathways after chronic SCI. All experiments were approved by the IACUC at WUSM. The study included 5 neurologically intact, adult male Sprague-Dawley rats (shams) and 9 rats with chronic SCI (T8 contusion). Neuronal activity was recorded from microelectrode arrays implanted into the spinal cord at the L5 dorsal root entry zone *in vivo*. Electrode arrays spanned the dorso-ventral extent of the gray matter, enabling simultaneous characterization of neuronal activity in sensory-dominant, motor-dominant, and integrative networks. Paired stimulation was delivered by stimulating the ipsilateral sciatic nerve using cuff electrodes to elicit a spinal cutaneous reflex, followed by sub-motor threshold intraspinal microstimulation (ISMS) in the deep dorsal horn with delays of 2-10 msec. Our primary outcomes were the average magnitude of intraspinal cutaneous reflex responses to single pulses of peripheral stimulation prior to and after paired peripheral-intraspinal stimulation (changes in voltage using spike triggered averages). We found that spinal responses to peripheral stimulation in sham animals increased after paired stimulation (prior to stimulation: $0.75 \pm 1.31 \mu\text{V}$; after stimulation: $9.58 \pm 1.41 \mu\text{V}$, $p < 0.001$), whereas these responses decreased in chronically injured animals (prior to stimulation: $18.78 \pm 2.32 \mu\text{V}$; after stimulation: $8.88 \pm 2.18 \mu\text{V}$, $p < 0.001$). These findings indicate that our paired stimulation paradigm has the capacity to acutely alter spinal cutaneous reflexes, which could drive therapeutic benefits for SCI-NP by reducing neuronal transmission in spinal sensory pathways.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

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Program #/Poster #: PSTR277.09/K12

Topic: E.01. Somatosensation – Pain and Itch

Support: TWU Division of Biology
NSF PRIME Grant (#19853448)

Title: Neuronal Arborization in the Dorsal Horn of Townes Sickle Cell Disease Mice

Authors: *C. D. BROOMFIELD¹, K. E. SADLER², D. L. HYND¹;

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Abstract: Sickle cell disease (SCD) impacts around 100,000 individuals in the United States. SCD, initially described in 1910, is caused by a mutation in the HBB gene. This gene provides instructions for making beta-globin, a protein part of hemoglobin. The mutation results in an abnormal form of hemoglobin called hemoglobin S (HbS), and HbS causes red blood cells to become stiff and crescent-shaped under certain physiological conditions. These sickled red cells negatively impact essentially every organ system in affected individuals, including the somatosensory nervous system. Indeed, individuals and mouse models with SCD experience life-long debilitating acute and chronic pain. Thirty to forty percent of adolescents and young adults who live with SCD experience episodic pain, and 50% experience chronic pain. The development of SCD mouse models has enabled researchers to begin characterizing the cellular and molecular basis of chronic SCD pain. For example, leading hypotheses suggest that sickle-shaped red blood cells cluster and block small blood vessels, decreasing blood and oxygen flow to tissues and organs. Increased ischemia contributes to increased inflammation and tissue damage, ultimately leading to nociceptive and neuropathic pain. Peripheral molecular targets, including cation channels TRPV1, TRPV4, and TRPC5, have all been identified as contributors to chronic SCD pain, mainly through their ability to increase the excitability and mechanical sensitivity of primary sensory neurons. Significantly less attention, however, has been placed on the anatomical and physiological integration of primary sensory neurons within the spinal cord. Thus, in these studies, we used immunohistochemistry to stain for two specific proteins: growth-associated protein (GAP-43) and beta III tubulin, in SCD mice and control animals. These markers help visualize and examine the neuron arborization in the afferent neurons of the lumbar spinal nerve tracts. We hypothesized that SCD mice would exhibit increased axon arborization in the dorsal horn of the spinal cord. Dorsal horn arborization was indeed increased in Rexed lamina layers I and II (n = 6), suggesting that amplified pain signals from peripheral nociceptors occur at the spinal level in SCD. In ongoing experiments, we anticipate observing increased C-fiber branching in layers I and II, and A-fiber branching in layers I and V, which is expected to result in synaptic plasticity with interneurons and ultimately lead to SCD-induced hyperalgesia.

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Poster

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Topic: E.01. Somatosensation – Pain and Itch

Support: DoD Grant W81XWH2211100

Title: Non-nociceptive cutaneous reflexes engage multimodal spinal networks and influence nociceptive transmission

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Abstract: Non-nociceptive cutaneous reflexes engage A β sensory afferents and a medley of spinal interneurons to rapidly correct movement in response to abrupt, non-painful perturbations. While the spinal networks mediating these responses remain intact below a spinal cord injury, it is not well understood if (and how) they are reorganized. The first objective of this work is to define the spinal networks mediating cutaneous reflexes, both in the neurologically intact spinal cord and following spinal cord injury (SCI). The second objective of this work is to determine whether activation of cutaneous reflexes concurrently modulates neural transmission in spinal pain pathways. This study utilized adult male Sprague-Dawley rats with and without chronic SCI and SCI-related neuropathic pain (SCI-NP). All procedures were approved by the IACUC of Washington University in St. Louis. We electrically stimulated the sciatic nerve *in vivo* while using intraspinal microelectrode arrays to simultaneously record neural activity from the dorso-ventral extent of the spinal gray matter at the L5 dorsal root entry zone. This allowed us to simulate natural non-nociceptive cutaneous reflexes and identify the associated patterns of intraspinal neural transmission. In separate trials, we also elicited nociceptive responses before, during, and after electrically evoked cutaneous reflex activation. To functionally define the classes of spinal neurons recruited by cutaneous reflex activation (with and without concurrent nociceptive sensory feedback), we characterized single- and multi-unit responses as nociceptive specific, low-threshold, or wide dynamic range. We then combine the spatial and temporal information afforded by the intraspinal microelectrode array with information about functional neuron classifications to reveal the intraspinal structure of the cutaneous reflexes. In keeping with their primary role as a sensorimotor corrective response, we find robust neural activity in sensory dominant, sensorimotor integrative, and deep motor regions of the spinal gray matter during cutaneous reflex activation. We also find that nociceptive neural transmission is indeed modulated during activation of non-nociceptive cutaneous reflexes, reflecting some convergence of pathways mediating these spinal networks. Although the specific interneurons implicated in cross-modal actions on nociceptive transmission remain unidentified, they may involve parvalbumin-expressing inhibitory interneurons, which are known to pre- and post-synaptically gate non-nociceptive primary afferent transmission at its synapses with neurons in spinal pain processing pathways.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

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Topic: E.01. Somatosensation – Pain and Itch

Support: UT Southwestern Endowed Scholars Program (A.P.)
Japan Society for the Promotion of Science Overseas Research
Fellowships (S.I.)

Title: Characterization of the spinal projection neuron type map of somatosensory states

Authors: *S. ISHISHITA¹, L. CANO GOMEZ¹, A.-H. POOL^{1,2,3};

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Abstract: Spinal projection neurons (SPNs) link the spinal cord (SC) to higher-order brain centers and are required for all central pain perception for tissue injuries below the neck. Both acute and chronic pain are centrally relayed by SPNs making them attractive therapeutic targets for pain management. However, the precise cell-type composition and specific molecular features of SPNs have remained ambiguous. To reveal the SPN repertoire, we first used spatial single-molecule transcriptomics to comprehensively characterize the cellular diversity within the intact SC. Our analysis revealed at least 110 anatomically and genetically distinct neuron types, exceeding previous estimates by several fold. Building on this, we performed cell type-informed retrograde viral tracing to map SPNs arborizing in higher-order brain centers implicated in pain processing. As a result, we identified 12 neuronal classes as SPNs (SPN1-12). Furthermore, we performed high-throughput stimulus-to-cell type mapping to characterize the responses of each SPN class to a diverse array of pain stimuli including thermal, mechanical, and inflammatory pain states across both superficial (skin) and deep (visceral, muscle, joint) tissues, as well as non-noxious somatosensory stimuli. This analysis revealed an SPN cell-type map of somatosensory states within the SC outlining the logic of SPN class tuning to individual tissue injury types. Our findings allow for a functional dissection of SPN classes in the regulation of supraspinal pain behaviors and inform therapeutic avenues to target this system for central pain control.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

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Topic: E.01. Somatosensation – Pain and Itch

Support: UTSW Endowed Scholars Program
NIH Grant R01MH134799

Title: Anatomy, projectome and functional annotation of mouse spinal cord neural diversity

Authors: *L. E. CANO GOMEZ¹, H. POLDSAM¹, H. C. LAI¹, A.-H. POOL^{1,2,3};
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Abstract: Spinal cord gives rise to central somatosensation and orchestrates autonomic and motor control. How its cellular diversity achieves physiological functions along the longitudinal axis and is wired to higher order brain centers remains poorly understood. Here we used retrograde viral tracing with spatial transcriptomics and multiomic profiling of >800,000 mouse spinal cord neurons to define its output and functional cellular repertoire. We identified two dozen spinal projection neuron classes that link the spinal cord to hind-, mid-, and forebrain centers implicated in itch, touch, and pain revealing the cellular output logic of the spinal cord. Furthermore, we characterized >500 anatomically and transcriptomically distinct neuron types, including many previously obscured by tissue dissociation artifacts and limited sampling. We found that nearly 40% of neuron types show rostro-caudal specificity reflecting different sensory, motor and autonomic functions in cervical, thoracic, lumbar and sacral spinal cord as well as several sexually dimorphic neuron classes. Finally, we assembled the first neuron-class-to-phenotype map of the spinal cord based on genetically defined interventions from existing literature. As a result, we deliver an anatomically and functionally annotated cellular atlas of the complete adult mouse spinal cord to guide future interrogation of the organ.

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Poster

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Topic: E.01. Somatosensation – Pain and Itch

Support: DARPA HR001123S0038-ABC-FP-00
NSF GRFP 2034836
HHMI Hanna H. Gray Fellowship

Title: Coding properties of spinal cord projection neurons before and after nerve injury

Authors: *M. ROSA CASILLAS¹, A. J. CROWTHER³, B. O. AHANONU⁴, A. I. BASBAUM²;

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Abstract: Chronic pain is one of the most common and debilitating medical conditions. Understanding the neural mechanisms that drive the transition from acute to chronic pain is critical for the development of novel treatments. Sensory information is processed by spinal cord projection neurons (SCPNs) before being sent to the brain to produce percepts and behavior. However, in chronic pain it is thought spinal circuits change, leading to non-painful stimuli being perceived as painful. To address how spinal circuits change as pain becomes chronic, we took advantage of our recent methodological advances (Ahanonu et al, 2024) that allow us to monitor the same population of SCPNs in mice over time, before and after injury. In our studies, we longitudinally recorded the activity of genetically defined SCPNs (using Phox2a-Cre floxed-GCaMP6 mice), during development of the neuropathic pain phenotype in the spared nerve injury (SNI) model. Repeated imaging of large numbers of individual SCPNs revealed that in awake animals, SCPNs are predominantly polymodal, responding to different modalities of pain-inducing stimuli. Individual response profiles were consistent over months before injury. Furthermore, in contrast to recordings in anesthetized mice, in the awake state, SCPNs exhibit significant spontaneous and movement-related activity. Consistent with the behavioral observation of mechanical allodynia and cold hypersensitivity in the SNI model, our initial findings are that peripheral nerve injury in the SNI model indeed causes an increase in the responsiveness of SCPNs to mechanical and cold evoked stimuli. By monitoring the activity of the same population of neurons, before and after peripheral nerve injury, we are now able to address longstanding questions about the transition from acute to chronic pain in the awake behaving mouse. Of particular interest is our plan to assess the impact of existing and novel analgesics on the activity of these projection neurons, during the development of chronic pain.

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Poster

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Program #/Poster #: PSTR277.14/L1

Topic: E.01. Somatosensation – Pain and Itch

Support: Eli Lilly & Company

Title: Investigating the central role of Nav1.7 in spinal cord pain pathways

Authors: *C. LIMA-TRUAX, L. YANG, B. L. ADAMS, E. O'NEIL, J. WANG, B. LI, J. KRAJEWSKI;
Eli Lilly & Co., Indianapolis, IN

Abstract: Background: Mutations in SCN9A, gene encoding the voltage-gated sodium channel Nav1.7, lead to either congenital insensitivity to pain or severe pain syndromes. Despite failed trials of Nav1.7 small molecule antagonists, recent findings of Nav1.7 in projection neurons

suggest new strategies to modulate pain centrally, offering renewed potential for effective analgesia beyond the peripheral nervous system. This study aimed to determine whether spinal Nav1.7 contributes to pain modulation by assessing the effects of its knockdown (KD) on pain-related behaviors in an oxaliplatin-induced cold hyperalgesia model. To investigate the central-specific role of Nav1.7 in sensory processing, we utilized a conditional knockout mouse line enabling targeted SCN9A gene deletion in dorsal root ganglion (DRG) neurons. **Methods:** Four commercially available SCN9A-targeting small interfering RNAs (siRNAs) were screened in cultured DRG neurons from wild-type C57BL/6 mice and the most potent was delivered intrathecally *in vivo*. Adult male Pirt-Cre/Nav1.7 mice (8-14 weeks) received a 40 µg unilateral intraplantar oxaliplatin injection to induce cold hyperalgesia. Behavior was assessed on a 10°C cold plate, with nocifensive responses scored by blinded observers. Following behavioral testing, DRG and spinal cord tissues were collected for quantitative polymerase chain reaction (qPCR) and normalized to a housekeeping gene (*Hprt1*). All animal procedures used in this study were approved by the Eli Lilly Institutional Animal Care and Use Committee. Data were analyzed by repeated measures analysis of variance (ANOVA) and unpaired t-tests, with significance at $p < 0.05$. **Results:** *In vitro*, the lead siRNA achieved 79% KD at 1 µM with no off-target effects on SCN10A (encoding Nav1.8). *In vivo*, oxaliplatin administration elicited significant increases in nocifensive flinching 24 hours post-injection ($p < 0.05$). Intrathecal delivery of the selected siRNA achieved approximately 45% reduction of spinal SCN9A expression but failed to produce significant attenuation of pain-related behaviors compared to controls ($p > 0.05$). **Conclusion:** Despite robust *in vitro* siRNA efficacy and significant spinal SCN9A KD, Nav1.7 does not appear to play a key role in mediating oxaliplatin-induced cold hyperalgesia in this model. If a role for spinal Nav1.7 does exist, a more extensive degree of KD may be required to reveal it. Additionally, potential species differences in spinal Nav1.7 function cannot be excluded. These findings highlight the importance of continued investigation into alternative therapeutic targets for oxaliplatin-induced cold hyperalgesia.

Disclosures: C. Lima-Truax: A. Employment/Salary (full or part-time): Eli Lilly & Company. L. Yang: A. Employment/Salary (full or part-time): Eli Lilly & Company. B.L. Adams: A. Employment/Salary (full or part-time): Eli Lilly & Company. E. O'Neil: A. Employment/Salary (full or part-time): Eli Lilly & Company. J. Wang: A. Employment/Salary (full or part-time): Eli Lilly & Company. B. Li: A. Employment/Salary (full or part-time): Eli Lilly & Company. J. Krajewski: A. Employment/Salary (full or part-time): Eli Lilly & Company.

Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.15/L2

Topic: E.01. Somatosensation – Pain and Itch

Title: Molecular and Functional Characterization of hiPSC-Derived Dorsal Horn Neurons for Central Pain Target Discovery

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Abstract: Central sensitization, defined by heightened excitability within the spinal dorsal horn, is a key driver in the development and maintenance of chronic pain. While recent advances—such as the FDA approval of the NaV1.8 inhibitor Suzetrigine—have advanced analgesic discovery targeting peripheral nociceptors, therapeutic strategies addressing central mechanisms remain underexplored. One major barrier is the lack of physiologically relevant human models to validate central nervous system (CNS) pain targets. Here, we present the molecular and functional characterization of spinal dorsal horn neurons (DHNs) derived from human induced pluripotent stem cells (hiPSCs), offering a novel and scalable in vitro platform to investigate central nociceptive pathways and support CNS-targeted drug discovery. Using a defined small molecule and growth factor based differentiation protocol, we generated multiple batches of DHNs and assessed reproducibility via immunocytochemistry and qPCR. Immunolabeling confirmed a neuronal population composed of ~80% glutamatergic and ~20% GABAergic neurons. qPCR analyses showed consistent expression of dorsal horn specific genes including BRN3A, LMX1B, PAX2, GBX1, DMRT3, and LHX5 across independent lots. Bulk RNA sequencing revealed a profile similar to primary human dorsal horn tissues, supporting the regional specificity of the model. To assess relevance for pain research, we profiled key nociceptive genes including ion channels, neurotransmitter receptors, neuropeptides, and central sensitization markers such as GRIN1, SCN9A, TAC1, and OPRM1. Functional maturation was evaluated over time using both multi-electrode array (MEA) recordings and manual whole-cell patch clamp. MEA analysis on Axion CytoView 24-well plates revealed spontaneous and synchronous network activity emerging by week two, reflecting functional interplay between excitatory and inhibitory neurons. Whole-cell patch clamp revealed that 72% of neurons had resting membrane potentials more negative than -50 mV, 86% fired spontaneous action potentials, and 57% exhibited repetitive spiking upon depolarization. All neurons displayed large voltage-gated sodium currents (~3,000 pA), and 81% exhibited miniature excitatory postsynaptic currents (mEPSCs), indicating synaptic activity and advanced maturation. Collectively, these results establish hiPSC-derived DHNs as a robust and scalable human platform for modeling central pain mechanisms, enabling high-content screening and therapeutic discovery for chronic pain conditions.

Disclosures: **V. Truong:** A. Employment/Salary (full or part-time): Anatomic Incorporated. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Anatomic Incorporated. **A. Arendt-Tranholt:** None. **R.E. Petroski:** None. **T.J. Price:** None. **P. Walsh:** A. Employment/Salary (full or part-time): Anatomic Incorporated. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Anatomic Incorporated.

Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.16/L3

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH NSR35097306
NIH F32 5F32DE029384
NSF Graduate Research Fellowship 2034836
HHMI Hanna H. Gray Fellowship

Title: Branching axons of dorsal horn projection neurons establish contralateral and multisegmental spinal networks

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Abstract: Unilateral nerve injury can produce bilateral reports of chronic, mirror-image pain, a clinically significant phenomenon termed allochiria, which manifests in conditions such as Complex Regional Pain Syndrome (CRPS). Previous research has identified polysynaptic commissural pathways that can restore pain signaling following anterolateral tract cordotomy, despite the complete transection of ascending spinothalamic and spinoreticular pathways. Dorsal horn projection neurons (DH-PNs) are well-established for transmitting nociceptive information via ascending anterolateral tract connections to supraspinal centers—the very pathways interrupted by cordotomy—yet anatomical studies suggest these neurons also possess local axonal collaterals that may engage additional spinal circuit elements during injury states. Here, we not only provide functional evidence of contralateral DH-PN activation provoked by unilateral noxious stimulation, but also reveal striking neuroanatomical evidence for contralateral connectivity. Using whole tissue clearing with sparse labeling and pseudorabies virus and wheat germ agglutinin (WGA)-based transneuronal tracing, we characterized the complete three-dimensional axonal architecture of individual Phox2a+ dorsal horn projection neurons and documented transsynaptic spread to contralateral Phox2a+ PNs across multiple spinal segments. Our results demonstrate that projection neurons, in addition to sending contralateral ascending nociceptive signals to supraspinal targets via the anterolateral tract, form extensive local networks spanning multiple segments both ipsilaterally and contralaterally. These findings establish anatomical substrates that may account for compensatory bilateral nociceptive circuit activation following anterolateral tract cordotomy, as well as the bilateral pain characteristic of CRPS and other chronic pain conditions.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.17/L4

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH R01AT011517
Richard Stickel Donation

Title: Terpenes from *Cannabis sativa* relieve neuropathic pain in mice via the Adenosine A2a Receptor expressed in Pax2+ spinal inhibitory interneurons

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Abstract: Chronic pain is a debilitating condition that affects one in five people in the United States adult population. Limited efficacy and/or harsh side effects limit many pain drugs, driving the need for alternate analgesics. In our past research, we have shown that terpenes, small hydrocarbons found in *Cannabis sativa*, are capable of reducing nociception in mouse models via multiple receptor targets. Furthermore, we showed the terpenes geraniol, linalool, β -pinene, α -humulene, and β -caryophyllene are efficacious in the treatment of mouse chemotherapy-induced peripheral neuropathy (CIPN). By utilizing CRISPR knockdown and the small molecule inhibitor istradefylline, we showed that terpenes act in CIPN through the activation of the Adenosine A2a Receptor (A2aR) in the spinal cord. However, the cell type and spinal circuitry for A2aR-induced antinociception by terpenes are unknown. Given the known role of the A2aR as a stimulatory GPCR, we hypothesized that the A2aR is localized in inhibitory interneurons in the spinal dorsal horn and act by inhibiting neuronal pain transmission. For our model we used male and female CD-1 mice treated with paclitaxel (2 mg/kg) to induce CIPN. Using immunohistochemistry, we found that the A2aR was strongly upregulated in the spinal cord by CIPN. We further observed that this A2aR expression was found in 95% of neurons, as measured by colocalization with the neuronal marker NeuN. Following this, we found that the A2aR is expressed in 56% of inhibitory interneurons, as measured by colocalization with the marker Pax2. We then used Pax2+ cell-selective CRISPR knockdown of the A2aR by using the Pax2 promoter to drive Cas9 expression along with a gRNA targeted to the A2aR. This resulted in a complete loss of terpene antinociception in CIPN, strongly suggesting that terpenes work through the A2aR in this specific cell population to relieve pain. To further confirm these findings, we used Fos staining in spinal cord, showing that Fos activation was increased by CIPN and repressed by terpene treatment. Future experiments will include measurement of evoked GABA release in response to terpenes and activity analysis of ascending nociceptive neurons. Taken together, we show that terpenes have antinociceptive effects in neuropathic pain via the A2aR on Pax2+ inhibitory interneurons in mouse spinal cord. With this work, we aim to identify the detailed mechanism by which terpenes relieve neuropathic pain, and establish a scientific basis for their use as a novel non-opioid pain therapeutic.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR277.18/L5

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH Grant NS113189
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Title: Activation of TRPA1 and TRPM3 triggers Ca^{2+} waves in central terminals of sensory neurons and facilitates synaptic activity in the spinal dorsal horn

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Abstract: Transient receptor potential ankyrin 1 (TRPA1) and melastatin 3 (TRPM3) are transduction channels of sensory neurons that play major roles in peripheral mechanisms of somatosensation, including thermosensation, chemosensation and nociception. Recent studies suggest that both channels also contribute to central mechanisms of pain processing at the spinal cord level. TRPA1 and TRPM3 are highly permeable for Ca^{2+} , suggesting that they could regulate Ca^{2+} signalling at spinal synapses. However, information about TRPA1- and TRPM3-induced Ca^{2+} signalling in the dorsal horn (DH) of the spinal cord is lacking. Here, we describe a dual-colour technique for simultaneously measuring Ca^{2+} concentration ($[\text{Ca}^{2+}]_i$) in central terminals of sensory neurons and in spinal DH neurons by green (GCaMP3) and red (jRGECO1a) Ca^{2+} indicators, using two-photon imaging in isolated mouse spinal cord with attached dorsal roots (DR). DR stimulation elicited $[\text{Ca}^{2+}]_i$ transients in axonal boutons of primary afferents and in cell bodies of DH neurons. The antagonists of AMPA and NMDA glutamate receptors, CNQX and AP5, inhibited $[\text{Ca}^{2+}]_i$ transients in DH neurons, but not in sensory axonal boutons. Selective agonists of TRPA1 and TRPM3, ASP7663 and CIM0216, induced complex $[\text{Ca}^{2+}]_i$ responses in distinct but partially overlapping subsets of sensory axonal boutons. Concomitant $[\text{Ca}^{2+}]_i$ elevations were observed in DH neurons, which were blocked by CNQX and AP5. Patch clamp recordings from DH neurons showed that ASP7663 and CIM0216 markedly enhanced excitatory synaptic activity. In summary, our findings suggest that TRPA1 and TRPM3 on central terminals of sensory neurons regulate presynaptic $[\text{Ca}^{2+}]_i$ and synaptic transmission in the spinal DH.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

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Program #/Poster #: PSTR277.19/L6

Topic: E.01. Somatosensation – Pain and Itch

Support: NIDA Grant DA057195
NIDA Grant DA050530

Title: Characterization of novel astrocyte subtype in HIV-induced neuropathic pain

Authors: *A. DIBUA¹, S.-J. TANG²;

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Abstract: An estimated 30-60% of people living with Human Immunodeficiency Virus 1 (HIV-1) experience chronic pain despite the use of antiretrovirals to suppress viral load. The HIV-1 envelope glycoprotein 120 (gp120) is known to promote neuroinflammation and disrupt neuronal circuitry in the spinal cord. One such mechanism by which this occurs is through the activation of reactive astrocytes. Single nuclei RNA sequencing (snRNA-seq) uncovered a novel astrocyte subtype characterized by its specific expression of Galectin 3 (Gal3) that is only present in HIV⁺ patients with chronic pain (Zheng et al., 2022). The discovery of this HIV pain-associated astrocyte (HIPA) was recapitulated in a HIV-1 gp120 transgenic (Tg) mouse model that expressed the same molecular and behavioral pain phenotype observed in humans. Through snRNA-seq, we found that the ablation of Gal3 restored the astrocyte population and pain behavior back to the wild-type phenotype. However, the critical pain-related role of this astrocyte subtype remains unclear. Previous data suggests that HIPAs may originate from the central canal (CC) of the spinal cord in the presence of gp120. We investigated this by developing a novel murine model, gp120Tg/Nestin-GFP^{Tg}, for lineage tracing of ependymal cell progeny in the CC which is visualized through immunofluorescence staining and confocal microscopy. Nestin activity is marked with green fluorescent protein (GFP) and is selectively expressed in neural stem cells, thus ependymal cell activation from the CC will express GFP and can be used for lineage tracing of progenitor cells (Mignone et al., 2004). We found Gal3⁺ expression emerging from differentiating cells in the CC and colocalizing with Nestin-GFP⁺ progenitor cells throughout the grey matter. Gal3⁺ expression also colocalizes with glial fibrillary acidic protein (GFAP) in both gp120Tg and gp120Tg/Nestin-GFP^{Tg} mice, but not in the wild-type. Additionally, Gal3⁺ expression does not colocalize with ionized calcium binding adaptor molecule 1 (Iba1) or neuronal nuclei (NeuN). The results show that the Gal3⁺ HIPA cells likely originate from the ependymal cells of the CC and retain its neural stem cell properties as it migrates through the grey matter in the spinal cord. Additionally, we see that HIPA is also colocalized with GFAP, validating that Gal3⁺ cells are astrocytic. Together, these findings

suggest that the presence of gp120 can stimulate the genesis of HIPAs from ependymal cells, and HIPA may exhibit both neural progenitor and activated astrocyte-like behavior, providing clarity into the role it may play in the spinal cord of patients living with HIV and pain.

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Poster

PSTR277: Spinal Pain Mechanisms and Circuitry

Location: SDCC Halls B-H

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Topic: E.01. Somatosensation – Pain and Itch

Support: NIH Grant R25 DA057802
Genentech Innovation Fund

Title: Intrathecal agmatine maintains EAAT2 immunoreactivity in the dorsal horn of nerve-injured mice

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Abstract: Introduction. Our previous studies show that administration of agmatine, endogenous decarboxylated L-arginine, attenuates neuropathic pain responses dependent on the GluN2B subunit of the NMDA receptor in spinal cord. This suggests that the effects of agmatine relate to reduction of glutamate activation of spinal NMDA receptors. Several research groups have reported that the excitatory amino acid transporter 2 (EAAT2) is reduced under conditions of nerve injury, leading to elevated glutamate concentrations. In the present study, we tested the hypothesis that agmatine treatment prevents the reduction of spinal EAAT2 under conditions of neuropathic pain.

Methods. Von Frey thresholds were collected on female ICR-CD1 mice (21-24 g), which were divided into five groups: 1) naive, 2) saline-treated, 3) agmatine-treated, 4) saline-treated with spared nerve injury (SNI), 5) agmatine-treated with SNI. Saline or agmatine (30 nmol) was delivered intrathecally during surgery (or day 0, groups 1-3), and on days 2, 4, 6. Tactile hypersensitivity was monitored for 7 days. Mice were transcardially perfused with 4% paraformaldehyde. Transverse spinal cord tissue was processed for EAAT2 immunohistochemistry (RB @ EAAT2 IgG, Abcam). We evaluated the colocalization of EAAT2 with neuronal and astrocytic markers and the relative changes in EAAT2-ir between treatment groups. All observations were confirmed by two blinded observers.

Results. EAAT2-ir was evident in the spinal cords of all treatment groups in a pattern consistent with glial expression, as expected. EAAT2-ir appeared decreased in SNI mice treated with saline

relative to saline-treated, uninjured mice. EAAT2-ir in SNI mice treated with agmatine appeared comparable to that of mice not subjected to SNI.

Conclusion. Our observations suggest that repeated i.t. agmatine during and after nerve injury prevents the reduction of EAAT2-ir observed in mice with SNI. These findings provide insight into the mechanisms by which agmatine reduces manifestations of nerve injury-induced pain in mice.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR278.01/L8

Topic: E.01. Somatosensation – Pain and Itch

Title: Applying digital measures to assess dose-dependent effects of a neuroactive compound in mice

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Abstract: Traditional behavioral assessments in preclinical research often rely on manual observations that can be subjective and prone to variability. Digital home-cage monitoring systems offer an objective, continuous, and non-invasive method for quantifying behavioral changes in animal models. This study aimed to evaluate the ability of digital behavioral phenotyping to detect dose-dependent effects of neuroactive compounds.

EnvisionTM by the Jackson Laboratory is an advanced digital in vivo monitoring system designed to assess mouse behavior and physiology in the home cage environment and was used to monitor mice continuously throughout the study. The study design configured in Envision included continuous digital measurement of Activity Suite (Activity, Activity Classification, and Distance Travelled in the Locomotor Activity Bouts) as well as Respiratory Rate for individual mice. Female 11-week-old C57BL/6J female mice (n=18) were housed in Allentown DiscoveryTM IVC cages and were acclimated to the cages for 7 days. Mice were treated with 5 or 25 mg/kg

morphine or vehicle, which was administered at the beginning of both the light and dark cycle and were monitored for 48 hours post-injection.

Using the Envision Activity Suite, dose-dependent behavioral and physiological effects of morphine were clearly identified. The system effectively classified animal behavior into distinct categories or activity: active, inactive, locomoting, climbing. Specifically, mice administered 25 mg/kg exhibited locomotor behavior for a median of 78.6% of the three-hour post-dose period, compared to 17.1% for the 5 mg/kg dose and 2.8% for the vehicle group. Additionally, mice treated with 5 mg/kg morphine spent significantly more time active than locomoting, in contrast to both the vehicle and the 25 mg/kg treatment groups. Respiratory rate analysis revealed a similar dose-dependent effect. Significant decreases in respiratory rate were observed within the first three hours following treatment at both doses. In the 25 mg/kg group, these reductions were sustained for 3-6 hours post-dose. All groups demonstrated a return to baseline respiratory rates within 6-9 hours.

This study demonstrates that computer vision based digital measures from the home cage were effective at detecting dose-dependent effects of morphine on the activity and respiration in mice, representing a powerful advancement in preclinical research and offering rich, high-resolution insights that can improve decision-making in drug development and accelerate the translation of findings into clinical success.

Disclosures: **N. Bratcher-Petersen:** A. Employment/Salary (full or part-time): TLR Ventures. **J. Beierle:** A. Employment/Salary (full or part-time): The Jackson Laboratory. **A.E. Clipperton-Allen:** A. Employment/Salary (full or part-time): The Jackson Laboratory. **J. Oberhauser:** A. Employment/Salary (full or part-time): TLR Ventures. **J. Ostmeyer:** A. Employment/Salary (full or part-time): TLR Ventures. **N.E. Peltier:** A. Employment/Salary (full or part-time): TLR Ventures. **M.C. Saul:** A. Employment/Salary (full or part-time): The Jackson Laboratory. **M. Ruidiaz:** A. Employment/Salary (full or part-time): TLR Ventures. **T. Robertson:** A. Employment/Salary (full or part-time): TLR Ventures.

Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR278.02/L9

Topic: E.01. Somatosensation – Pain and Itch

Title: Investigating the Effect of Xylazine on the Duration of Fentanyl's Antinociceptive Action

Authors: ***E. ZOLALI**¹, J. ZUARTH GONZALEZ¹, T. TORGBENU¹, M. A. SAKIB², L. R. MCMAHON³, S. OBENG⁴;

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Abstract: Fentanyl is a synthetic opioid that accounts for most overdose deaths due to its high potency and pharmacological efficacy, as well as increasing availability and relatively low cost. Co-administration of fentanyl with other drugs such as xylazine (tranq dope) has become a major concern. Anecdotally, xylazine is combined with fentanyl to increase duration of action. Here, we evaluate the separate and combined effects of xylazine and fentanyl by measuring antinociceptive and hypothermic effects in female (N=4) and male (N=4) Sprague Dawley rats. Rectal temperature (°C) was measured with a digital thermometer and antinociception was measured with latency (s) to respond on a hot-plate apparatus set to 52 °C with a 60-s cutoff. After recording baseline values, rats received vehicle (saline), fentanyl (0.32 and 1 mg/kg), xylazine (1 and 3.2 mg/kg), or 3:1 fentanyl: xylazine combinations intended to mimic real-world ratios. Fentanyl at 0.32 mg/kg produced antinociceptive effects (57.4% MPE) lasting 30 min, whereas 1 mg/kg produced maximal effects (100% MPE) lasting 90 min. Xylazine (3.2 mg/kg) produced modest though statistically significant antinociceptive effects (19% MPE). Notably, xylazine (1 mg/kg) attenuated the antinociceptive effects of fentanyl (0.32 mg/kg), reducing MPE from 57.4% to 17.8%. The higher xylazine dose (3.2 mg/kg) did not extend the duration of fentanyl (1 mg/kg)-induced antinociception. Lower doses of fentanyl (0.32 mg/kg) and xylazine (1 mg/kg), whether administered alone or in combination, did not produce significant hypothermia. In contrast, higher doses of fentanyl (1 mg/kg) and xylazine (3.2 mg/kg) each produced significant hypothermia, and their combination produced greater hypothermia than that obtained from either drug alone. These results indicate that xylazine does not prolong the antinociceptive effects of fentanyl and may antagonize some effects of lower fentanyl doses. Although xylazine potentiates the hypothermic effects of fentanyl, it does not extend its duration of action. Our findings do not support the hypothesis that xylazine increases the duration of action of fentanyl. Rather, these results suggest that the combined effects of xylazine and fentanyl may be antagonistic or additive/supra-additive for different effects.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR278.03/L10

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH GRANT R01-NS055159

Title: Trpm3 inhibition by $g\beta\gamma$ is an important mediator of opioid-induced analgesia in inflammatory pain

Authors: *C. BURTON¹, N. KIM¹, Y. YUDIN¹, H. HU², T. ROHACS¹;

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Abstract: TRPM3 is a heat-activated cation channel that is an important detector of noxious heat in peripheral nociceptors and TRPM3 activation is sufficient to induce pain. Previous studies have shown that activation of G_i-coupled G-protein coupled receptors (GPCRs) causes TRPM3 inhibition via a direct interaction between TRPM3 and Gβγ. The physiological relevance of TRPM3 inhibition by Gβγ has not previously been established in a behavioral model. Using CRISPR-Cas9 genome editing, we generated a mouse line (TRPM3^{ΔEx17}) without *Trpm3* exon 17, which encodes the Gβγ binding site, thus disinhibiting TRPM3 following activation of G_i-coupled GPCRs. Calcium imaging of dorsal root ganglion (DRG) neurons isolated from TRPM3^{ΔEx17} mice shows loss of TRPM3 inhibition following activation of G_i-coupled mu-opioid receptors (MOR), thus confirming the physiological relevance of exon 17 in Gβγ-mediated inhibition. Using TRPM3^{ΔEx17} mice, we show that inhibition of TRPM3 is an important mediator of exogenous-opioid-induced analgesia in heat hyperalgesia, but not mechanical allodynia, in the Complete Freund's Adjuvant (CFA) model of inflammatory pain. Based on this finding, we developed a second novel mouse line (TRPM3-Ex17^{fl/fl}) in which *Trpm3* exon 17 is flanked by *loxP* sites to allow for cell-type-specific disinhibition of TRPM3. We crossed TRPM3-Ex17^{fl/fl} mice with *Trpv1*^{Cre} mice to selectively disinhibit TRPM3 in peripheral nociceptors and validated exon 17 deletion using calcium imaging of DRG neurons. Behavior experiments using *Trpv1*^{Cre}; TRPM3-Ex17^{fl/fl} mice show significant reductions in opioid-induced analgesia to heat hyperalgesia caused by CFA-induced inflammation when TRPM3 in peripheral nociceptors is disinhibited. Collectively, our data show that inhibition of TRPM3 in peripheral nociceptors is a key mediator of opioid-induced analgesia in inflammatory heat hyperalgesia.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR278.04/L11

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH/NIDA Grant R01 DA059978

Title: Structure-based design of low-efficacy mu-opioid agonists as overdose-resistant analgesics

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Abstract: Effective pain management remains a critical clinical need, but the widespread use of opioids has contributed to an ever-growing overdose epidemic due to their high potential for abuse, dependence, and respiratory depression. Opioid ligands with low intrinsic efficacy at the μ -opioid receptor (μ OR) offer a promising strategy for separating analgesic properties from these liabilities. In this study, we identified BP1-94 (10-Cyclopropyl MP), a semi-synthetic analog of mitragynine pseudoindoxyl (MP), developed using structure-based design. In BRET Gi/o/z, cAMP, Nb39 recruitment, GTP turnover and [³⁵S]-GTP γ S assays, BP1-94 displays exceptionally low intrinsic efficacy and exhibits pharmacological properties intermediate between traditional μ OR agonists and antagonists. Despite its low efficacy, BP1-94 produced μ OR-dependent antinociception in multiple mouse models of pain, including thermal, inflammatory, cancer, and neuropathic assays, without respiratory depression or conditioned place preference in mice. Moreover, BP1-94 shows promise in alleviating morphine withdrawal, blocking fentanyl respiratory depression as well as morphine self-administration consistent with its low efficacy profile. BP1-94 alone was not intravenously self-administered by rats, but attenuated morphine self-administration, and mitigated physical dependence and withdrawal symptoms. A cryo-EM structure was solved of BP1-94-bound in μ OR in both the active (G protein-bound) and inactive (Nb6-bound) states, this showed that BP1-94 engages a subpocket made up of transmembrane domain (TM) 1, 2, and 7. BP1-94 engagement in this subpocket leads to a conformational rearrangement of TM1 and TM7, specifically the 'YYH' motif (Y^{1.39}, Y^{2.64} and H^{7.36}). Molecular dynamics simulations and single molecule FRET indicate that the conformational change in this YYH motif may translate to reduced TM6 outward swing, thereby providing a mechanistic basis for BP1-94's reduced efficacy. These findings highlight BP1-94 as a low-efficacy μ OR ligand with adequate analgesic potential and markedly attenuated opioid-related side effects, supporting further development as a safer therapeutic alternative for pain treatment.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

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Program #/Poster #: PSTR278.05/L12

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH R01 DA057790

Title: Delta opioid receptor bitopic ligand C6-Quino produces biased partial agonism, safer analgesia, and reduces voluntary ethanol consumption

Authors: *J. P. MCLAUGHLIN¹, S. EANS¹, S. MURPHY¹, S. TOURNESY¹, B. VARGA², S. BERNHARD², V. KATRITCH³, T. CHE⁴, S. MAJUMDAR⁵;

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Abstract: Aims: Delta opioid receptors (DOR) are known to regulate pain, mood and alcohol consumption. The therapeutic development of DOR full agonists for pain and alcohol abuse has been thwarted by related seizure activity. In contrast, DOR partial agonists, biased against arrestin signaling, reportedly lack seizures while retaining therapeutic efficacy. However, partial agonism is poorly understood. Whereas agonists activate GPCR signaling by interacting with conventional orthosteric binding sites, ligands interacting at putative allosteric binding sites modulate the activity of agonists. High-resolution crystal structures of several inactive-state Family A GPCRs reveal a sodium binding pocket centered around the highly conserved D2.50 residue of the second transmembrane domain which may be responsible for allosteric modulation. Guided by new insights of DOR structure and signaling obtained by structure-aided design, we designed and explored the functional selectivity of bitopic ligand C6-Quino to simultaneously target the classical orthosteric and allosteric site of the DOR. We then examined if C6-Quino acted as a DOR partial agonist, producing analgesia and reducing alcohol consumption without drug-induced liabilities like seizures. Methods/Results: Optimal interactions with both sites were validated by cryo-electron microscopy for DOR-selective C6-quino. In cellular in vitro testing, C6-Quino displayed potent G-protein partial agonism with reduced arrestin signaling. When tested in vivo in mice, C6-quino demonstrated dose-dependent, non-maximal antinociception in the 55°C warm-water tail-withdrawal assay, with an ED₅₀ (and 95% confidence interval) value of 12.0 (9.14-16.4) mg/kg s.c. mediated by the DOR. C6-Quino further demonstrated dose-dependent anti-allodynia mediated by the DOR in the chronic constriction injury (CCI) assay of neuropathic pain. Unlike morphine, C6-Quino demonstrated no respiratory depression or conditioned place preference, and whereas conventional DOR agonist SNC80 produced robust seizures, the DOR-selective bitopic ligand C6-Quino showed no seizure activity. Finally, in preliminary testing, this DOR partial agonist reduced voluntary ethanol consumption in a two-bottle choice (TBC) assay in a dose-dependent manner. Conclusions: Engaging both the orthosteric and allosteric site leads to a unique G-protein and arrestin recruitment profile, with C6-Quino displaying biased partial agonism and in vivo antinociceptive efficacy against multiple types of pain without adverse seizures typical of DOR full agonists, and reduced voluntary ethanol consumption.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Program #/Poster #: PSTR278.06/L13

Topic: E.01. Somatosensation – Pain and Itch

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R01 NS134647-01A1

Title: Novel therapeutic actions of LSD1 inhibition for chronic pain in sickle cell disease

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Abstract: Sickle cell disease (SCD) is an inherited blood disorder with debilitating pain as its hallmark feature. Previous studies indicated that inhibition of lysine-specific demethylase 1 (LSD1) induced fetal hemoglobin (HbF) production and reduced disease pathology in a mouse model of SCD. The aim of this study was to determine whether LSD1 inhibition could alleviate persistent chronic pain in SCD. In a targeted knock-in mouse model of sickle cell anemia, we characterized ongoing spontaneous pain, as well as evoked hypersensitivity to mechanical and thermal stimuli using the conditioned place preference (CPP) paradigm, von Frey filaments assessment, and Hargreaves radiation heat test, respectively. Chronic treatment with RN-1 (2.5 mg/kg/day, *i.p.*, 10 days), a selective irreversible inhibitor of LSD1, completely abolished clonidine-elicited chamber preference in SCD mice carrying human sickle hemoglobin, indicating the effective reduction of spontaneous affective pain component associated with SCD. Meanwhile, chronic RN-1 treatment significantly attenuated mechanical allodynia and heat hyperalgesia in mice with SCD, but not non-sickle control littermates. Moreover, acute spinal administration of RN-1 (10 nmol, *i.t.*) was effective in reversing evoked hypersensitivity in SCD mice without affecting baseline sensitivities in control mice. Furthermore, a single trial conditioning with RN-1 produced CPP in SCD mice similar as clonidine, implying the anti-nociceptive effect of RN-1 may not rely on HbF induction. These findings suggest the functional participation of LSD1 in the development of both non-evoked ongoing pain and evoked pain in SCD, which highlighted the possibility of a new intervention target to treat chronic pain in SCD.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Title: Convergent state-control of endogenous opioid analgesia

Authors: *B. A. KIMMEY¹, L. EJOH¹, L. SHANGLOO¹, J. WOJICK¹, S. NASSER CHEHIMI¹, N. M. MCCALL¹, C. OSWELL¹, M. MAHMOOD¹, L. YANG², V. K. SAMINENI², K. DEISSEROTH³, R. CRIST¹, B. C. REINER¹, L. TIAN⁴, G. CORDER¹;
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Abstract: Pain is a dynamic and nonlinear experience shaped by injury and contextual factors, including expectations of future pain or relief. Increasing evidence suggests that the midbrain periaqueductal gray plays a central role in endogenous analgesia and pain expectancy as the ventrolateral column (vLPAG) receives ascending nociceptive signals from the periphery and descending pain representations arising in forebrain cortical areas. Neurons expressing the μ opioid receptor (MOR+) and the MOR ligand enkephalin (Enk) are crucial to the nociceptive and pain-relieving functions of the vLPAG. However, significant gaps remain in our understanding of the vLPAG cell-type identities, the sub-second neural dynamics involved in pain modulation, the contribution of endogenous peptide neuromodulators, and the contextual factors influencing these processes. We hypothesized that opioidergic signaling dynamics in the vLPAG represent injury- and expectancy-induced pain states that can be targeted for on-demand pain relief. To this end, we used spatial mapping with RNA sequencing of pain-active neurons, alongside activity-dependent and cell type-specific genetic tools for *in vivo* optical recordings and modulation of neural activity in mice, to identify pain state-related opioidergic signaling in vLPAG. TRAP labeling and Projection-TAGging revealed that fore- and hindbrain projecting MOR+ glutamatergic vLPAG neurons are recruited during acutely nociceptive states, while their real-time calcium activity is modulated by opioid agonists and pain behavior. Enk-releasing afferents densely innervate vLPAG, arising from, among other sources, local neurons. In contrast to the activation of MOR+ neurons, we observed acute pain-related suppression of Enk release detected with the δ Light sensor. Interestingly, protracted inflammatory pain, learned predictions of pain onset, and expression of placebo analgesia all produced tonic elevations of Enk. By leveraging the functional effects of pain relief expectation, we used direct optogenetic activation of vLPAG Enk-expressing neurons to drive opioid peptide release onto MOR+ neurons, resulting in a robust reduction in pain. These findings show that diverse biological and contextual factors converge on shared midbrain circuitry that releases endogenous opioids to suppress nociceptive activity and promote analgesia. Harnessing vLPAG Enk signaling offers a promising avenue for future pain therapies that achieve effective pain relief without the harmful side effects associated with prescribed opioid analgesics.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR278.08/L15

Topic: E.01. Somatosensation – Pain and Itch

Title: Peripheral neuroimmune drivers of opioid-induced hyperalgesia

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Abstract: The use of opioids in pain management is limited by adverse side effects, including opioid-induced hyperalgesia. The neuroinflammatory effects of opioids are one pathway that is known to generate hyperalgesia, but the mechanisms through which this inflammation is generated is unclear. Notably, opioid receptors are found throughout the body, and in the periphery are expressed on enteric neurons, where they disrupt gastrointestinal motility and the gut microbiome, as well as directly on immune cells and sensory neurons. Consequently, the peripheral actions of opioids may represent a novel route through which opioid-induced inflammation can be generated and sustained. To test this, male and female C57Bl6J mice were treated with escalating doses of morphine (10-40 mg/kg, i.p.) for 4 days. Pain behaviour, spinal tissue collection, and 16S rRNA sequencing of fecal samples was performed 1- 28 days after morphine treatment to assess withdrawal syndromes, alongside measurements of gut motility and permeability. Single-cell RNA sequencing of mouse spinal cords was used to interrogate transcriptomic changes associated with withdrawal. Centrally- or peripherally-restricted opioid administration was used to determine the contributions of opioids in each compartment, while germ-free (GF) mice were used to assess the role of the gut microbiome. We found that peripheral, but not central, opioid administration was sufficient to produce hyperalgesia in mice. Both acute and prolonged opioid withdrawal produced morphological and transcriptomic changes in spinal microglia, and peripherally-restricted opioid administration alone was sufficient to recapitulate the alterations in microglial morphology observed with systemic opioids. Microbiome sequencing revealed that opioid withdrawal induced significant alterations in microbial diversity that persisted for up to 1-week post-drug and correlated with the expression of hyperalgesia, neuroinflammation, and impaired gut motility and permeability. However, microbiota transfer from opioid-withdrawn mice to GF donors was not sufficient to produce hyperalgesia, indicating that the altered composition of the gut microbiome itself does not generate increased hypersensitivity. These results suggest that opioid withdrawal produces profound changes in the periphery, which are sufficient to drive hyperalgesia.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Program #/Poster #: PSTR278.09/M1

Topic: E.01. Somatosensation – Pain and Itch

Support: R37 DA039997
CDMRP W81XWH-21-1-0771
T32 TR004764

Title: The Mu-Opioid Receptor Modulator BMS-122 Enhances the Anti-Allodynic Action of Opioid and Non-Opioid Analgesics in Neuropathic Pain

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Abstract: *Motivation:* Opioid therapy for chronic pain is effective but carries severe risks, particularly as efficacy decreases and duration of treatment extends. Adjunct medications to enhance opioid analgesia without enhancing adverse effects could provide a useful method for opioid sparing. Positive allosteric modulators (PAMs) interact with agonist-occupied receptors to increase agonist affinity and/or signaling. BMS-122 is a PAM of the μ -opioid receptor and has been shown to enhance opioid agonists *in vitro*. This molecule also enhances opioid activity *in vivo* without increasing negative effects like constipation, respiratory depression, and reward. However, these studies were performed in tests of acute antinociception. Our objective is to determine the capacity of BMS-122 to enhance opioids in chronic neuropathic pain and to potentially enhance the activity of non-opioid analgesics. *Methods:* Studies were conducted in male and female Sprague-Dawley rats with spared nerve injury (SNI). Rats developed tactile allodynia as measured with von Frey filaments. Animals received a 15-minute pretreatment of BMS-122 (10 mg/kg, s.c.) or vehicle, followed by various doses of analgesics. Tactile thresholds were measured over 2 hours. *Results:* Various doses of the opioids (R)-methadone (0.1, 0.32, or 1 mg/kg, s.c.) or morphine (1, 3.2, or 10 mg/kg, s.c.) were tested after pretreatment by BMS-122. Doses of opioid with minimal effects on allodynia were enhanced by pretreatment with BMS-122 to a similar or greater extent than a fully efficacious dose. To test more clinically relevant therapeutics, we repeated the study using the opioid partial agonist buprenorphine (0.01, 0.032, or 0.1 mg/kg, s.c.) or the non-opioid analgesic gabapentin (30 or 100 mg/kg). BMS-122 enhanced the action of even the highest dose of buprenorphine, supporting the capacity for PAMs improve the efficacy of partial agonists. In addition, BMS-122 enhanced the activity of gabapentin at a low dose to that of a fully efficacious dose. Gabapentinoids and opioid drugs synergize in nerve injury, and these data suggest a possible interaction between gabapentin and

endogenous opioid peptides. *Conclusion:* Overall, our data suggest that opioid PAMs may rescue opioid analgesia in neuropathic pain. These molecules could provide opioid sparing or serve as adjuvants to non-opioid analgesics. These studies were supported by R37 DA039997 (J.R. Traynor), DOD CDMRP W81XWH-21-1-0771 (S.W.P. Kemp), and T32 TR004764 (B.M. Clements).

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Topic: E.01. Somatosensation – Pain and Itch

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Title: Meningeal regulatory T cells gate nociception through an endogenous opioid circuit

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Abstract: T cells have emerged as orchestrators of pain chronification, but the mechanism by which T cells control pain sensitivity is unresolved. Here, we demonstrate an influence of regulatory T cells (Tregs) on pain processing that is distinct from their canonical functions of immune regulation and tissue repair. Site-specific depletion or expansion of meningeal Tregs (mTregs) leads to profound female-selective and sex hormone-dependent modulation of mechanical hypersensitivity. Specifically, mTregs express the endogenous opioid enkephalin that exerts an antinociceptive action through the delta opioid receptors expressed by MrgprD⁺ sensory neurons. Although enkephalin restrains nociceptive processing, it is dispensable for Treg-mediated immunomodulation (Midavaine et al., 2025 *Science*). Notably, the mTreg modulation of pain thresholds depends on sex-hormones and expansion of enkephalinergic mTregs during gestation imparts a remarkable pregnancy-induced analgesia in a pre-existing, chronic, unremitting neuropathic pain model. These results uncover a fundamental sex-specific, pregnancy-pronounced, and immunologically-derived endogenous opioid circuit for nociceptive regulation with critical implications for pain biology and maternal health.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Program #/Poster #: PSTR278.11/M3

Topic: E.01. Somatosensation – Pain and Itch

Support: Howard Hughes Medical Institute (HHMI)
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Title: Investigating epidermal signaling mechanisms in a mouse model of Atopic Dermatitis

Authors: *L. N. MURPHY¹, M. BUDRON¹, C. COOK¹, Y. SCOTT², E. A. LUMPKIN¹, D. M. BAUTISTA²;

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Abstract: Chronic itch is a debilitating condition that affects approximately 20% of individuals during their lifetime.¹ Many chronic itch disorders, such as Atopic dermatitis (AD), initiate in the skin where signaling between peripheral sensory neurons, skin epithelial (epidermal) cells, and infiltrating immune cells occurs. Epidermal cell-derived inflammatory and excitatory signaling molecules play an important role in both the initiation and progression of AD. However, the molecular signaling mechanisms by which epidermal cells release these molecules are not well understood in AD. Thus, we used a genetic mouse model to selectively inhibit conical SNARE-mediated vesicle release from epidermal cells by expressing tetanus neurotoxin (TeNT) under keratin 14 (K14) cre promoter (K14^{cre};R26^{TeNT/+} mice). To induce AD, MC903 was applied for seven consecutive days to the shaved cheek of 8-14 week old male and female littermates. Itch behavior was scored for 20 minutes blinded to treatment and genotype from videos taken on day 8. We photographed MC903-treated skin to record lesion severity then collected the skin for flow cytometry to measure immune cell infiltration. Lesion severity was assessed for redness, dryness, and scabbing with scorers blinded to genotype and treatment. We found MC903-treated K14^{cre};R26^{TeNT/+} mice had diminished itch behaviors compared to MC903-treated littermate controls. However, lesion severity is not dependent on epidermal vesicle release. Moreover, epidermal vesicle release is required for recruiting specific immune cell populations to the skin. In order to determine which population of skin cells are mediating these effects, we utilized mice that lack Merkel cells, a specialized subset of epidermal cells (K14^{cre};Atoh1^{LacZ/Fl} mice). MC903-treated mice lacking Merkel cells show a significant decrease in time spent scratching compared to MC903-treated littermate controls with no difference in average bout length or latency to first scratch bout. Together, our data suggest that epidermal cells use TeNT-dependent vesicle release to modulate itch and facilitate immune cell recruitment, as well as indicate that Merkel cells are required for itch in the MC903 mouse model of AD. Reference: ¹ Butler, D. C. et al. Chronic pruritus: A review. JAMA 331, 2114-2124 (2024).

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Topic: E.01. Somatosensation – Pain and Itch

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Title: Lateral hypothalamus directs stress-induced modulation of acute and psoriatic itch

Authors: *J. PRAJAPATI¹, A. BARIK², G. SAHU⁴, A. HOQUE³, M. PATTANAYAK¹;
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Abstract: Stress and anxiety are well-known modulators of both physiological and pathological itch. Acute stress suppresses itch, while chronic stress exacerbates it. These effects are mediated by neural circuits within the brain, though the precise mechanisms remain poorly understood. In this study, we investigate the role of stress-sensitive lateral hypothalamic area (LHA) neurons in modulating itch. Using neural activity-dependent genetic labeling and chemogenetic tools, we selectively engaged a population of LHA neurons (LHA^{stress-TRAP} neurons) responsive to stress. Transient stimulation of these neurons induced anxiety-like behaviors, conditioned place aversion, and suppressed acute (chloroquine-induced) and chronic (psoriatic) itch. Conversely, the inhibition of the LHA^{stress-TRAP} neurons enhanced acute and chronic itch. Interestingly, LHA^{stress-TRAP} neurons did not respond to acute itch stimuli, but their activity was temporally correlated with scratching episodes in mice with psoriasis. Ex vivo whole-cell patch-clamp recordings revealed that these neurons exhibit heightened excitability in psoriatic animals. Anterograde viral tracing demonstrated that LHA^{stress-TRAP} neurons project to brainstem regions implicated in itch modulation, including the periaqueductal gray (PAG), rostral ventromedial medulla (RVM), and lateral parabrachial nucleus (LPBN). Furthermore, chemogenetic activation and optogenetic silencing of LHA^{stress-TRAP} axon terminals revealed that bidirectional modulation of itch is primarily mediated through projections to the PAG. Together, these findings identify a previously unrecognized central mechanism by which stress modulates itch, centered on a specific population of LHA neurons and their downstream brainstem targets.

Disclosures: **J. Prajapati:** A. Employment/Salary (full or part-time): Centre for Neuroscience, Indian Institute of Science, Bengaluru, India, Pin-560012. **A. Barik:** A. Employment/Salary (full or part-time): Centre for Neuroscience, Indian Institute of Science, Bengaluru, India, Pin-560012. **G. Sahu:** A. Employment/Salary (full or part-time): Molecular Biophysics Unit, Indian Institute of Science, Bengaluru, India, Pin-560012. **A. Hoque:** A. Employment/Salary (full or part-time): Molecular Biophysics Unit, Indian Institute of Science. **M. Pattanayak:** A. Employment/Salary (full or part-time): Indian Institute of Science.

Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Program #/Poster #: PSTR278.13/M5

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH/NINDS R01;NS125413

Title: The participation of endocytic mediators to itch transmission

Authors: ***M. DE AMORIM FERREIRA**¹, R. TONELLO², D. D. JENSEN³;

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Abstract: Pruritus, or chronic itch, significantly impacts the quality of life for one in four adults and is associated with increased anxiety, depression, and suicide. Despite its widespread burden, the precise mechanisms driving chronic itch remain poorly understood. Neurotransmitter release via synaptic vesicles (SVs) regulate neurotransmission and play a key role in itch transmission. SV exocytosis releases neurotransmitters to propagate signals and SV endocytosis retrieves vesicles from the presynaptic membrane. SV movement is governed by key mediators like clathrin, adaptor associated kinase 1 (AAK1), and Dynamin (Dnm). Previous studies have shown that Dnm and AAK1-mediated endocytosis at nociceptor terminals (pain-sensing neurons) and can be targeted to block pain. In this study, we hypothesize that endocytic mediators regulate synaptic transmission in itch pathways and can be targeted to alleviate itch. C57BL/6 mice (n=8/group, 4 male, 4 female) were treated with siRNAs (1.25 µg/5 µL intrathecally at cervical region), targeting AAK1, Dnm1, or Dnm3 to knockdown SV regulators. After 48 hours, itch was induced by intradermal chloroquine injection (10 mM/10µL), and scratching behavior, locomotor activity, and anxiety-like behaviors were quantified for 30 minutes. The effectiveness of the siRNA knockdown was confirmed using RNA Scope in situ hybridization in spinal cord and DRG (n=4/group). This study also assessed the effect of pharmacological inhibition on these behaviors using the AAK1 antagonist LP935509 (10µg/5µL) and the dynamin antagonist Dyngo4a (50nM/5µL). AAK1 siRNA significantly reduced scratching behavior by 48% (± 5.73) without affecting locomotor or anxiety-like behaviors. This reduction correlated with confirmed *Aak1* mRNA knockdown in both the spinal cord (72% ± 9.23) and DRG (50% ± 2.36). The AAK1 inhibitor, LP935509, also decreased scratching by 62% (± 4.27) compared to the control

group. Similarly, targeting Dnm1 and Dnm3 with siRNA reduced scratching by 46% (± 10.65) and 69% (± 3.83). No significant changes in locomotor activity or anxiety-like behaviors occurred for these treatments. Knockdown of Dnm1 and Dnm3 was observed exclusively in the DRG ($57.01\% \pm 7.42$ for Dnm1 and $52.84\% \pm 5.05$ for Dnm3). The collective knockdown of Dnm1, 2, and 3 by combined siRNA injection resulted in a substantial reduction of scratching by 80% (± 4.38). This effect was mirrored by the dynamin antagonist Dyngo4a, which decreased scratching by 53% (± 4.30). These findings suggest that the disruption of synaptic transmission reverses itch behavior without affecting motor functions and provide new insights into the mechanisms regulating itch.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

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Topic: E.01. Somatosensation – Pain and Itch

Support: NIH R01AR074062
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Title: Characterization of itch-responsive neurons which project from the PBN to the Amygdala

Authors: *D. PAVLENKO¹, H. ISHIDA², T. AKIYAMA³;

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Abstract: The parabrachial nucleus (PBN) is known as a “relay station” of the brain because it receives sensory input from the spinal cord and sends it to other regions of the brain including the amygdala, thalamus, and periaqueductal gray. Both the PBN and the amygdala are involved in itch processing, but the precise neural pathways remain poorly understood due to their heterogeneous cell populations. Notably, a subpopulation of calcitonin gene-related peptide-positive (CGRP+) PBN neurons has been linked to itch, with their silencing reducing scratching behaviors. However, the specific projections of these CGRP+ PBN neurons involved in itch have not been identified. Using the targeted recombination in active populations (TRAP2) system, we were able to identify and activate itch-responsive neurons which project from the PBN to the amygdala. Optogenetic activation of these neurons significantly increased spontaneous scratching, but it did not alter pruritogen-induced scratching, anxiety-like behaviors, thermal sensitivity, or mechanical sensitivity. Additionally, in atopic dermatitis like chronic itch model, inhibiting these itch-responsive neurons significantly reduced scratching bouts, but no change in locomotion or anxiety-like behavior. To characterize the itch-responsive neurons, we used a combination of TRAP2, retrograde tracing, and immunohistochemistry. First, we captured the itch-responsive neurons by pairing an i.p. 4-hydroxytamoxifen injection with an i.d. serotonin

injection in the nape of the neck of a mouse which induces expression of tdTomato in those cells. Next, we injected cholera toxin subunit B (CT-B), a retrograde tracer, into the central amygdala (CeA). We analyzed the overlap between CGRP⁺ cells and TRAPped cells, CT-B⁺ cells and TRAPped cells, and cells that were positive for all three. Our analysis revealed that CGRP⁺ cells were only found in the external lateral area of the PBN. Additionally, only 8% of the CGRP⁺ cells were TRAPped, and of those, 72% projected to the CeA. Of all the TRAPped cells in the PBN, 17% of them projected to the CeA. These findings suggest that the itch-related PBN-amygdala pathway comprises heterogeneous neurons with a small percentage being CGRP⁺.

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Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

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Topic: E.01. Somatosensation – Pain and Itch

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Title: Intracellular Signaling of Mas-related G-protein Coupled Receptor X2 (MrGPRX2) And Its Role in Transient Receptor Potential Vanilloid (TRPV) Sensitization in Itch Pathways

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Abstract: G protein-coupled receptors (GPCRs) and TRPV channels are fundamental to signal transduction in sensory processes such as pain and itch, where the activation of GPCRs often results in the sensitization of TRPV channels expressed on the same neurons. While canonical GPCR signaling occurs at the cell surface, compartmentalized signaling from intracellular organelles is increasingly recognized to regulate cellular responses and itch and pain sensation. We explored this phenomenon for human Mas-related G-protein coupled receptor X2 (MrGPRX2), a pruritus-linked GPCR without prior evidence of intracellular signaling. Our study reveals that the biased agonism of MrGPRX2 regulates receptor trafficking and the ability of MrGPRX2 to signal from endosomes. Immunofluorescence analysis showed that MrGPRX2 internalizes following Substance P (SP) stimulation, but not BAM8-22. BRET assays demonstrated SP-induced Gαq protein recruitment at both the plasma membrane and early endosomes. Compartmentalized FRET biosensor studies showed that SP induced, endosomal MrGPRX2 signaling regulates nuclear ERK activation. Pharmacological and genetic inhibition of endocytosis prevented SP-induced MrGPRX2 internalization and downstream ERK activation. Furthermore, calcium imaging revealed MrGPRX2-mediated TRPV sensitization specifically after SP stimulation. These findings unveil a crucial role for MrGPRX2 endosomal

signaling and TRPV sensitization, offering valuable insights into itch mechanisms for the development of targeted therapeutic interventions to treat chronic itch.

Disclosures: P. Duran: None. D.D. Jensen: None.

Poster

PSTR278: Pain and Itch: Opioid and Non-opioid Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR278.16/M8

Topic: E.01. Somatosensation – Pain and Itch

Support: NIH Grant NS111643

Title: Touch signaling maintains cell survival in the somatosensory dorsal horn

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Abstract: Touch information performs diverse functions in somatosensation, including but not limited to the discrimination of shapes and textures and the modulation of itch and pain. In addition, it directs the morphogenesis of sensory structures in the skin and the wiring up of central circuits during development. Here, we demonstrate a novel role for touch signaling in the maintenance of neuronal survival in the somatosensory dorsal horn of adult mice. Chronic abrogation of Merkel cell signaling in young adult (P56) *Atoh1*^{CreER}; *Piezo2*^{fl/fl} mice results in the loss by apoptosis of molecularly defined subsets of both excitatory and inhibitory spinal neurons that constitute circuits for somatosensory processing. Affected populations include neuropeptide Y (NPY)-expressing inhibitory neurons that have been shown to gate itch and pain signaling. We show that as a consequence of a substantial reduction in NPY⁺ neurons, mice exhibit increased rates of spontaneous and mechanically evoked itch, as well as elevated inflammatory pain responses. We go on to demonstrate that the degeneration of Merkel cells that occurs during normal aging in wild type mice results in substantial cell loss in the dorsal horn, with the selective loss of NPY⁺ neurons but not of Merkel cells per se accounting for the elevated sensitivity of geriatric mice to itch and pain stimuli. Finally, we provide evidence that cell survival in the dorsal horn of adult mice is maintained in part by BDNF-TrkB signaling. Sensory dysfunction is one of the most striking characteristics of aging and has a major impact on quality of life, with the incidence of chronic itch and pain conditions increasing as animals progress into old age; however, the central circuit mechanisms underlying these changes have not previously been described. Our data support a novel mechanism whereby somatosensory circuits are reorganized during normal aging in an activity-dependent manner, exacerbating itch and pain.

Disclosures: D. Acton: None. S. Pimpinella: None. M.D. Goulding: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR279.01/M9

Topic: E.03. The Chemical Senses

Support: NSF CAREER 2238686
NSF Graduate Research Fellowship

Title: Classification of multiple per- and polyfluoroalkyl substances (PFAS) at environmentally relevant concentrations

Authors: *S. MCLANE-SVOBODA¹, C. STOUT², A. MCLANE-SVOBODA¹, D. SAHA¹;
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Abstract: Per- and polyfluoroalkyl substances (PFAS) are becoming increasingly widespread worldwide due to their persistence and resistance to environmental degradation. As a result, these compounds tend to bioaccumulate and have been associated with various health issues, posing a threat to human well-being. Recently, there has been a growing demand for more sensitive and efficient PFAS detection methods. While current technologies such as liquid chromatography-mass spectrometry (LC-MS) and molecularly imprinted polymers (MIPs), have been used for PFAS detection, they often struggle to accurately identify these substances at environmentally relevant concentrations (e.g., parts per trillion [ppt]) and exhibit limited detection ranges. This underscores the urgent need for improved PFAS detection devices. Chemical sensing in biological olfactory systems, particularly insects (locust), has evolved extensively across the animal kingdom, demonstrating exceptional sensitivity to a wide range of compounds. Locust antennae contain olfactory receptor neurons (ORNs) that detect volatile chemicals through combinatorial coding, allowing the insect to process trillions of odorants. When odor molecules bind to ORNs, they generate electrical signals (i.e., neural spikes) that are transmitted to the antennal lobes (ALs), where excitatory and inhibitory neurons refine the signals before relaying to higher-order brain centers. Using spatiotemporal coding, the locust's olfactory system generates unique neural fingerprints for each odorant and its concentration, enabling highly sensitive and specific compound detection at ppt concentrations. By leveraging biological olfaction as a novel gas sensor, the limitations of current PFAS detection methods can be overcome. Here, we used biological olfaction for PFAS detection by conducting in-vivo extracellular neural recordings from locust ALs and analyzing odor-evoked neural responses. Multi-channel electrodes were employed to record neural activity in response to eight PFAS compounds (e.g., PFOS, 6:2 FTOH, 8:2 FTOH, 6:2 FTAc, MeFOSE, EtFOSE, MeFOSA, PFOSA) at varying concentrations. Selected PFAS were serially diluted to 0.01% and 0.0001% to simulate environmental concentrations (e.g., ppt). We achieved a high classification success rate for all stimuli and concentrations. Distinct spatiotemporal differences were observed in neural response to varying concentrations of the same PFAS compound, indicating unique information encoding for compound identification and concentration. Our results demonstrate the first biological olfaction based PFAS sensor.

Disclosures: S. McLane-Svoboda: None. C. Stout: None. A. McLane-Svoboda: None. D. Saha: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR279.02/M10

Topic: E.03. The Chemical Senses

Support: NSF Grant 2323240
NSF Graduate Research Fellowship
NIH 1R01DC020892

Title: Neural basis of associative learning and generalization in locust olfaction

Authors: *A. MCLANE-SVOBODA¹, S. MCLANE-SVOBODA², S. JOSHI³, M. V. BAZHENOV⁴, D. SAHA²;

²Biomed. Engin., ¹Michigan State Univ., East Lansing, MI; ³Univ. of California San Diego, San Diego, CA; ⁴Dept. of Med., UCSD, La Jolla, CA

Abstract: Associative learning enables organisms to generalize information and adapt behavior based on reinforced experiences. While some organisms require extensive repeated exposures to form associations, others, such as locust, learn and generalize information from only a few interactions. Locusts can associate specific odors with food, generalizing learned information across different stimulus conditions. This ability stems from the complexity of the locusts' olfactory system, where a plastic network of projection neurons (PNs) and local neurons - the Antennal Lobe (AL) - is engaged in early odor information processing. Engineered systems have yet to replicate the ability to generalize stimuli across various concentrations and contexts. By studying the locust olfactory system, we can uncover associative learning mechanisms, leveraging them to develop novel machine learning models with similar generalization capabilities. Here, we address how the AL neuronal activity changes to a target odor once associated with a reward and how that learned association changes the neural response to other untrained odors. Odor-evoked AL responses were collected from neurons before and after associative learning. Minimally invasive surgery exposed the ALs for electrode insertion while retaining basic motor functions (palp movement) for training. Five odors were presented to the locust: a target odor, a chemically similar odor to the target, a perceptually similar odor, an odor control, and experimental control. Baseline odor-evoked activity was recorded by presenting odors to the locust before training. Associative training consisted of a sucrose solution reward during target odor presentation for six trials. After training, the odors were re-presented to the locust. The electrode maintained its original position in the AL throughout the entire experiment. Individual neurons were analyzed to identify differences in odor-evoked response pre- and post-training. Using the population of neurons collected, we have uncovered neural activity changes for specific odors based on chemical and perceptual similarity to the target odor. Interestingly,

trends in neural activity differ throughout odor presentation, highlighting distinct responses for odor onset (transient and steady state) vs offset. These results demonstrate the locusts' ability to quickly learn and generalize information over chemical and perceptual structure and give insight into how responsiveness changes during stimulus evolution.

Disclosures: **A. McLane-Svoboda:** None. **S. McLane-Svoboda:** None. **S. Joshi:** None. **M.V. Bazhenov:** None. **D. Saha:** None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR279.03/M11

Topic: E.03. The Chemical Senses

Support: NIH Grant 1R01DC020892
NSF Grant 2223839
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NSF Grant 2323240

Title: Distinct roles of inhibitory motifs in odor discrimination: Insights from an RNN model of the locust antennal lobe

Authors: ***S. JOSHI**¹, G. NAVAS², A. MCLANE-SVOBODA⁴, D. SAHA⁴, M. V. BAZHENOV³;

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Abstract: Inhibitory circuits in the first relay of the insect olfactory system - the antennal lobe (AL) - shape olfactory representations through complex interactions between inhibitory local neurons (LNs) and excitatory projection neurons (PNs). While inhibition is crucial for odor coding, how different inhibitory motifs contribute to circuit dynamics and function remains poorly understood. Here, we systematically manipulated inhibitory connections in a biologically constrained RNN model of the locust AL (830 PNs, 300 LNs), trained on in vivo data, to investigate how LN-PN and LN-LN inhibition differentially regulate olfactory processing. Mean PN firing rate proved remarkably stable across a wide range of global inhibition, revealing an antagonistic balance between direct LN-to-PN inhibition and LN-mediated disinhibition. Inhibition strength, however, exhibited a non-linear relationship with coding dimensionality: weak inhibition compressed neural trajectories into a low-dimensional space driven by receptor neurons activity while preserving stimulus decodability, whereas strong inhibition expanded the coding space but severely impaired discrimination accuracy. Motif-specific manipulations further revealed functionally distinct effects. Increasing mutual inhibition between LNs (LN-to-LN) elevated response dimensionality but negatively impacted information coding. In contrast, amplifying feedforward inhibition (LN-to-PN) suppressed both firing rates and AL response

dimensionality, demonstrating direct control over network activity. Temporal analysis revealed that stimulus onset responses remained relatively robust to inhibitory perturbations, while late-phase responses showed marked vulnerability. The system exhibited a transition at intermediate inhibition levels, beyond which both coding capacity and classification accuracy degraded catastrophically, suggesting that the AL operates near a critical regime. These findings reveal fundamental principles of inhibitory circuit design: the AL operates in a regime where inhibition balances high-dimensional representations facilitating stimulus discriminability with accurate stimulus encoding. While increasing LN mutual inhibition can further expand coding dimensionality, it does so at the cost of encoding accuracy. This work demonstrates how data-driven computational models can uncover circuit-level mechanisms underlying neural coding.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

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Program #/Poster #: PSTR279.04/M12

Topic: E.03. The Chemical Senses

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Dr. Heinz Lux-Stiftung (SP 2023/12-01)

Title: Olfactory integration in distinct amygdaloid nuclei

Authors: *M. M. NESSELER, L. BÜSCHING, M. SPEHR;
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Abstract: The central olfactory system comprises at least two neural circuits, i.e., the main and accessory olfactory pathways. Both rely on transduction of chemical stimuli in separate peripheral sensory organs and the integration of this sensory information in the main and accessory olfactory bulbs, respectively. Within both primary processing stages, the main and accessory olfactory pathways remain separated. Here, we ask whether the main and accessory olfactory pathways converge at downstream nuclei. We use Cre-dependent expression of stereotactically injected rAAV genomes to label and trace principal neurons of the main or accessory olfactory bulbs. Notably, we identify both divergent and convergent projections. Next, we investigate the neurophysiological composition of amygdaloid nuclei that receive convergent input. We use single-cell patch-clamp recordings in acute brain slices to describe the electrophysiological and morphological properties of neurons within these nuclei. We identify both unique and shared electrophysiological properties among these neurons. Additionally, we provide comprehensive insights into individual neuron morphological characteristics. In

summary, we describe convergence of the main and accessory olfactory pathways and characterize the electrophysiological and morphological properties of neurons within shared amygdaloid target nuclei.

Disclosures: M.M. Nessler: None. L. Büsching: None. M. Spehr: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

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Program #/Poster #: PSTR279.05/M13

Topic: E.03. The Chemical Senses

Support: DFG Grant HE 9996/1-1
ERS Grant (DE-82)EXS-SF-SFBSS006

Title: A cellular atlas of the mouse accessory olfactory bulb: where pheromones meet the brain

Authors: *A. HERNANDEZ-CLAVIJO¹, U. RANGASWAMY², R. SANGES², M. SPEHR¹;
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Abstract: Chemical communication plays a crucial role in regulating behavior in most mammals, including mice. Pheromones and other socially relevant chemical signals are primarily detected and processed by the accessory olfactory system (AOS). The AOS consists of the vomeronasal organ (VNO), which serves as the sensory receptor, and the accessory olfactory bulb (AOB), which receives input from the VNO via the vomeronasal nerves. Located in the dorsal-posterior region of the olfactory bulb, the AOB processes pheromonal information and transmits it directly to the amygdala-hypothalamic complex, a key center for controlling behavior and internal physiological states. Despite its pivotal role in pheromone processing and behavioral regulation, the cellular composition and functional organization of the AOB remain largely unexplored. In this study, we employed a multifaceted approach to investigate the cellular diversity of the AOB. We performed single-cell transcriptomic analysis in both male and female mice, generating a robust dataset that captures the cellular heterogeneity of this brain region. To spatially contextualize gene expression, we applied in situ spatial profiling techniques to map the distribution of hundreds of marker genes at subcellular resolution. By integrating single-cell transcriptomic data with spatial gene expression maps, we revealed the spatial organization of distinct AOB cell populations. To further characterize individual cell types, we conducted patch-clamp recordings combined with high-resolution microscopy to assess their electrophysiological properties and morphological features. Post-hoc analysis of marker gene expression in the recorded cells enabled a comprehensive integration of transcriptomic, spatial, electrophysiological, and morphological data, resulting in a multidimensional cell atlas of the murine AOB. This study identifies previously uncharacterized neuronal subtypes within the AOB and presents one of the most comprehensive analyses of cellular diversity in this region to

date. Our findings provide a critical foundation for future research into the full cellular composition and functional circuitry of the mouse AOB, deepening our understanding of its role in pheromone processing and the regulation of social behavior.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

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Program #/Poster #: PSTR279.06/M14

Topic: E.03. The Chemical Senses

Support: NIH Grant DC015525

Title: Semilunar cells in the mouse piriform cortex are necessary for the acquisition, but not retrieval, of odor memories

Authors: *M. JACOB¹, S. NAGAPPAN², E. WEINGARDT², K. M. FRANKS²;
²Neurobio., ¹Duke Univ., Durham, NC

Abstract: How and where memories are acquired, stored, and retrieved is a fundamental question in neuroscience. In the mammalian olfactory system, the piriform cortex (PCx), a sensory-associative cortex with extensive recurrent intracortical circuitry, is thought to be required for odor memory. To test this, we examined the role of distinct cell types in the acquisition, storage, and retrieval of odor memories. The PCx has two classes of principal excitatory neurons: semilunar cells (SLs) and pyramidal cells (PYRs), which differ in their connectivity. SLs receive strong input from the olfactory bulb but little to no recurrent excitation. PYRs also receive olfactory bulb inputs, along with extensive recurrent excitation from other PYRs and SLs, and top-down inputs from other brain regions. We used a classical conditioning paradigm in which mice were exposed to pseudorandom 10-second pulses of two odors (CS+ and CS-) over a 25-minute period, with a foot shock delivered at the end of each CS+. Seven to ten days later, we assessed odor memory in a two-chamber arena by measuring place preference before and during odor presentation. Control mice avoided the CS+ chamber when the odors were delivered. To evaluate the role of SLs in odor memory, we selectively eliminated them using NetrinG1-Cre mice which, in the PCx, express Cre exclusively in SLs. We first injected AAVs encoding the diphtheria toxin receptor into the PCx. SLs were then ablated by systemic administration of diphtheria toxin (DT). In one cohort, SLs were ablated before odor-shock pairing. In a second, SLs were eliminated after pairing but before memory testing. Eliminating SLs before odor-shock pairing impaired memory formation, as mice showed no preference for the CS+ or CS- chamber. In contrast, eliminating SLs after conditioning did not impair retrieval, as mice avoided the CS+ chamber. These data suggest that SLs are necessary for odor memory acquisition but not for their storage or retrieval. SLs constitute about half the principal neurons in

the PCx. To ensure that this deficit was specific to SL loss, we repeated these experiments in Emx1-Cre mice, which express Cre in both SLs and PYRs. By titrating AAV injections, we non-selectively ablated ~50% of PCx principal neurons. In these mice, DT administered before odor-shock pairing did not impair odor memory formation as they still avoided the CS+ chamber. Together, these results indicate that SLs are necessary for odor memory acquisition but not for storage or retrieval. Identifying which SL connections support memory acquisition and determining where and how odor memories are stored and retrieved are important next steps.

Disclosures: M. Jacob: None. S. Nagappan: None. E. Weingardt: None. K.M. Franks: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

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Program #/Poster #: PSTR279.07/M15

Topic: E.03. The Chemical Senses

Support: NIH Grant

Title: A novel head-fixed paradigm for dissecting neural mechanisms of odor localization

Authors: *S. DASTE, M. FOGLESONG, Z. YAO, K. M. FRANKS;
Duke Univ., Durham, NC

Abstract: Accurately localizing odors in space is a fundamental yet poorly understood function of the mammalian olfactory system. Unlike vision or touch, olfactory cues lack a spatially mapped receptive field, requiring the brain to infer location by comparing odor signals across space and time. The anterior olfactory nucleus (AON) is uniquely positioned within the olfactory circuit to support such computations: it is the only olfactory cortical region receiving strong bilateral input. This makes it a likely site for comparing nostril-specific information and mediating odor localization. However, progress has been limited by the inability to deliver spatially precise, repeatable odor stimuli while monitoring the rapid sensorimotor dynamics underlying spatial olfactory behaviors. To overcome these barriers, we developed a head-fixed behavioral paradigm in which mice were trained to lick an odorized water droplet delivered by a lick port attached to a motorized arm. On each trial, the spout was moved to different locations around the snout. After a variable delay (1-3 seconds), an odorized water droplet was delivered and the mouse could lick for a reward. Trials in which the mouse licked before the water droplet was presented were excluded. After one week of training, mice reliably learned to report droplet location by licking toward the water droplet. This paradigm minimizes the turbulence and temporal variability of traditional airborne stimuli, enables highly reproducible spatial odor delivery, and allows simultaneous tracking of respiration using integrated high-speed cameras. After the training period, we recorded spiking activity in the AON during task performance. We identified odor-responsive, location-selective neurons that responded more strongly to droplets presented on one side of the snout than the other, consistent with encoding stimulus location.

These neurons exhibited sharp response onsets as the odorized droplet emerged and showed clear differences between ipsilateral and contralateral presentations. Interestingly, other AON units exhibited pre-motor activity, with ramping activity that began before and peaked at the time of the lick. Unlike odor-responsive neurons, all premotor-like neurons identified thus far only responded when the mouse licked to the contralateral side. The presence of premotor-like signals suggests the AON may play a role in transforming spatial odor signals into directed motor output, consistent with its proposed role as a hub for integrating bilateral sensory information and guiding spatially targeted actions.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

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Topic: E.03. The Chemical Senses

Support: NIH Grant R01DC015525
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Title: Region-specific readout of temporally coded information in the olfactory system

Authors: *E. WEINGARDT¹, R. BLAZING³, K. M. FRANKS²;

²Neurobio., ¹Duke Univ., Durham, NC; ³Duke Univ., Duke Univ. Neurobio., Durham, NC

Abstract: Temporal codes are a hallmark of sensory signaling, yet how downstream circuits read out these codes can vary markedly between cortical areas. In the olfactory system, odors evoke spatiotemporal activity patterns in the olfactory bulb (OB), in which glomeruli respond to odors at distinct phases of the respiration cycle, forming odor-specific sequences. Previously, our lab has shown that neurons in the piriform cortex (PCx) are “phase-tuned” such that they selectively respond to glomerular activity occurring at a specific phase of the sniff. Phase tuning in the PCx is invariant to glomerulus identity, and across the population, the distribution of phase tuning preferences uniformly tiles the respiration cycle. It remains unclear, however, whether other olfactory cortical areas employ similar temporal tuning strategies. The anterior olfactory nucleus (AON) receives direct ipsilateral and contralateral OB input, yet little is known about how it represents the temporal structure of odor signals. To test this, we optogenetically stimulated glomerular-sized spots on unilateral dorsal OB during distinct phases of the respiration cycle while recording in ipsilateral lateral AON in awake, head-fixed mice. In contrast to the sharp and uniformly distributed phase tuning observed in PCx, AON neurons showed broader individual tuning curves, with preferred phases across the population concentrated during exhalation and early inhalation. That few AON neurons responded during the inhalation-exhalation transition suggests a suppression of excitability at this phase. In both regions, phase preferences were largely invariant to stimulation site, indicating that temporal

phase and glomerular spatial location input can be independently decoded. These findings reveal that PCx and AON extract different features from the same temporally structured input. The narrow and sharply defined phase tuning in PCx allows for high-resolution temporal discrimination, enabling precise encoding of glomerular-phase combinations. In contrast, that AON neurons are more broadly tuned and biased toward specific phases suggests a role in integrating input over extended portions of a single sniff. Thus, the PCx emphasizes fine temporal distinctions, responding selectively to brief, precisely defined phases of the sniff cycle. The AON, by contrast, integrates input over broader segments of the cycle, making it less sensitive to exact timing but potentially more robust to variability. This functional divergence suggests that PCx supports high-resolution temporal discrimination, whereas AON contributes to more stable, time-averaged representations of odor input.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

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Program #/Poster #: PSTR279.09/N1

Topic: E.03. The Chemical Senses

Support: Monell Chemical Senses Center - Startup Funding
NIH R01-DC020927
NIH T32-DC000014

Title: Age-related pattern separation deficits in olfactory bulb and piriform cortex

Authors: *I. F. CHAPMAN, K. A. BOLDING;
Monell Chem. Senses Ctr., Philadelphia, PA

Abstract: The risk of cognitive decline rises with age, driven by underlying changes in neural circuit function. Odor recognition memory impairments are early indicators of cognitive decline in both normal aging and Alzheimer's disease and are observed in humans and rodents. These symptoms are consistent with impaired pattern separation, a canonical neural circuit computation transforming overlapping sensory or synaptic inputs into distinct neural representations. Aging-associated failures of pattern separation are apparent in humans, monkeys, and rodents and across multiple sensory domains. The olfactory system presents a tractable model for studying how aging affects neural computations as sensory inputs reach cortical circuits through a short, two-step synaptic pathway from peripheral receptors. Pattern separation operations have been observed in both olfactory bulb (OB) and olfactory cortex (piriform cortex, PCx). To test whether olfactory pattern separation is affected by aging, we acquired acute neural population recordings simultaneously in OB and PCx of awake, head-fixed mice using Neuropixels 2.0 probes. We obtained recordings in young (~3-month, n=4) and aged (~13-month, n=3) B6 mice to investigate how olfactory processing is affected by aging. Four recording shanks are

positioned to span the dense principal cell layer in each structure, sampling multiple spatially separated populations (OB: 53 +/- 7.6; PCx: 273 +/- 15.9 stable, well-isolated units per recording), and enabling single-trial, within-animal characterization of neural population correlations during odor stimulation. We presented mixture stimuli composed of 8 monomolecular components, systematically varying overlap with a 'standard' mixture. In young mice, population correlations rose with overlap in OB and PCx. A linear SVM classifier showed increased latency and decreased accuracy distinguishing mixtures from standard at higher overlap levels (~200 ms latency shift from 7- to 4-component overlaps). Aged mice showed elevated correlations for overlapping mixtures, with corresponding effects on classifier latency and accuracy. Preliminary analyses suggest these specific deficits in stimulus separation may vary between the OB and PCx depending on stimulus overlap. Ongoing work will extend these observations to additional aging timepoints and use optogenetic identification of cell types in PCx to determine circuit-level mechanisms underlying altered pattern separation in aged mice.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

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Topic: E.03. The Chemical Senses

Support: PICT 2020/00360
FOCEM-Mercosur (COF 03/11)
SNSF SPIRIT 216044

Title: Spatial encoding in the piriform cortex is the result of odor-place associative learning

Authors: *L. SALOMON^{1,2}, N. FEDERMAN¹, S. A. ROMANO¹, A. MARIN-BURGIN¹;
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Abstract: Olfactory processing depends on prior experiences, present context, and the animal's internal state. To investigate how learning dynamically changes responses in olfactory cortical circuits, we used a context-dependent olfactory discrimination GO/NO-GO task within a virtual reality setting. Mice explored virtual corridors in which they were presented with two possible odors combined with two possible visual contexts, and were trained to discriminate only one of the four odor-context combinations to obtain a water reward. In previous experiments with this task, we have shown that odor-responsive primary piriform cortex (PCx) neurons become mixed-selective after learning, encoding positional, contextual, and associative information. Nevertheless, it is still unclear how this shift emerges in the PCx encoding scheme. Here, we

performed PCx neuronal recordings throughout the learning process of the task, and combined behavioral analysis together with neuronal encoding and decoding models to address this open question.

We show that animals learn the task sequentially, first discriminating odors (intermediate session) to then also discriminating contexts a few sessions later (expert session), when they show licking responses only to the rewarded odor-context combination. We found that neuronal encoding in PCx parallels this sequential process. While expert animals encode both odor and contextual information in the PCx, animals from intermediate sessions that behaviorally discriminate odors but not yet contexts only encode odors in the PCx. Neuronal decoding analyses confirm this result. Moreover, we observed that neurons from expert animals showed enhanced multiplexing, becoming modulated by a larger number of task parameters compared to first session and intermediate session animals.

This evidence indicates that the observed contextual encoding in the PCx depends on the associative learning process in which odorant cues are combined with positional information to produce correct behavioral responses in the task. Ongoing experiments focus on the neural mechanisms responsible for making this association in the PCx.

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Poster

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Program #/Poster #: PSTR279.11/N3

Topic: E.03. The Chemical Senses

Support: NIH F31 DC0201323
NIH R01 GM130842

Title: Laterality and interhemispheric integration in the larval *Drosophila* olfactory system

Authors: *D. M. ZIMMERMAN^{1,2}, B. L. DE BIVORT^{3,2}, A. D. T. SAMUEL^{1,2};
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Abstract: All animals with bilateral symmetry must integrate the sensory input from the left and right sides of their bodies to make coherent perceptual decisions. In *Drosophila* larvae, connectomic evidence suggests that olfactory signals from the two sides of the head are initially processed independently (as in the mammalian olfactory system) before being combined in the central brain. By pairing volumetric calcium imaging with microfluidic odor delivery and side-specific laser ablation at the sensory periphery, we have mapped the propagation of olfactory signals between the two brain hemispheres, across successive layers of the larval olfactory system. This approach implicates the mushroom body (MB) as a key substrate for

interhemispheric integration of odor representations. Whereas the two larval MBs appear to process odors largely independently at the level of their intrinsic neurons (Kenyon cells), the modulatory neurons of the MB show strongly symmetrized responses to asymmetric olfactory stimuli. Nevertheless, odor responses in some MB output neurons (MBONs), up to 5 synapses downstream from the sensory periphery, preserve information about stimulus laterality. Moreover, we show that asymmetric activation of these MBONs can modulate the animal's turning behavior in a side-biased manner. These findings suggest that the deeply lateralized architecture of the larval olfactory system balances the need for interhemispheric integration with the advantages of parallel sensory processing.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR279.12/N4

Topic: E.03. The Chemical Senses

Support: SFB 1328

Title: Role of the p_{2y1} receptor in neuromodulation of olfactory signal processing

Authors: *S. SHAHMORAD, C. LOHR, D. HIRNET;
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Abstract: Adenosine 5'-triphosphate (ATP) plays an important role as a co-transmitter in the nervous system. ATP activates purinergic signaling by binding to P_2 receptors, which are divided into two subgroups, P_2X and P_2Y . While P_2X work as ionotropic receptors and are activated by ATP, P_2Y are metabotropic receptors which respond to various nucleotides such as ADP, UTP, UDP, ATP, and UDP-glucose. In the central nervous system, purinergic signaling modulates neuronal communication in the brain circuits. Here, we want to investigate the role of purinergic neuromodulation for the processing of olfactory signals. Olfactory receptor neurons (ORNs) in the olfactory epithelium detect odors and project to the olfactory bulb, the first relay station in the olfactory pathway. There, within separate processing units called glomeruli, the ORN axons release glutamate and ATP, exciting mitral cells (MCs), which integrate excitatory input from ORNs and inhibiting interneurons in this processing circuit and serve as the output neurons of the olfactory bulb. Using patch-clamp recording of mitral cells in acute brain slices revealed that the uncaging of caged ATP in the glomerulus significantly depolarizes mitral cells, with the selective P_2Y_1 antagonist MRS2179 reducing the uncaging response. Therefore, mitral cells (MCs) in the olfactory bulb are modulated by ATP/ADP through a P_2Y_1 receptor (P_2Y_1R)-dependent pathway. Furthermore, approximately 50% of ATP-induced excitation was mediated by glutamate, since the ATP effect was reduced in the presence of glutamate receptor blockers, suggesting a multisynaptic excitatory pathway induced by purinergic signaling. As activation of

astrocytes results in ATP-induced ATP/glutamate release and therefore might play a role in this P₂Y₁-dependent pathway, we examined astrocyte-specific P₂Y₁R knockout mice in ATP uncaging. Notably, examining the ATP response by confocal Ca²⁺ imaging in parallel with mitral cell recording by bypassing the astrocytic network did not result in a reduction of the mitral cell excitation. Application of TTX strongly reduced mitral cell depolarization upon ATP release, suggesting a direct depolarization of Mitral/Tufted cells by ATP inducing release of glutamate and excitative responses within this population. Yet, mitral and tufted cell-specific P₂Y₁R knockout mice showed no reduction in the ATP response compared to wild-type controls. In conclusion, a multisynaptic pathway induced by ATP release and resulting in mitral cell excitation highlights a modulatory role of ATP in olfactory processing, involving both direct purinergic receptor-mediated and indirect glutamatergic excitation of MCs.

Disclosures: S. Shahmorad: None. C. Lohr: None. D. Hirnet: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

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Program #/Poster #: PSTR279.13/N5

Topic: E.03. The Chemical Senses

Support: NIHR01DC018075

Title: Time-course of odor coding in the human brain - a single-neuron perspective

Authors: *G. N. DIKECLIGIL¹, M. S. KEHL², J. A. GOTTFRIED¹, F. MORMANN³;
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Abstract: Odor information arising from the olfactory bulb is projected in parallel to the piriform cortex, amygdala, entorhinal cortex, and other subregions of the primary olfactory cortex. These brain areas, which receive direct monosynaptic input from the olfactory bulb, are highly interconnected and functionally linked to higher-order regions, including the hippocampus. However, the precise latency and temporal evolution of odor coding within and across these regions in the human brain remain largely unexplored. To address this, we conducted single-neuron recordings in epilepsy patients undergoing invasive intracranial seizure monitoring with Behnke-Fried electrodes. We recorded neural activity from the piriform cortex, amygdala, entorhinal cortex, hippocampus, and parahippocampal cortex during an odor task (517 units across 7 sessions). Participants performed an odor-identification paradigm in which short, 200-ms odor pulses (one of four complex odorants or odorless air) were delivered via a computer-controlled olfactometer. Participants identified odors in a forced-choice task (odor identity judgment). Breathing was monitored in real time, and odors were delivered using a closed-loop system at consistent phases of the inhalation cycle to ensure temporal precision. Participants successfully identified odorants across sessions. Preliminary results show that

neurons in the piriform cortex exhibit robust, odor-specific firing rate modulations as early as 100 ms after odor onset. Additionally, we observed a higher proportion of odor-modulated neurons in the piriform cortex compared to the amygdala, entorhinal cortex, hippocampus, and parahippocampal cortex. Piriform neurons also demonstrated broader tuning to odor stimuli than neurons in these medial temporal lobe regions. Together, these initial findings demonstrate rapid and precise processing of complex odors by human piriform neurons, laying the foundation for exploring interregional transmission and temporal dynamics in the human olfactory network.

Disclosures: G.N. Dikeçligil: None. M.S. Kehl: None. J.A. Gottfried: None. F. Mormann: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

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Program #/Poster #: PSTR279.14/N6

Topic: E.03. The Chemical Senses

Title: Sharpen tuning along with post-embryonic neurogenesis in fly olfactory center

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Abstract: The number of mushroom body intrinsic neurons (Kenyon cells, KCs) increases ten times during the larval stage of *Drosophila*. However, little is known about the functional consequence of this dramatic neurogenesis event. To address this question, we combine microfluidics, genetics, and calcium imaging to track KCs' responses to a large panel of odorants. We developed a series of microfluidic channels that could fit the size of the larval body across the development. We successfully imaged all KC's responses to over 100 odor stimuli across the three larval developmental stages. Additionally, we have also developed a transparency technique for larvae, which allows us to complete the imaging of bilateral mushroom bodies. With the enriched dataset, we can identify individual KC's diverse receptive fields in the olfactory receptor coding space. We found that at the early development stages, a significant proportion of KCs have simple tuning properties, e.g. activated by individual stimulus channels. However, less of this type of KCs could be found in the late development stages. Furthermore, at late development stages, more KCs have complicated tuning properties, e.g. requiring multiple stimulus channels to be activated simultaneously. Overall, the KCs' tuning profiles become more and more specific and diverse during the development. Another interesting finding is that odor representations from the bilateral mushroom bodies of the same individual exhibit greater similarity, a difference that is significantly more pronounced than random comparisons between different individuals. Taken together, we are able to describe the KC's odorant responses across the development, which pave the road to study the evolution of a central olfaction representation.

Disclosures: W. Li: None. G. Si: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

Location: SDCC Halls B-H

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Program #/Poster #: PSTR279.15/N7

Topic: E.03. The Chemical Senses

Support: Intramural Grant Support from NICHD

Title: Feedback from a higher olfactory center shapes sensory processing

Authors: *M. P. BROWN, M. A. STOPFER;
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Abstract: Sensory systems translate the physical environment into electrical signals that the brain can process. In many sensory systems, specialized projection neurons relay all the information of a given sensory modality from the periphery to the rest of the brain. To understand how our brains represent our sensory environments, it is critical to know how these projection neurons encode this information into discrete electrical signals. Here, we leverage genetic, connectivity-mapping, and electrophysiological tools in *Drosophila* to understand how sensory information is compressed and transmitted through the brain.

In *Drosophila*, olfactory sensory neurons detecting odors from the environment synapse onto antennal lobe projection neurons (ALPNs). ALPNs are the sole carriers of information about odor identity, concentration, timing, and location to the rest of the brain, including areas termed the mushroom body and lateral horn (LH). A key aspect of ALPN function is their remarkable ability to produce diverse firing patterns in response to varying odor types and concentrations. Understanding how ALPNs generate signaling patterns is important for understanding how the brain encodes massive amounts of information with limited resources.

Newly available serial electron microscope (SEM) connectomes of the *Drosophila* central brain reveal what appears to be previously unknown circuitry: local neurons within the lateral horn (LHLNs) that receive direct input from ALPNs and then immediately provide feedback onto ALPN terminals. We hypothesize these newly revealed LHLN-ALPN connections help ALPNs create efficient representations of the olfactory environment. To test this idea, we are characterizing the synaptic connections between ALPNs and LHLNs with precise genetic tools for evaluating connectivity and function. We found that ALPNs are indeed postsynaptic targets of LHLNs and that they express a broad range of neurotransmitter receptors at their terminals. This diversity may enable them to utilize different signaling speeds and regulatory mechanisms to modulate the strength and firing patterns of their odor responses.

We also found evidence suggesting different types of LHLNs express different neurotransmitters. We are now planning *in vivo* patch clamp electrophysiology and voltage imaging of ALPNs and LHLNs to characterize the roles the novel LHLN circuitry play in

olfactory processing. Our work will lead to a clearer picture of how the nervous system represents the physical world.

Disclosures: M.P. Brown: None. M.A. Stopfer: None.

Poster

PSTR279: Olfaction: Higher-Order Circuits

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Topic: E.03. The Chemical Senses

Support: PICT 2020/00360
FOCEM-Mercosur (COF 03/11)
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CONICET PIP 2787

Title: Assessing spatial information coding in the olfactory cortex

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Abstract: The hippocampal-entorhinal network is well known to play an essential role in spatial cognition. Surprisingly, other brain regions not involved in this canonical spatial encoding network have been recently shown to modulate their neuronal activity according to the subject's spatial location. This includes the remarkable discovery of spatial information encoded in the mouse primary visual cortex (V1) and, more recently, in the primary piriform olfactory cortex (PCx) of both rats and mice. Indeed, in a previous study and in work from our lab, rodents were trained in a task to associate olfactory and spatial cues to obtain a reward, and machine learning classifiers were applied to PCx activity to successfully classify the spatial context of each trial. This has been interpreted as spatial maps encoded in the olfactory cortex.

These findings open new important questions. For example, these studies could classify spatial context, but can the animal's spatial location across time be precisely decoded from PCx activity? If so, do the spatial maps of the PCx depend on learning the spatial location of relevant olfactory cues while animals explore the environment? Or is it independent of associating places with olfactory experiences?

To start to address these issues, we study PCx and hippocampal neuronal activity of mice in different learning stages of this olfactory-spatial association task. We perform a series of spatial location decoding analyses using feed-forward and recurrent artificial networks, and combine them with Generalized Linear Models (GLM) regression and simulations to study the

contributions of behavioral, motor and task variables to decoding performance. Our preliminary results indicate that the spatial location of the animal is encoded in the PCx once animals learn the task, but not before. Moreover the spatial encoding precision increases around behaviorally relevant locations. Spatial location decoding performance in the PCx is comparable to decoding performance in the hippocampus. We discuss how these results could contribute to our understanding of the role of the olfactory cortex in olfactory-cued spatial learning.

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Poster

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Topic: E.03. The Chemical Senses

Support: ERC grant Sensational Tethers 789128
Deutsche Forschungsgemeinschaft CRC 958 grant

Title: Anterior olfactory nucleus: an intrinsically mechanosensitive relay for olfaction?

Authors: *A. BALOMENOS, S. CHAKRABARTI, W. LUO, R. PROCE, G. IELACQUA, V. BEGAY-MUELLER, M. PAMPOLS PEREZ, A. HAMMES, H. HÖRNBERG, G. R. LEWIN; Max Delbrück Ctr. for Mol. Medicine, 13125, Berlin, Germany, Berlin, Germany

Abstract: Mammalian olfaction depends on inhalation and the perception of smell may result from convergent processing of chemosensory (odors) and mechanosensory (airflow) information. Numerous studies highlight the putative role of mechanosensation in the olfactory system, but the relationship between the two is not well understood. Using single molecule Fluorescent in Situ Hybridisation (smFISH), we detected the expression of the mechanically gated ion channel *Piezo2*, in many cells of the olfactory system, including high levels of expression in neurons within the Anterior Olfactory Nucleus (AON). Expression of this mechanically gated ion channel in AON neurons was especially prominent in the first two post-natal weeks. Using mice in which *Piezo2-Cre* drives *tdTomato* expression in *Piezo2* expressing cells, we found that *Piezo2* fate mapped cells exist in the olfactory system, most prominently in the AON. These observations prompted us to ask whether robust mechanically activated currents (MA currents) can be found in CNS areas that process olfactory information. We established a primary culture protocol for AON neurons in order to examine whether AON neurons are mechanosensitive. Using single nucleus RNA sequencing we detected expression of *Piezo2* and other mechanically gated ion channels in specific cell clusters from our neuronal cultures. Patch clamp electrophysiological recordings from AON neurons showed that mechanical stimulation via substrate deflection was able to generate robust MA currents. The proportion of *tdTomato*⁺ neurons (reading out *Piezo2* promoter activity) with MA currents was significantly higher than *tdTomato*⁻ neurons. We

sought to understand the role of AON mechanosensitivity in vivo and using functional ultrasound imaging in anesthetized mice we observed activation of AON neurons upon airflow-driven mechanical stimulation. We established a gain-of-function approach by overexpressing Stoml3, a known modulator of mechanosensitivity in Piezo2+ cells within the AON, which was associated with impaired odor discrimination in mice without altering the AON structure. Future work will investigate the airflow-driven activation in this gain-of-function model.

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Poster

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Program #/Poster #: PSTR279.18/N10

Topic: E.03. The Chemical Senses

Title: Neural connectivity constrains computation in the insect Lateral Horn

Authors: *H. GIAFFAR¹, P. JIANG², M. AOI³;

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Abstract: Careful dissection of circuit connectivity, the backbone of network based computation, has been instrumental in developing our understanding of neural algorithms in both brains and machines. The recent release of hemi-, and full brain connectomes from D. Melanogaster has already informed our interpretation of circuits across a wide range of areas, from the visual, taste, and olfactory systems, to the central brain and goal directed navigation systems. The Mushroom Body (MB) - a structure which is crucial in mediating online olfactory learning - has been a major focus of this work. The circuits of the Lateral Horn (LH) - an area thought to mediate innate olfactory processing - have received comparatively little attention, perhaps due to it's relative complexity, with many cell types, diverse inputs/outputs and complex recurrent components. Studies of LH to date have focused on characterizing aspects of the neuropil's structure and response properties, but have not yielded any strong algorithmic insights, on par with those gained from MB. We will present recent work characterizing shared structure, relating to the processing of food odor signals, in the inputs to both LH and MB. This shared structure is consistent across hemispheres and individuals. Furthermore, we characterize co-tuning of glomerular sensitivities and connectivity patterns, finding a significantly stronger shared tuning in the LH pathway vs the MB, with implications for the recently proposed primacy theory of olfactory processing.

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Poster

PSTR279: Olfaction: Higher-Order Circuits

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Program #/Poster #: PSTR279.19/N11

Topic: E.03. The Chemical Senses

Title: Towards understanding host odor processing in mosquito higher olfactory circuits

Authors: *J. ZHENG¹, C. MCBRIDE¹, Z. ZHAO², L. WEISS¹;

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Abstract: Many animals show robust, innate responses to ecologically relevant odors, but how central olfactory circuits evolve to recognize and discriminate these complex stimuli is poorly understood. For example, female dengue mosquitoes (*Aedes aegypti aegypti*, *Aaa*) are strongly attracted to human odor, which they consistently prefer over the odor of non-human animals. However, they evolved from animal-preferring ancestors (*A. a. formosus*, *Aaf*) around 5000 years ago. Understanding this rapid behavioral adaptation requires studying how mosquitoes' brains process different natural odor blends. Previous work from our lab shows that *Aaa* have distinct representations for human and animal odors in the antennal lobe, the first relay for olfactory information in the insect brain. However, it's still unclear how these patterns of activity are integrated in higher olfactory circuits, such as the mushroom body and lateral horn. Is human odor represented by a unique and stereotyped pattern of neural activity in these regions? What functional and neural computational insights can we get from this pattern? How does this pattern differ in homologous brain regions of *Aaf*? To address these questions, we are generating genetic tools that will allow us to reveal odor-evoked activities at the level of the lateral horn and whole brain. We have constructed and validated a driver line for olfactory projection neurons (PNs) equivalent to the widely used *GHI46* line in *Drosophila*, which allows us to image activity in second order olfactory neurons. We are also constructing a PN marker line that would allow direct visualization of the lateral horn (tdTomato) while imaging activity in all neurons (pan-neuronal GCaMP). Besides, we are developing a tethered-flight platform compatible with two-photon imaging where we can motivate the mosquito and record its response to defined olfactory stimuli. Later, we will try to construct artificial neural networks (ANN) that may help us analyze brain imaging data. Together, these tools will allow us to characterize and contrast brain activity evoked by naturalistic and complicated olfactory cues like human odor, animal odors, human odor synergized with CO₂, floral odors, etc. Our data will provide insight into the key neuropils and patterns of activity associated with innate host-seeking behaviors in mosquitoes.

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Poster

PSTR280: Neural Coding in Auditory Cortex

Location: SDCC Halls B-H

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Program #/Poster #: PSTR280.01/N12

Topic: E.05. Auditory and Vestibular Systems

Support: MOTIE-20012355
IITP-2025-RS-2024-00436857

Title: Representation and population drift in mouse auditory cortex in response to passive human monosyllabic speech stimuli

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Abstract: Recent studies suggest that sensory cortical representations drift over time, even under sensory conditions. While representation drift has been observed in the mouse auditory cortex (ACx), it remains unclear whether similar drift occurs in human monosyllabic speech stimuli. To address this, we performed longitudinal single-photon calcium imaging in CaMKII-GCaMP8f-expressing excitatory neurons in the ACx of C57BL/6J mice passively exposed to human monosyllabic utterances (“ba” and “ga”) over twelve days. Neural responses were recorded on day 1, day 6, and day 12. Over time, we observed a gradual increase in the number of active neurons, indicating population-level drift. In parallel, representational expansion emerged, with broader population responses to stimuli. To dissociate whether this drift was driven by tuning changes or baseline shifts, we computed representation dissimilarity matrices (RDMs) using both raw and normalized spike activity. RDMs based on normalized responses showed greater stimulus separation, suggesting that tuning alterations, rather than baseline shifts, played the dominant role. This interpretation was further supported by longitudinal single-cell tracking, which revealed that tuning profiles changed across days, while baseline activity remained largely stable. Together, our findings suggest that representation drift in the ACx is driven by both population drift and tuning reorganization, with minimal contribution from baseline fluctuations.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Topic: E.05. Auditory and Vestibular Systems

Support: NIH/NIDCD R01 DC020459
Klingenstein-Simons Fellowship Award in Neuroscience

Title: Canonical cortical architecture supports the emergence of noise-invariant auditory representations

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Abstract: A key challenge for the auditory system is to maintain reliable representations of sound despite interference from background noise (BN). Although the ascending auditory pathway contributes to constructing noise-invariant representations, the underlying mechanisms and neural subpopulations involved remain poorly understood. In the primary auditory cortex (ACtx), the canonical cortical microcircuit, thalamus-> layer (L)4 -> L2/3 -> L5, transmits sensory information across cortical layers. Here, we examined how neural representations of sound evolve through this microcircuit to achieve noise invariance.

Using cell-type-specific two-photon calcium imaging in mouse ACtx, we recorded sound-evoked activity from axonal inputs of the auditory thalamus (MGBv) and somata of L2/3, L5 intratelencephalic (IT), and L5 extratelencephalic (ET) neurons. Mice were presented with pure tones either in silence or embedded in white BN. To assess noise-invariance, we applied information-theoretic metrics, population decoding, manifold geometry, and pairwise correlation analyses across single-cell and population levels. Tone-evoked responses in MGBv and L2/3 were significantly reduced in the presence of BN, leading to degraded stimulus encoding. In contrast, L5 IT and ET neurons maintained robust single-cell and population-level responses, suggesting the emergence of a noise-invariant output code in the deep layers of ACtx.

To investigate mechanisms supporting this transformation, we developed a holographic optogenetics paradigm to probe functional connectivity within L2/3. We transiently activated small groups of neurons that were either co-tuned to the same frequency or co-modulated by BN. Co-modulated ensembles more effectively drove similarly co-modulated neurons than non-modulated or differently modulated ones. These findings suggest that local connectivity in L2/3 is structured to selectively reinforce noise-invariant subnetworks. Together, our results demonstrate that noise-dependent thalamic inputs are progressively transformed into noise-invariant auditory representations through canonical cortical circuits, with L5 projection neurons serving as a noise-invariant broadcast output.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Topic: E.05. Auditory and Vestibular Systems

Support: R01 DC020459, NIH/NIDCD
Klingenstein-Simons Fellowship Award in Neuroscience

Title: Layer 5 and 6b extratelencephalic neurons encode distinct sound features in the auditory cortex

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Abstract: Extratelencephalic (ET) neurons in layers (L)5 and 6b of the primary auditory cortex (ACTx) project to subcortical targets and contribute to auditory-guided behavior and learning-related plasticity. Although both subtypes share major projection targets, including the inferior colliculus, thalamus, striatum, and amygdala, they exhibit striking differences in morphology and intrinsic physiology. L5 ET neurons are large pyramidal cells with a prominent apical dendrite extending toward L1, and many are burst-spiking. In contrast, L6b ET neurons often have non-pyramidal, radially oriented somata with widely branching dendrites that span more than a millimeter and are predominantly regular-spiking. Despite these differences, the in vivo functional properties of these neurons remain poorly understood. To address this, we used an intersectional viral strategy to express GCaMP8s in both L5 and L6b ET neurons within the same animal. Two-photon calcium imaging was used to record activity during the presentation of pure tones, sinusoidally amplitude modulated (sAM) noise, and ripples. L5 ET neurons exhibited predominantly excitatory responses, with higher response sparsity and trial-to-trial reliability, especially for pure tones and sAM noise. In contrast, L6b ET neurons showed a greater proportion of suppressive responses, particularly to complex stimuli, and displayed enhanced functional connectivity within their population. Unsupervised clustering revealed distinct response motifs and tuning properties across laminar subtypes. L5 ET neurons were more likely to exhibit monotonic, single-peaked tuning, whereas L6b ET neurons displayed non-monotonic and complex tuning profiles. Together, these findings suggest that L5 and L6b ET neurons support complementary coding strategies, with L5 neurons optimized for precise auditory relay and L6b neurons for integrating and broadcasting higher-order features.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Topic: E.05. Auditory and Vestibular Systems

Support: NIH Grant DC009836
Seth Robbins Mass General Neuroscience Transformative Scholar in
Tinnitus

Title: Awakening normal cortical sound processing and perception in deaf adult mice lacking all developmental auditory experience

Authors: *K. K. CLAYTON^{1,2}, K. SUDANA¹, J. LI¹, E. NORRIS³, E. LAWLER¹, P. GRATIAS^{1,2}, E. HALE¹, A. A. INDZHYKULIAN^{1,2}, D. B. POLLEY^{1,2};
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Abstract: The development of sensory systems proceeds sequentially from activity-independent genetic programs which wire up afferent circuits to a series of activity-dependent critical periods where neural tuning is shaped by sensory experience. While altering sensory input during these critical periods can dramatically reshape cortical maps and behavior, it remains unclear whether initiating sensory transduction in adulthood—after lifelong deprivation—can result in normal cortical coding and perception. One possibility is that lifelong deprivation leads to irreversible deficits due to cross-modal plasticity or degeneration of afferent pathways. Alternatively, the basic cortical architecture established prenatally may provide a scaffold for functional sensory processing, even without developmental experience. In the auditory system, mutations in the *Otof* gene prevent synaptic transmission from inner hair cells to auditory nerve fibers and thus completely block afferent sound transmission, while crucially, all other components of the sensory transduction apparatus remain intact. Here, we asked whether initiating afferent sensory transduction in a deaf adult *otof*-deficient mouse using a well-established cochlear gene therapy (Chung et al. 2023 ASGCT) would be sufficient for the emergence of normal cortical sound encoding and perception despite a total lack of auditory experience, as measured through videographic and operant behavioral assays, in tandem with high-density single unit recordings and chronic calcium imaging in auditory cortex (ACtx). Three weeks following gene therapy, treated mice could perform basic sound detection, complex sound discrimination, and even sound detection in noise (N = 6 treated mice, N = 6 normal hearing controls). Sound-evoked facial movements were present, albeit with lower amplitudes (N = 6 treated mice, N = 8 WT). Cortical single unit recordings (N = 4 mice, n = 549 units) revealed strikingly normal receptive fields and tonotopic organization. Further, single units showed normal sound level encoding and even faithfully represented temporally modulated stimuli like click trains. Using a triple transgenic mouse (*VGlut1-Cre;jGCaMP8s:Otof^{f/-}*), we have tracked the emergence of sound responses in populations of L2/3 pyramidal neurons after gene therapy (n = 7,786 neurons). Well-organized tone-evoked receptive fields emerge within five days of gene therapy, suggesting that pre-existing circuit architectures, not experience, drive the rapid emergence of sensory coding and perception following adult-onset sensory transduction.

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Poster

PSTR280: Neural Coding in Auditory Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR280.05/N16

Topic: E.05. Auditory and Vestibular Systems

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Klingenstein-Simons Fellowship Award

Title: Systematic inactivation reveals functional diversity of higher-order cortical feedback to auditory cortex

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Abstract: Perception is an active process in which primary sensory cortices are dynamically modulated by behavioral goals, environmental context, and internal state. Top-down corticocortical feedback is a key mechanism underlying this modulation, and prior studies have shown that individual feedback projections can shape sensory processing and influence behavior. Primary auditory cortex (ACtx) receives feedback from multiple higher-order cortical areas, including prefrontal, motor, and association cortices, and its responses vary markedly with behavioral context. To dissect the functional contributions of distinct top-down sources, we developed a clear-skull preparation enabling temporally precise, unilateral optogenetic inactivation of nine cortical areas within a single recording session. In awake mice, we used high-channel-count extracellular electrophysiology to record single-unit activity across the ACtx laminae during passive presentation of amplitude-modulated pure tones. On randomly interleaved trials, we inactivated each cortical target individually to assess the contribution of distinct feedback sources. This within-session design allowed for direct, trial-wise comparisons of how top-down inputs modulate both spontaneous and evoked activity at the single-neuron level. In the absence of sound, inactivation of prefrontal regions consistently reduced spontaneous firing rates in ACtx, whereas inactivation of motor and association areas had minimal effect, suggesting a dominant role for prefrontal feedback in setting baseline excitability. In contrast, during sound presentation, inactivation of posterior prefrontal, motor, and association regions enhanced tone-evoked responses, revealing a context-dependent shift in top-down influence. Standardized major axis regression showed that inactivation of higher-order regions broadly increased response magnitude, and cosine-angle differences revealed substantial changes in tuning shape. Notably, anterior prefrontal inactivation selectively suppressed responses at each neuron's best frequency, indicating a specialized role in frequency-specific gain control. These findings demonstrate that higher-order cortical inputs modulate ACtx activity in functionally distinct ways, differentially influencing spontaneous and evoked activity in a context-dependent, region-specific manner.

Disclosures: M.I. Malina: None. R.S. Williamson: None.

Poster

PSTR280: Neural Coding in Auditory Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR280.06/O1

Topic: E.05. Auditory and Vestibular Systems

Title: The geometry of cortical sound processing in slow wave sleep

Authors: *A. MULLER^{1,2}, A. FILIPCHUK², S. BAGUR², B. BATHELLIER²;

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Abstract: During wake, sound-evoked and spontaneous neural activity of the auditory cortex evolve in distinct subspaces whereas anesthesia disrupts sound responses and merges these spaces. To evaluate if similar modifications of the sound representation geometry explain sensory disconnection during sleep, we followed large neural populations of the mouse auditory cortex across slow wave sleep and wakefulness. We observed that sleep dampens sound responses but preserves the geometry of sound representations which remain separate from spontaneous activity. Moreover, response dampening was strongly coordinated across neurons and varied throughout sleep spanning from fully preserved response patterns to population response failures on a fraction of sound presentations. These failures are more common during high spindle-band activity and more rarely observed in wakefulness. Therefore, in sleep, the auditory system preserves sound feature selectivity up to the cortex for detailed acoustic surveillance, but concurrently implements an intermittent gating mechanism leading to local sensory disconnections.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Support: NIH/NIDCD R01 DC020459
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Title: Active sensorimotor strategies for sound localization in complex acoustic scenes

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Abstract: In natural environments, animals must react appropriately to behaviorally relevant sounds, such as those signaling predators or food sources, while ignoring competing auditory signals. Effective responses require identifying both the identity and location of sounds. The

psychophysics of sound localization have been well characterized by presenting stimuli at varying positions relative to an animal's head, which is kept stationary for precise spatial control. However, such approaches do not capture the sensorimotor strategies animals use during active navigation. In real-world settings, auditory spatial perception is shaped by environmental noise, behavioral context, and self-motion. Animals frequently orient their heads and bodies to reduce perceptual ambiguity and improve localization accuracy. To investigate the behavioral strategies that support naturalistic sound localization, we developed a freely-moving auditory discrimination task. C57BL/6J mice were trained to localize a target sound along the perimeter of an open circular arena while ignoring a distractor presented simultaneously from another azimuthal position. Stimuli were bandpassed white noise of varying center frequencies. After trial initiation, both stimuli played continuously until mice approached and licked a water spout beneath the perceived target. Since mice were free to explore before making a decision, this design enabled observation of how animals integrate dynamic sensory input with self-motion. Over several weeks of training, mice adopted characteristic sensorimotor strategies that facilitated task learning, including rapid head and body "scanning" movements at stimulus onset and 180° turns to correct errant approaches towards the distractor. These behaviors became more frequent with learning and coincided with increased accuracy, shorter reaction times, and more stereotyped trajectories. These results indicate that mice resolve spatial ambiguity with dynamic movement strategies and continuously update motor plans in response to ongoing acoustic input. Our ongoing work investigates the neural circuits underlying this dynamic sensorimotor loop. While auditory cortex encodes spectral features and learned associations, it lacks a map of egocentric space. In contrast, its projection targets in the midbrain, such as the superior and inferior colliculi, are spatially tuned and mediate orienting behaviors. We are currently using pharmacological and optogenetic perturbations to dissect how corticocollicular circuits contribute to spatial decision-making, offering new insight into how animals transform auditory input into adaptive action.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Support: R01 DC020459, NIH/NIDCD
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CMU Rales Fellowship

Title: Learning sound-reward associations through auditory corticostriatal and dopaminergic signaling

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Abstract: For all mammals, survival depends on the ability to learn which environmental features predict rewards and to use that information to guide behavior. The caudal “tail” of the dorsal striatum (Ts) plays a central role in this process by integrating inputs from sensory cortices and midbrain dopaminergic neurons to link sensory cues with motivated actions. In the auditory system, the tail of the striatum receives dense excitatory input from both layer (L)5 intratelencephalic (IT) and extratelencephalic (ET) neurons in primary auditory cortex (ACtx), each of which adaptively signals different reward-related features of sounds during learning [Schneider et al., *in prep.*].

Here, we examined how cell type-specific activity in ACtx and dopaminergic signaling in Ts jointly support auditory-guided learning. Mice were trained on a head-fixed associative learning task in which one of two sinusoidal amplitude-modulated (sAM) noise bursts predicted a water reward after a short (0.5 s) delay. While mice learned to discriminate between the two stimuli, we simultaneously recorded calcium activity from L5 IT neurons and dopamine transients in Ts using fiber photometry.

Our preliminary results (n=4) show that sound-evoked activity in L5 IT neurons is prolonged and reliably followed by rapid, robust dopamine transients in Ts. These dopamine responses are time-locked to stimulus onset, occur independently of movement, and consistently precede licking behavior. Ongoing experiments aim to characterize how L5 IT, L5 ET, and dopamine signals evolve with learning and whether they encode trial-by-trial reward predictions or errors. We hypothesize that L5 IT neurons facilitate early sound-reward associations, while L5 ET neurons contribute to the execution of learned behaviors in later stages.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Title: Pupil-linked arousal differentially modulates cell-type-specific sound processing in auditory cortex

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Abstract: Arousal state, indexed by pupil diameter, dynamically shapes cortical activity and sensory processing. While prior studies have documented arousal-dependent changes in neural responsiveness, reliability, and tuning in the auditory cortex (ACtx), most have treated the excitatory population as functionally uniform. In reality, ACtx contains diverse excitatory cell types, including intratelencephalic (IT), extratelencephalic (ET), and corticothalamic (CT) neurons, with distinct anatomical and physiological properties. These differences suggest that arousal may influence sensory coding in a cell-type-specific manner.

To test this, we combined pupillometry with two-photon calcium imaging in awake mice, targeting L2/3 (n = 3,023), L5 IT (n = 2,447), L5 ET (n = 2,876), and L6 CT (n = 3,311) neurons. A multivariate regression model revealed that sound-evoked responses were modulated by arousal in a linear or non-linear fashion. The shape and magnitude of this modulation varied by cell type: L2/3 and CT neurons showed peak responsiveness at intermediate arousal levels, whereas L5 ET neuron responses increased monotonically and IT neuron responses remained largely unaffected. Hierarchical clustering identified four distinct arousal-modulation motifs (linear increasing, linear decreasing, U, and inverted-U) that were present across all cell types but differed significantly in prevalence. Tuning width analyses revealed that increased arousal led to pronounced multiplicative and additive gain changes in L5 ET neurons, indicating sharper and enhanced tuning compared to other populations.

To assess how arousal shapes sensory encoding, we trained a neural network decoder to classify stimulus identity from population activity across pupil states. Decoding performance followed an inverted-U profile for L2/3 and L6 CT neurons, increased monotonically for L5 ET neurons, and remained unchanged for L5 IT neurons. Decorrelating population activity had no impact on decoding accuracy, but response reliability strongly predicted decoding performance and mirrored the same arousal-dependent trend. Together, these findings demonstrate that arousal exerts distinct effects across excitatory subtypes, dynamically reconfiguring auditory cortical representations in a cell-type specific manner.

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Title: Deep-layer projection neurons develop representations of perceptual categories and behavioral choice

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Abstract: Auditory categorization, the transformation of continuous acoustic features into discrete perceptual categories, is critical for guiding decisions and actions in daily life. While the auditory cortex (ACtx) is necessary for categorical behavior, the contributions of specific excitatory cell types remain unclear. Nestled amongst several populations of cells, layer 5 (L5) extratelencephalic (ET) neurons are uniquely positioned to influence behavior, as they project to subcortical structures involved in decision-making, motor control, and reward. To investigate their role in perceptual categorization, we trained head-fixed water-restricted mice to categorize the rate of sinusoidal amplitude-modulated (sAM) noise bursts as “fast” or “slow” by licking left or right to receive a water reward. Using cell-type-specific GCaMP8s expression and two-photon calcium imaging, we then recorded from L5 ET, as well as L2/3 and L5 intratelencephalic (IT) neurons.

In expert mice, L5 ET neurons exhibited robust categorical preferences for fast or slow sAM stimuli during active task engagement. These categorical representations were absent in untrained mice and emerged gradually with learning, as revealed by longitudinal imaging. Notably, the same neurons did not exhibit categorical selectivity during passive listening to identical stimuli, suggesting top-down modulatory input. In contrast, L2/3 and L5 IT neurons lacked categorical selectivity under all conditions. To ensure these effects were not confounded by movement, we fit a generalized linear model to dissociate sound-driven and movement-related activity. Categorical tuning in L5 ET neurons persisted even after excluding movement-related neurons, confirming that these signals reflect auditory processing rather than motor output.

Finally, we investigated how population activity encoded behavioral choices. On ambiguous trials at the category boundary, principal component analysis revealed choice-predictive trajectories in all three populations, with the strongest and most distinct activity in L5 ET neurons. Together, these results suggest that L5 ET neurons in ACtx acquire task-dependent representations of both perceptual categories and behavioral choices, positioning this projection system as a key conduit for relaying behaviorally relevant auditory information to downstream targets.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Title: Inversion of auditory cortex feature encoding by behavioral state

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Abstract: Sound features are encoded by the central auditory system through a combination of maps, metronomes, and meters—that is, which neurons are active, when spikes occur, and the overall rate of population activity, respectively. While most sound features are encoded via maps and metronomes, we recently showed that loudness perception is encoded in the auditory cortex using a simplified meter-based code that depends only on the population spiking rate, irrespective of spike timing or neuron identity (Clayton et al., 2024). We found that parvalbumin-expressing inhibitory neurons (PVNs) in the auditory cortex act as a volume knob on this meter, shifting the neural and behavioral boundary between “soft” and “loud” by up to 20 dB. Here, we used 2-photon calcium imaging to record from layer 2/3 excitatory neurons in the primary auditory cortex (A1) of five well-trained mice (N=16 sessions; 4,514 neurons). Imaging was conducted during a two-alternative forced-choice (2AFC) loudness classification task and again during a passive listening session immediately afterward. Passive recordings replicated our earlier electrophysiology findings, revealing heterogeneous sound level tuning across neurons that summed to a linearly increasing population code. Mean population activity from a single trial could classify stimulus sound level within ~3 dB. However, recordings from the same neurons during the behavioral task showed that in ~50% of sessions, this relationship inverted: population activity decreased with increasing sound level. Focusing on stimuli near the perceptual boundary, we found that behavioral choices could also be decoded from the population activity rate, but the direction of this relationship (i.e., more or less activity when mice reported “loud”) was evenly split across sessions. The key variable that explained this inversion was the spatial mapping between loud vs. soft and left vs. right lick spouts, with stimuli mapped to the contralateral spout eliciting the greater population responses. These findings caution against assuming that neural correlates of perception are best—or only—revealed during active behavior. They also underscore the critical importance of spatial counterbalancing in AFC designs. A1 qualifies as a higher-order cortical area in that its activity reflects sound features as well as cognitive and motor variables. Disentangling cognitive and motor influences to isolate the neural encoding of low-level perceptual features is essential but is not trivial, particularly when using slower calcium-based signals to monitor neural activity.

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Title: I contain multitudes: Information processing by individual neurons depends on ensemble synchrony

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Abstract: Neural information processing has traditionally been studied by recording single neuron spike trains and quantifying changes in firing associated with specific sensorimotor events and other processes. Modern tools make it possible to simultaneously record many neurons and detect subsets of coincidentally active neurons. Using these methods, recent studies suggest that information processing by individual neurons differs from moment to moment depending on which other neurons are active at the same time. In our own work, we find many neurons in anesthetized rat auditory cortex (ACtx) are coactivated within multiple, independent coordinated neuronal ensembles (cNEs), which comprise subsets of neurons with reliably synchronous activity. By separating single neuron spike trains into subsets of spikes that are tightly synchronized with members of one or another cNE, we recently found individual neurons are sensitive to different sound features in dynamic moving ripples depending on which cNE they were synchronized with at a given time. This result suggests classic auditory receptive fields derived from single neuron spike trains may in fact reflect an amalgam of multiple underlying stimulus subspaces to which a neuron may respond depending on coactivation with other cNE members. This form of cNE-dependent multiplexing at the level of individual spikes could increase both the encoding capacity of individual neurons and the efficiency with which neural networks represent sensory and other information spaces. Considering the potential broad relevance of this finding for understanding how neural networks process information, the aim of the current study was to determine whether and how cNE-dependent coding is observed in neurons across diverse brain regions, processing a wide range of sensory features and other events. We conducted a series of experiments using Neuropixels probes to record from awake mouse ACtx, thalamus, and hippocampus. We replicated our earlier finding that single unit responses to dynamic moving ripples depend on cNE synchrony, show that this phenomenon generalizes to other sound types, and that it also occurs in auditory thalamus. We next demonstrate that cNE synchrony similarly influences how single units in ACtx respond to non-auditory events such as visual stimuli and locomotor feedback. Finally, we show that hippocampal units similarly respond to sound in ways that critically depend on cNE synchrony. Together, our findings suggest cNE-dependent coding - in which individual neurons play

“different roles on different teams” - is a ubiquitous strategy for processing diverse types of information across brain regions.

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Poster

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Title: Auditory-guided inference and belief-state updating in dynamic foraging behavior

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Abstract: While model-free associative learning allows animals to rapidly link stimuli with outcomes, flexible goal-directed behavior in dynamic environments requires processing uncertainty and the ability to generalize associations. These capabilities emerge from inferring a model-based causal representation of task states (belief states). Belief states are represented and updated in the orbitofrontal cortex (OFC), which encodes value-tagged cognitive maps. However, how these representations are updated through learning from ambiguous auditory cues remains poorly understood. To investigate this, we developed a novel closed-loop, auditory-guided dynamic foraging task in mice, designed to probe interactions between OFC and auditory cortex (ACtx) during belief updates in inference-based decision making. The task includes two phases: in the inference phase, mice learn solely from reward outcomes; in the auditory phase, they learn a cue-target mapping. In a linear track with speakers and lick spouts at left (L) and right (R) ports, mice initiate trials by maintaining position in a central location to trigger onset of an auditory cue (identical from both speakers), followed by a fixed delay after which reward is available at a target with a high probability. In the inference phase, the auditory cue (8 Hz, sAM noise) is non-informative, and the hidden target (L/R) reverses probabilistically after a correct lick. This probabilistic structure is formalized as a partially observable Markov decision process (POMDP), and prior work shows it engages the OFC during optimal inference. In the auditory-guided phase, the cue identity (2 Hz and 32 Hz, sAM noise) is perfectly informative about the target, rendering the reward state fully observable given the cue. Mice trained on the inference task show choice behavior consistent with belief states computed by an ideal observer, consistent with a model-based strategy that updates using state prediction errors. In the auditory-guided phase, trained mice perform with high accuracy and movement trajectories reveal signatures of

evidence accumulation and decision confidence. Notably, 20-30% of trials are “early trials”, where mice violate the post-cue delay and receive no reward. Accuracy on early trials improves with training but plateaus below completed trials, suggesting a dissociation between task acquisition and expression. Using Bayesian Q-learning, we show that uncertainty-guided exploration accounts for these learning dynamics. Ongoing experiments aim to directly compare model-based and model-free learning driven by auditory cues, enabling mechanistic dissection of belief-state updating in OFC-AC circuits.

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Poster

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Topic: E.05. Auditory and Vestibular Systems

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Title: A generic deep learning architecture for bio-inspired encoding of an auditory cortical implant

Authors: *C. LAMOTHE¹, B. BATHELLIER²;

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Abstract: Cochlear implants (CIs) are the most successful sensory neuroprosthesis and help almost a million patients to recover functional hearing. However, their relatively poor spectral and temporal accuracy limits speech intelligibility in noise and music perception. Moreover, CIs do not address pathologies involving auditory nerve loss or cochlear malformation. Because auditory cortex stimulation leads to sound perception, a possible approach to circumvent these issues could be to implant the neuroprosthesis at this level. However, cortical implants would require high-level encoding models to transfer information in a format similar to sound representations in the auditory cortex. To address this challenge, we have developed an AI-based encoder allowing us to recreate and map key features of the biological auditory code observed in the cortex while ensuring a high information throughput, which is closely measured by reconstructing the waveforms of the encoded sounds. Our approach uses an autoencoder whose latent space matches the number of electrodes on the stimulator and the temporal resolution of auditory cortex activity. The autoencoder is combined with a classifier of human-labelled sound categories that enables the generation of structured pre-semantic representations. The encoder and decoder are based on modern 1D convolution layers explicitly developed for sound processing, yielding efficient training and excellent performance. We demonstrate that this novel hybrid architecture trained on millions of natural sounds can perform sound encoding on hundreds of channels with almost no information loss while generating stimulation patterns matching multiple neural code properties of the auditory cortex. Preservation of information is

established by the high perceptual similarity of reconstructed sounds compared to original sounds, for speech, music and natural sounds. The similarity with natural auditory cortex representations is established by using representational similarity analysis on cortical data and on the representation of our encoder. In addition, specific constraints in the training process allow us to reproduce tonotopy. In addition, we demonstrate that single channel activities for well-known sounds have temporal profiles similar to cortical neurons with a variety of onset, offset and sustained responses, as well as tuning properties observed commonly in vivo. Finally, we show that mice can perceive and use the representations generated by our models with good precision. Hence, our approach allows for precisely delineating the design of a potentially successful auditory cortical implant for auditory restoration.

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Poster

PSTR280: Neural Coding in Auditory Cortex

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Topic: E.05. Auditory and Vestibular Systems

Support: CNRS

Title: Ramped pulses as a reliable alternative solution to the use of rectangular pulses in cochlear implants: An electrophysiological study in the guinea pig auditory cortex.

Authors: *E. PARTOUCHE¹, V. ADENIS², C. HUETZ³, J.-M. EDELINE³;

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Abstract: For decades, cochlear implant (CI) is the most successful neuro-prosthetic device allowing more than a million of patients to recover hearing sensation and speech understanding. The performance is usually good in silence, but CI patients have difficulties in the presence of background noise. Potentially, these limitations stem from the large spread of currents diffusing in the cochlea's perilymph when the different electrodes are activated at high current levels. To reduce the spread of current, one potential strategy is to change the shape of the electrical pulses. In human, studies have tested asymmetric rectangular pulses (Macherey et al., 2006, 2008). A new pulse shape, called "ramped pulse", has been proposed (Ballesterio et al., 2015) but only two studies have showed that such pulses elicited responses with lower thresholds than rectangular pulses (Navntoft et al 2020, Partouche et al 2023). Here, we report the consequences of using different types of ramped pulses on responses of auditory cortex (ACx) neurons in anesthetized guinea pigs.

The intracochlear stimulating array was a shortened version of the EVO electrode array used by Oticon Medical and was composed of 6 ring-shaped Platinum-Iridium electrodes with a 0.0046mm² surface. Four shapes of ramped pulses were tested, each at 20 charge levels (from 3

to 31.5nC), to obtain the growth functions of ACx neurons. Recordings in ACx were collected by 16-channels electrodes composed of two rows of 8 electrodes. Two types of doubles ramped pulses were tested: one with the ramps on the 1st and 4th phases, a second with the ramps on the 1st and 3rd phases. We also tested single ramp pulses with the ramp on the 3rd phase. Compared with the responses obtained with rectangular pulses, all ramped pulses generated (i) responses with lower thresholds (ii) higher maximal firing rates (FR) (iii) higher FR at threshold and (iv) longer response latencies. These effects were always significant with cathodic-first phase, and were in the same direction (but less often significant) with anodic-first phase. These results indicate that ramped pulses should be considered as a good alternative to rectangular pulses, even if polarity of the first phase matters to benefit from this shape.

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Title: A corticofugal gated memory model captures auditory cortical responses to natural sounds

Authors: *L. MAZZASCHI, A. J. KING, B. D. B. WILLMORE, N. S. HARPER;
Dept. of Physiology, Anat. and Genet., Univ. of Oxford, Oxford, United Kingdom

Abstract: To make sense of dynamic natural sounds, cortical neurons must be sensitive to sequential dependencies in sounds at various timescales. This suggests that the mammalian auditory cortex uses memory processes that retain information over seconds and fractions of seconds. To understand this neural system, we built computational models to predict responses of auditory cortical neurons. Existing models use long delay lines as memory, which is biologically unrealistic. We therefore developed new models with memory mechanisms arguably more consistent with biology. Specifically, we incorporated gated recurrency - a powerful concept from machine learning. This involves units with switchable memory components (gates) that enable relevant information to be retained whilst forgetting irrelevant information. Our aim was to improve the performance and biological plausibility of auditory models by capturing neural sensitivity to sequential stimulus dependencies. We fitted gated recurrent models to extracellularly recorded spiking activity of neurons recorded in primary auditory cortex (A1) and higher auditory cortex (posterior ectosylvian gyrus, PEG) of awake ferrets in response to natural sounds. We compared the capacity of these models to predict neuronal responses, relative

to standard delay-line models. Notably, we developed a gated recurrent model, the corticofugal long short-term memory (f-LSTM) model, which incorporated input gating resembling that described for corticofugal projections to subcortical regions. The f-LSTM performed the best of all the models for both A1 and PEG responses. We further improved the biological plausibility and performance for the f-LSTM model in A1 by adding an initial feedforward expansion layer. In A1, the deep cortical layers 5-6, the source of descending corticofugal projections, receive extensive input from the superficial layers 2-4, which have a higher neuronal density than layers 5-6. This added expansion therefore mirrors high-dimensional processing in superficial cortical layers. Analyzing our models, we also found substantially longer memory retention in PEG than A1, suggesting a central role for PEG in memory-mediated processing. We also identified two kinds of stimulus features that evoked responses for which gated recurrency may be particularly relevant: sharp changes in sound intensity and periods of quiet. These results suggest that the auditory cortex may use a form of gated recurrency and that corticofugal projections and superficial-layer dimensionality expansions are important in understanding auditory cortical memory systems.

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Topic: E.05. Auditory and Vestibular Systems

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Title: Uncovering neuron type diversity in the auditory cortex of marmosets through unsupervised machine learning analysis

Authors: *K. DOXEY¹, Y. ZHOU², S. M. CROOK³;

²Speech and Hearing Sci., ³Sch. of Mathematical and Statistical Sci., ¹Arizona State Univ., Tempe, AZ

Abstract: Recent advances in electrophysiological, morphological, and transcriptomic techniques have enabled increasingly detailed cell type characterizations in cortical circuits (e.g., Allen Mouse Brain Atlas). However, it is challenging to classify neuron types based on extracellular action potentials (EAPs) recorded from high-density electrode arrays, particularly in the absence of “ground truth” labels. Current approaches rely on extracted features and machine learning (ML) algorithms that result in varying numbers of neuron type classes (Jia et al., 2019, Lee et al., 2021, Haynes et al., 2024). Here, we used an unsupervised ML framework to cluster EAP types based on both single- and multi-channel features extracted from extracellular recordings in the auditory cortex of two awake common marmosets (*Callithrix jacchus*). We

analyzed neural recordings from 70 sessions across two adult marmosets, identifying thousands of spiking events. We constructed multi-channel 2D kernel waveforms with a 1.5 ms temporal window and ~400 μ m depth spread from spatial clusters of the spiking events. We applied KMeans and Agglomerative clustering on two representations of the data: (1) extracted spatiotemporal features (e.g., event duration, vertical spread, amplitude, etc.) and (2) vectorized channel waveforms. Both input types yielded meaningful cluster separations, but clustering based on extracted features results in more distinct EAP classes. In particular, event duration and vertical spread across channels were the most robust discriminators among clusters. This suggests the presence of diverse neuronal subtypes with distinct extracellular signatures in the marmoset auditory cortex. Our data-driven approach demonstrates how unsupervised ML algorithms can uncover physiologically meaningful variation in EAP waveforms, providing a scalable tool for comparative and cross-modal analysis of cortical organization.

Disclosures: K. Doxey: None. Y. Zhou: None. S.M. Crook: None.

Poster

PSTR280: Neural Coding in Auditory Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR280.18/O13

Topic: E.05. Auditory and Vestibular Systems

Support: NIDCD R01DC019278

Title: Layer and frequency dependent propagation of local field potentials in the marmoset auditory cortex

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Abstract: Local field potentials (LFPs) are low-frequency signals reflecting the summed extracellular activity of local neuronal populations. While LFP response strength reflects the magnitude of these currents, the spatial spread and waveform shape are influenced by the geometry of dendrites and the curvature of cortical layers, particularly in regions with pronounced folding such as sulci and gyri. The common marmoset (*Callithrix jacchus*) has a lissencephalic (smooth) brain with only two prominent sulci—the lateral sulcus and the superior temporal sulcus—both located in the temporal lobe, with the auditory cortex situated between them. Although the cortical anatomy is simplified, difficulties with tonotopic mapping using LFPs arise due to high individual variability and uncertain signal origins. In this study, we examined LFP propagation across cortical depth and surface area surrounding the primary auditory cortex (A1) in two marmoset monkeys. Using high-density linear silicon probes, we recorded LFPs and single-unit activity (SUA) along both the laminar depth and rostral-caudal-lateral axes. Auditory stimuli included pure tones (2-32 kHz) and broadband noise (BBN) presented at multiple sound levels. We observed persistent LFP responses to pure tones around

18 kHz that propagated at least 2 mm caudally and 5 mm laterally from A1. These responses were strongest in superficial layers and diminished in deeper layers at distal sites. BBN elicited similar propagation patterns but only at higher intensities (10-30 dB SPL). In contrast, within A1, LFP strength increased with depth, spanned a broader frequency range, and was detectable at lower sound intensities. These findings suggest that LFP propagation in the marmoset auditory cortex is frequency- and layer-dependent, with distinct activation patterns within and beyond A1.

Disclosures: D. Nguyen: None. Y. Zhou: None.

Poster

PSTR280: Neural Coding in Auditory Cortex

Location: SDCC Halls B-H

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Program #/Poster #: PSTR280.19/O14

Topic: E.05. Auditory and Vestibular Systems

Support: NIH NINDS R01, NS137950
NIH NIDCD R01, DC020717

Title: Altered phonetic representation in tumor-afflicted auditory cortex during speech perception

Authors: *Y. LI¹, S. OTEN², S. HERVEY-JUMPER³, D. BRANG⁴;

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Abstract: Glial brain tumors form electrical and chemical synapses with healthy neurons to accelerate tumor proliferation, potentially altering local neural computations. Prior work has shown that glioma-infiltrated cortex remains similarly active during speech production but with decreased entropy, suggesting reduced information processing capacity. The current study investigates how tumor presence affects auditory information processing in the human superior temporal gyrus (STG) during naturalistic speech perception. Subdural electrocorticography (ECoG) data were collected from tumor patients during neurosurgery. Task-related changes in high-gamma power (HGp, 70-150 Hz) were used to measure neural selectivity for phonetic features. Distributed changes in population responses were quantified by the classification accuracy of support vector machine (SVM) classifiers trained to decode phonetic categories from HGp. Preliminary results show that while tumor-afflicted areas achieve a comparable mean decoding accuracy to normal-appearing regions, they exhibit greater variability in decodability, suggesting a noisier phonetic representation. These findings align with previous research on phonetic feature encoding in human STG and provide new insights into speech information processing in the human auditory cortex. This work highlights the partial preservation of neural computation in tumor-infiltrated auditory regions, which is both scientifically and clinically

significant as it offers a foundation for further understanding the mechanism of altered neural decodability as well as perceptual outcomes in patients following surgical resection.

Disclosures: Y. Li: None. S. Oten: None. S. Hervey-Jumper: None. D. Brang: None.

Poster

PSTR281: Higher Visual Areas

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Program #/Poster #: PSTR281.01/O15

Topic: E.06. Vision

Title: Robustness of task performance to neural manifold perturbation in artificial neural networks

Authors: *J. GUO, L. CHANG;
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Abstract: Neural manifolds, which are embedded in low-dimensional subspaces and encode behaviorally relevant neural population dynamics, are widely hypothesized to constitute fundamental representational bridges between brain activity and behavior. However, direct empirical validation of their functional necessity for cognitive computations remains challenging, with current evidence limited to indirect metrics, such as explained variance in neural population activity. To address this issue, we use artificial neural networks (ANNs) as experimentally tractable models for systematic perturbation of representational subspaces. Their explicit layer-wise activation spaces (representation/manifold) and quantifiable task performance (behavior) allow us to investigate the relationship between manifolds and behavior directly. Using image classification neural networks (AlexNet, VGG19, Resnet152, SwinTransformer) as examples, we quantified the change in classification accuracy by iteratively removing the principal components from the activation space of different layers. We observed remarkable robustness in task performance (i.e. classification accuracy) against subspace removal: the accuracy of AlexNet decreased by only 5% when principal components with the highest explained variance (up to 50%) were removed from the output layer. Similar results were found in other classification models. Further analysis revealed fundamental differences in the representational geometry: the representational dissimilarity matrix (RDM) between all pairs of image categories in the output layer of shallow neural networks (AlexNet, ResNet18, VGG19) exhibits a low-rank (smooth) structure, with its rank gradually increasing when principal components are removed. In contrast, the representation in the ultra-deep networks (Resnet152, SwinTransformer) is inherently high-rank. During network training, the low-rank structure emerges transiently before vanishing with continual training. Interestingly, retraining AlexNet on a small subset of the full dataset (50-class only) preserved a low-rank structure in the RDM that was similar to that of the well-trained AlexNet. These findings suggest that low-dimensional manifolds within ANN's deep-layer space may be non-essential for task performance, representing domain-general features inherited from preceding network stages. These domain-

general features undergo nonlinear transformations before evolving into task-specific features, with their relative importance decaying during training in ultra-deep architectures.

Disclosures: J. Guo: None. L. Chang: None.

Poster

PSTR281: Higher Visual Areas

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Topic: E.06. Vision

Support: NIH Grant R00EY031795
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Title: Neural representational geometry of border features in macaque area V4

Authors: Y. LU¹, F. DIDEHVAR², Z. YE³, M. AZADI², A. DAS², *T. P. FRANKEN²;
¹Washington Univ. Sch. of Med., Saint Louis, MO; ²Washington Univ. Sch. of Med., St. Louis, MO; ³Dept. of Physics, Washington Univ. in St. Louis, St. Louis, MO

Abstract: Visual scenes contain borders that separate foreground objects from background. Each border has a side of ownership: the border is perceptually owned by the side that belongs to the foreground object. Prior studies found that border ownership is prominent and computed early in Area V4 of the macaque brain, in which these signals are organized in columnar clusters. In addition, V4 neurons are also selective for other features of borders, such as their orientation and their luminance contrast. However, how V4 populations simultaneously represent these features is unclear. Here we characterize their neural representational geometry. We obtained recordings from columnar populations of tens to hundreds of neurons simultaneously using Neuropixels probes in Area V4 while a macaque performed a fixation task (n=11 sessions). We inserted the probes orthogonally such that the classical receptive fields of the neurons in the recorded population were clustered on one region in the visual field. During the task, we presented scenes with a luminance-defined square object such that only one of its borders overlapped with the classical receptive fields, and varied luminance contrast polarity and border ownership, for different border orientations. After spikesorting the data, we performed principal component analysis on the spike counts evoked by the stimuli to analyze the neural representational geometry for border orientation, border ownership and luminance contrast polarity, separately for each simultaneously recorded columnar cluster. We find that a substantial subset of columnar clusters (4 out of 11 penetrations) show an ordered representation of the responses to the different scenes in PCA space (first three principal components). In these penetrations, the neural representation forms two separate rings, which correspond respectively to the two luminance values that were used to define the squares. In each of those rings, there is an orderly

organization of border orientation, such that opposite values of border ownership and border contrast polarity occur at opposite sides of the rings. Together our data suggest that the parameter space is fully represented in individual V4 columns: they simultaneously encode border orientation, border ownership and border contrast polarity. These features are organized primarily by square luminance rather than by border ownership or contrast polarity. This suggests that the representation is surface-based rather than border-based.

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Poster

PSTR281: Higher Visual Areas

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Topic: E.06. Vision

Support: National Natural Science Foundation of China (Grant No. 32171033)
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Title: Domain-organized neural coding of black and white stimuli in macaque v4 revealed by two-photon imaging

Authors: *X. SUN, X. LU, T. ZHOU, M. LI, D. XING;
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Abstract: Previous research reveals asymmetry in encoding black and white stimuli in the visual system. Specifically, in macaque primary visual cortex layer 2/3, 93% of neurons strongly prefer black stimuli. However, this finding is confusing. Although a similar asymmetry exists in luminance perception, the preference for black is less pronounced, tending toward a more balanced coding for black and white stimuli. This inconsistency indicates unknown luminance-processing mechanisms in the macaque visual system. Furthermore, unlike macaques, the primary visual cortex of other species contains many neurons that strongly prefer white stimuli, forming ON-OFF cortical domains to encode black and white stimuli. These findings suggest that the ventral pathway in primates may employ a dynamic mechanism, with ON-OFF domains emerging at a certain stage, to adjust black-white encoding for a more balanced response. To validate this conjecture, our study investigates how V4 encodes luminance in macaques. Two awake adult macaques were trained to perform a passive fixation task. Using optical imaging methods, we found that V4, unlike V1, contains three domains: OFF domains preferring black stimuli, ON domains preferring white stimuli, and non-preferential domains, which together form the black-white polarity map of V4. More importantly, we employed two-photon imaging technology to characterize the response properties of V4 neurons, providing deeper insights into their functional organization. We recorded 19 ROIs, with around 3,500 neurons in total. We

found that V4 neurons are indeed divided into three response types: those preferring black, white, or neither, with these three types of neurons spatially clustered into corresponding domains. Within ROIs from stronger response regions in these three domains, the proportion of neurons matching the preferred type exceeded 90%. Notably, non-preferential neurons still responded strongly to black and white stimuli. These results were consistent for both monkeys. Together with previous studies, our results indicate a dynamic encoding mechanism that adjust the neural responses to black and white stimuli to be more balanced, although still biased to black, along macaque visual ventral pathway.

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Poster

PSTR281: Higher Visual Areas

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Program #/Poster #: PSTR281.04/P2

Topic: E.06. Vision

Support: JSPS KAKENHI 25H00576
JSPS KAKENHI 23KK0046

Title: Flicker-induced time dilation: the contributions of flicker frequency and semantic processing

Authors: *T. ODA¹, A. HOUSHMAND CHATROUDI², Y. YOTSUMOTO¹;

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Abstract: Flickering stimuli lead to an overestimation of temporal duration, a phenomenon termed Flicker-Induced Time Dilation (FITD). However, the underlying mechanisms of FITD remain controversial. Previous studies have primarily focused on flickering stimuli that activate the primary visual cortex and the dorsal visual pathway. More recently, however, some studies have suggested that semantic information may also contribute to time dilation. This study therefore aimed to investigate whether activation along the ventral visual pathway contributes to FITD by using various types of flickering stimuli presented at different frequencies. To this end, we generated scramble and semantic flickers by employing the Semantic Wavelet-Induced Frequency Tagging (SWIFT) method. Scramble flickers alternated between two different scrambled images, while semantic flickers alternated between a semantic image and its scrambled version. Importantly, both types of flickers preserved low-level visual features across frames, such as global luminance, contrast, and the local distribution of spatial frequencies. In each of Experiments I and II, eight participants were asked to compare the subjective durations of a standard stimulus and a comparison stimulus, allowing us to calculate the point of subjective equality (PSE). In Experiment I, scramble and semantic flickers at 2, 4, and 6 Hz were used as standard stimuli. While both types of flickers were designed to elicit only minimal activation in low-level visual regions, the semantic flickers were further intended to preferentially activate

higher-level regions of the ventral visual stream. Scramble and semantic static images were also included as baseline conditions. In Experiment II, we introduced luminance flickers—both scramble and semantic—at 2, 4, and 6 Hz to strongly engage low-level visual regions. In both experiments, flicker frequency had a robust effect on perceived duration, supported by inclusion Bayes factors of $BF_{\text{incl}} = 14.04$ (Experiment I) and > 1000 (Experiment II), indicating strong to extreme evidence. In contrast, stimulus type (scramble/semantic) exerted only a weak influence; anecdotal BF_{incl} values from both experiments offered limited support for its contribution. Collectively, these findings indicate that while higher flicker frequencies induce greater time dilation, semantic information does not appear to significantly modulate time perception. Furthermore, the observation that the frequency-dependent increase in time dilation is notably amplified when the low-level visual cortex is strongly entrained underscores its predominance in FITD.

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Poster

PSTR281: Higher Visual Areas

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Program #/Poster #: PSTR281.05/P3

Topic: E.06. Vision

Support: SNF Postdoc Mobility Grant

Title: Persistence of internal representation of rapidly presented natural images along the visual hierarchy in monkey

Authors: *E. REVOL, P. PAPALE, P. R. ROELFSEMA;
Vision and Cognition, Netherlands Inst. for Neurosci., Amsterdam, Netherlands

Abstract: In daily life, we are constantly exposed to brief and successive visual images, which can yield responses in the brain lasting beyond stimulus presentation. How are these images processed in the brain to form stable representation? In this study, we investigated the persistence of internal representation of brief visual images and their propagation along the visual hierarchy. For that, we recorded from multiple Utah Arrays (1024 electrodes) implanted in one rhesus monkey in the primary visual cortex (V1), V4 and the inferotemporal (IT) cortex. The monkey was trained to a fixation task, with a grey screen before/after the presentation of the stimuli. Firstly, we recorded the cortical signals while the monkey saw a single image for 100 ms, belonging to one of 4 categories (monkey, giraffe, key, pencil sharpener), for a total of 40 different stimuli. Secondly, we performed a rapid serial visual presentation (RSVP) experiment of 4 consecutive images from the same image set, randomized to obtain all possible combination of image category and position in the presentation sequence. Images were presented at different exposure times (25, 50, 100, 200 ms). We measured multi-unit activity (MUA) and trained multi-class linear discriminant classifiers to predict image category. For single image stimulus, peak

mean decoding accuracy (78-88%) occurred between 50-70 ms following stimulus onset, and decoding stayed above chance level for > 500 ms after stimulus offset in all visual areas. Decay of decoding was the slowest in IT (407 ms from stimulus onset to reach half of the max decoding accuracy, vs. 295 ms in V4 and 254 ms in V1). During RSVP stimuli, decoding capability also extended beyond stimuli exposure times, and accuracy globally increased along the visual hierarchy (+20%). With long exposure times (100 and 200 ms), each class could be decoded along the presentation sequence with similar decoding accuracy: one representation replacing the next, with some overlap, in a sequential fashion in all visual areas. Interestingly, for stimuli with short exposure times (25 and 50 ms), the first and last image were well decoded, while masking of the 2 images in the middle of the sequence occurred (18% drop in decoding accuracy in V1). This suggests mainly forward masking, where the processing of the next image is disrupted by previous ongoing processing if the next image is shown <50ms later. In this study, we examined how the brain processes brief natural images along the visual hierarchy in monkey. This dataset opens the way for further investigation on internal representations and its link to different forms of visual memory (iconic, fragile or working memory).

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Poster

PSTR281: Higher Visual Areas

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Program #/Poster #: PSTR281.06/P4

Topic: E.06. Vision

Support: National Eye Institute Intramural Research Program at the National Institutes of Health (ZIA EY000511)

Title: Primate superior colliculus contributes to face processing in temporal cortex

Authors: *G. YU¹, L. N. KATZ², R. J. KRAUZLIS¹;

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Abstract: We recently reported that neurons in the macaque superior colliculus (SC) exhibit a preference for face stimuli at extremely short latencies. This finding raises the question of what SC might contribute to the well-established selectivity for faces in the temporal cortex. To answer this question, we used multichannel probes to record activity from face-selective neurons in the temporal cortex, both before and during SC inactivation. Visual object stimuli—including faces, bodies, and human-made objects—were presented either within the visual field location affected by SC inactivation (5° away from fixation, contralateral to the recorded hemisphere) or outside it (5° ipsilateral). Results show that SC inactivation significantly reduced the selectivity of face-selective neurons in the temporal cortex. This effect was predominantly observed for stimuli presented in the visual field affected by SC inactivation and primarily restricted to the initial visual response following stimulus onset (~70-140ms), rather than the later phase (200-

270ms). In addition to face-selective neurons, our dataset included a population of neurons selective for bodies. SC inactivation reduced the selectivity of body-selective neurons in the early phase as well, but the effect was substantially weaker than that observed for face-selective neurons. Our findings demonstrate that activity from the SC contributes to object processing in the temporal cortex at short visual latencies, and this contribution is larger for faces than for bodies. Thus, the visual processing of faces and other objects may depend on a fast circuit from the SC to the temporal cortex, in addition to the feedforward hierarchy of areas in the visual cortex.

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Poster

PSTR281: Higher Visual Areas

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Topic: E.06. Vision

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Title: Reversible inactivation of macaque Anterior Intraparietal area affects activations during action observation in ventral premotor and prefrontal area

Authors: *S. RIVA, P. JANSSEN;
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Abstract: The macaque area AIP has a pivotal role in both visually-guided grasping and action observation. Reversible inactivation (RI) of this area causes an inability to pre-shape the hand to perform a grasping action (Gallese et al, 1994). Yet, the effects of RI of this area on other cortical areas are still unknown. We studied the functional activations and connectivity during action observation before and after RI of AIP using muscimol (10 mg/mL) during MION-enhanced functional Magnetic Resonance Imaging (fMRI). Two macaques (both males) fixated in a 4 x 4 visual degrees window while watching two sets of videos presented in a block design fashion. The first set was made of hands (namely action-hand) performing a grasping action, while the second set showed also monkey faces (namely action-hands-face) as well as hand actions. We used phase scrambled videos, static frames, and fixation as control conditions. Without muscimol injection, the contrast hand actions versus scrambled and static controls yielded significant activations of parietal areas AIP, LIP, VIP and MIP, as well as prefrontal (area 12, 45A/B, 46), premotor (area F5a/p/c) and temporal areas (widespread activations along the STS). RI of AIP unilaterally caused a reduced activation not only in AIP, but also significant reductions in action observation-related activations in prefrontal areas 45A, 12 and 44 and in ventral premotor area F5c. Surprisingly, RI of AIP also induced significant effects on the functional activations in early visual areas V1 and V2. Moreover, we found a significant increase in the functional connectivity between contralateral and ipsilateral prefrontal and premotor areas

during RI of AIP. Interestingly, those same areas showed a significant decrease in connectivity with the early visual, parietal and temporal areas during the action-hand stimuli. Our results showed that several prefrontal areas and ventral premotor area F5c are causally related to the functional activation of area AIP during action observation, which clarifies the flow of visual information in the macaque brain when observing action of others.

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Poster

PSTR281: Higher Visual Areas

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Topic: E.06. Vision

Support: ZIAMH002838
ZIAMH002898
ZIAMH002899

Title: Comparing neuronal responses to face stimuli in frontal and temporal face patches

Authors: *C. GREGG¹, K. W. KOYANO³, W. ROBISON¹, J. TAUBERT⁴, D. A. LEOPOLD²;
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Abstract: Facial recognition is a critical skill shared by humans and non-human primates, supported by specialized brain areas in both the temporal and frontal cortices. A commonly held face-recognition theory proposes a multi-dimensional “face space,” where distinctiveness cues for individual identities are encoded relative to an experience-dependent average face. We previously found that neurons in fMRI-defined face patches of the inferior temporal cortex, including the anterior medial (AM) face patch, exhibit tuning around the average face, consistent with this framework. However, neurons in the same face-selective regions did not respond to non-face images that strongly resemble faces, a phenomenon known as face pareidolia, suggesting that such tuning is specific to real faces. The aim of the present study is to replicate and extend these findings in additional face patches in the temporal and frontal cortex. In two male Rhesus macaques, we used fMRI to localize AM and the perirhinal (PRh) face patches in the temporal cortex and orbitofrontal cortex (PO) and frontal pole (FP) face patches in the prefrontal cortex. In each monkey, we implanted four 64-channel chronic brush electrodes, one into each patch. We simultaneously recorded responses of single-units from the four areas to morphed faces, pareidolia faces, and other image stimuli. For the morphing experiment, each animal viewed macaque and human faces across identity levels spanning full trajectories in face space, from caricatures (exaggerated identity) to antifaces (negative identity). Responses in

frontal vs. temporal lobe face patches were generally similar for these stimuli. A subset of neurons in AM, PRh and PO exhibited V-shaped tuning for morphed stimuli around the average face, which was rare among FP neurons. One difference between the temporal and frontal face patches was that area PO showed stronger tuning around the average for morphed macaque faces compared to morphed human faces, possibly reflecting its role in conspecific face analysis for social interaction and decision-making. For the pareidolia experiment, the animals viewed a large set of pareidolia images along with matched control images that did not elicit the perception of faces. Few face-selective neurons in any of the areas showed elevated responses for pareidolia stimuli compared to control images, supporting the idea that tuning for the average face is specific to real-world faces and shaped by prior experiences. Together, these results reveal largely shared elements of facial structural analysis expressed among neurons in fMRI-defined frontal and temporal cortex face patches.

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Poster

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Topic: E.06. Vision

Support: R21 Ey 037065

Title: Testing shape processing algorithms in V4 derived from neural network models

Authors: *A. M. CHEN¹, R. SRINATH³, C. E. CONNOR²;

¹Biomed. Engin., ²Krieger Mind/Brain Inst., Johns Hopkins Univ., Baltimore, MD; ³Neurobio., Univ. of Chicago, Chicago, IL

Abstract: Studies of neural coding in intermediate and higher level stages of the ventral/object pathway of primate visual cortex have revealed that neurons represent 2D and 3D shape in terms of object fragments, their geometric properties, and their object-relative spatial configuration. However, almost nothing is known about the neural algorithms that generate this information from lower level inputs. Here, we used analysis of visual neural network processing to derive hypotheses about these algorithms in area V4, an intermediate stage where individual neurons encode orientation, curvature, and object-relative positions of 2D contour fragments, 3D surface fragments, and 3D medial axis fragments. We have also tested these hypotheses with electrophysiological recording from V4 neurons in Rhesus Macaques. We analyzed AlexNet, a convolutional visual network trained on the ImageNet database. Previous work has shown that V4 responses are most closely modeled by layer 3 of such convolutional vision networks. We analyzed how layer 3 response patterns depend on differential inputs from neurons in layer 2 (conv2) and layer 1 (conv1), modulated through max pooling, rectification, and connection

weight patterns to the layer 3 neuron. One hypothesis that emerged from these analyses is that V4 neurons that encode 3D shape fragments may be driven mainly by achromatic contrast and low spatial frequencies. In contrast, V4 neurons selective for 2D shape would be driven mainly by chromatic contrast. We tested these hypotheses by comparing the sensitivity of 3D- vs. 2D-responsive neurons in V4 to achromatic vs. chromatic contrast gratings at low and high spatial frequencies. Our early results support the AlexNet-based hypotheses. 3D-responsive neurons were activated more by achromatic contrast and low spatial frequencies, in contrast to 2D-responsive neurons. This trend is consistent with the importance for 3D perception of shading, which depends on low-frequency achromatic contrast.

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Poster

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Topic: E.06. Vision

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Title: Putative inhibitory neurons play a dynamic role in border ownership computations in primate visual cortex

Authors: *M. AZADI¹, F. DIDEHVAR², T. P. FRANKEN³;

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Abstract: A prominent scene segmentation signal in primate visual cortex is generated by border ownership (BOS) neurons: they signal which side of a border in their classical receptive field (cRF) is foreground. BOS neurons occur in columns in macaque Area V4, in which neurons tend to share the same preferred side of BOS. It remains unclear how BOS is computed. Since borders are perceptually owned by one side at a time, inhibition may play a role to suppress the other side. One possibility is that inhibitory (I) neurons receive input from excitatory (E) neurons with opposite BOS preferences and inhibit both, which predicts that BOS selectivity is weaker for I than for E neurons. Another possibility is that BOS-selective I neurons help establish BOS by inhibiting nearby E neurons, which predicts that nearby I and E neurons are both BOS-selective and that BOS-signals occur earlier in I neurons than in E neurons. To distinguish between these possibilities we used Neuropixels to record from 10 columnar clusters of single neurons in V4 of one adult male macaque. We find a strongly bimodal distribution of spike duration, allowing us to classify narrow-spiking neurons as putative I and broad-spiking neurons as putative E (as in

Mitchell et al. (2007)). We measured receptive fields using subspace reverse correlation and identified laminar compartments using current source density analysis. As in prior studies we used square stimuli to measure BOS signals, positioning the square such that only one of its borders is centered in the cRF, and varied the side of ownership of this border, controlling for differences in contrast polarity. We used the BOS index (BOI, difference divided by sum of spike rates to stimuli with opposite BOS) to identify BOS-selective neurons (permutation test). We compared BOI as well as BOS-induced spike count differences between cell types. We find that both putative E and putative I neurons can be selective for BOS. BOS induces larger spike count differences for putative I neurons than for putative E neurons, even though higher rates across conditions results in lower BOI for putative I than for putative E neurons. The difference in spike counts between cell types is most pronounced in deep cortical layers. A simple spike count classifier for BOS is more accurate for putative I than for putative E neurons early after stimulus onset. Our data argue against the model that I neurons have weaker BOS selectivity than E neurons. Instead, they suggest that I neurons play an active, dynamic role in BOS computations, i.e. early after stimulus onset and mostly in deep cortical layers.

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Poster

PSTR281: Higher Visual Areas

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Topic: E.06. Vision

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Title: Brain encoding analysis with naturalistic movie fMRI in humans and macaques

Authors: *H. WANG¹, M. FEILONG², J. HAN², X. LI³, R. KIM⁴, Y. ZHANG², G. JIAHU⁵, M. FROESEL³, Y. LIU³, W. SHIM⁴, M. GOBBINI⁶, Q. ZHU⁷, J. V. HAXBY², W. VANDUFFEL³;

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Abstract: Understanding how humans and macaques process multimodal information in the brain yields critical insights into the evolution of the primate brain — a central question in systems neuroscience. Compared to traditional paradigms, naturalistic stimuli such as movies provide rich, ecologically-valid multimodal input that better reflects real-world experience. In this study, we used brain encoding models (Tour et al., 2025) to analyze fMRI data from 24 humans and 12 rhesus macaques who viewed the same documentary *Monkey Kingdom*. This movie featured dynamic scenes involving natural and man-made objects, various actions of macaques, humans, and other animal species, and synchronized audio. Objects and actions in the movie were annotated at 1-second resolution by 10 raters and subsequently quality-checked. These features were represented either as one-hot vectors or semantic embeddings derived from OpenAI’s text-embedding-3-large model. Compared to one-hot vectors, embeddings place semantically similar objects/actions closer together. Additional features — visual motion via pymoten and audio via Mel-frequency cepstral coefficients — were also extracted. For each subject, voxel-wise ridge regression modeled the relationship between fMRI signals and stimulus features. A nested leave-one-movie-clip-out cross-validation strategy was used to mitigate overfitting. Features — including objects, actions, visual motion, and audio — were incrementally added to the model to assess their individual contributions. Semantic embeddings of objects and actions consistently outperformed one-hot encodings in both species. Incorporating additional features significantly improved encoding performance, particularly in modality-specific cortical regions (Figure). This highly consistent pattern of modality-specific performance enhancements reveals conserved neural pathways for encoding complex multimodal information across human and macaque brains, validating naturalistic paradigms as powerful tools for advancing our understanding of primate brain evolution.

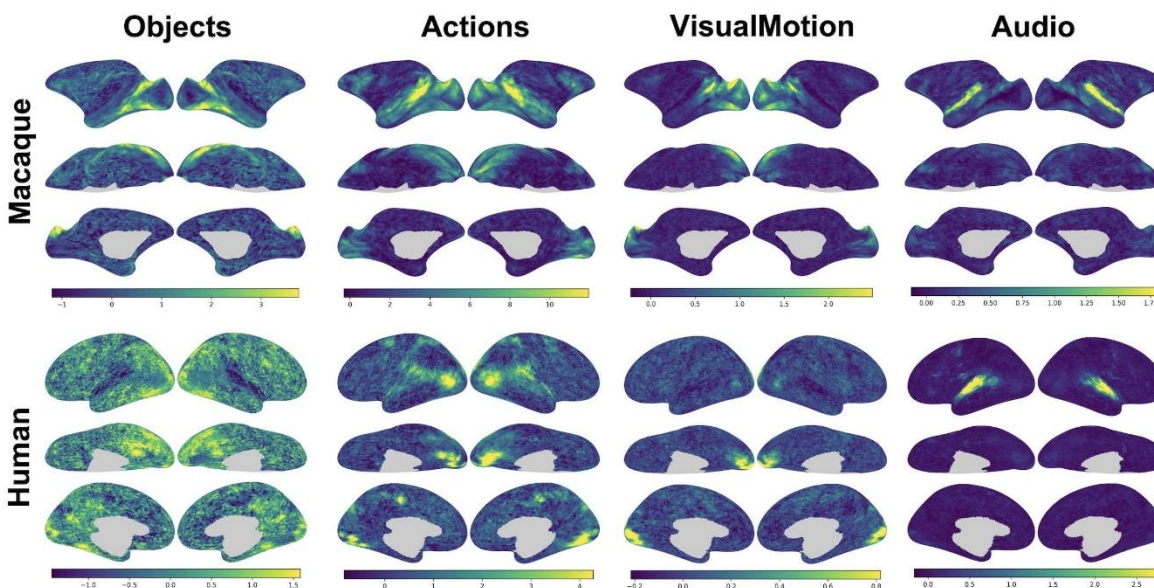


Figure. Brain regions showing increased encoding performance when specific stimulus features were added in macaques and humans. The color bar indicates t -values from two-sample t -tests comparing model prediction performance (correlation between predicted and actual fMRI time series, converted to z-scores) with and without each feature.

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Poster

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Title: Functional Segregation of Face Part Representations in Macaque Face Patch

Authors: *J. LUO, J. YANG, H. YAO, L. YIPENG, S. TANG, P. BAO;
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Abstract: The inferotemporal (IT) cortex of primates contains multiple category-selective regions. Among these, face-selective patches have been extensively studied, revealing both population-level and single-neuron tuning properties. Prior work has shown a progressive increase in viewpoint invariance along the posterior-to-anterior axis of the face-processing network, and the anterior lateral (AL) face patch in particular exhibits mirror-symmetric tuning to left and right profile views (Freiwald & Tsao, 2010). Despite this progress, it remains unclear whether individual face patches contain finer-scale spatial subdivisions that reflect tuning to specific facial features. In this study, we ask whether face patches, particularly AL, exhibit internal spatial structure that differentiates responses to different dimensions of the faces. To address this question, we used wide-field calcium imaging to record neural responses from a ~10 mm region overlapping the anterior lateral (AL) face patch in two macaques as they viewed systematically varied face stimuli—including manipulations of viewpoint, expression, gender, species, and isolated upper versus lower face parts. Among these stimulus dimensions, two prominent feature maps emerged: first, spatially distinct subregions showed strong selectivity for frontal versus profile views, consistent with previously reported viewpoint tuning in AL. More unexpectedly, both monkeys exhibited sharply delineated subregions that preferentially responded to either the upper or lower half of the face, revealing fine-scale spatial organization based on face part selectivity. Furthermore, to probe these part-based preferences at higher resolution, we performed large-scale extracellular recordings using Neuropixels probes targeting the ML and AL face patches. Across more than 1,000 face-selective neurons, over 70% exhibited robust and consistent preferences for either the upper or lower face parts. These findings reveal a previously unrecognized level of fine-scale functional organization within macaque face patches. The discovery of spatially segregated subregions tuned to specific face parts—alongside known

view-selective zones—suggests that face patches are not homogeneous modules, but contain structured subpopulations dedicated to encoding distinct facial features.

Disclosures: J. Luo: None. J. Yang: None. H. Yao: None. L. Yipeng: None. S. Tang: None. P. Bao: None.

Poster

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Title: Holistic vs. Partial Processing of Faces in Face-selective and Face-responsive Regions in the Fusiform Gyrus

Authors: *Z. LI^{1,2,3,4}, G. TAN^{1,2,3,4}, T. XIE^{2,3,4}, H. CHO^{2,3,4}, J. R. SWIFT^{2,3,4}, X. LIU^{1,2,3,4}, J. T. WILLIE^{1,2,3,4}, P. BRUNNER^{1,2,3,4};

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Abstract: The fusiform face area (FFA) is a specialized brain region in the human ventral occipitotemporal cortex associated with face perception. Overlapping regions in the fusiform that respond to both faces and words suggest a degree of domain-general processing. How face-selective (only responding to faces) and face-responsive (also responding to other categories of visual stimuli) regions in the fusiform process differ in processing faces remains unknown. Some studies indicate that FFA performs holistic processing of faces. In this study, we investigated whether face-selective and face-responsive regions in the fusiform gyrus play different roles in holistic or partial processing of faces. Seven patients with intractable epilepsy undergoing intracranial monitoring for the localization of their seizure onset zone within their temporal lobe participated in this study. Subjects attended to rapid serial visual presentation of images, randomly selected from face and non-face categories (faces, bodies, scenes, objects, scrambled objects, digits, line drawings, words, shuffled words, and character strings). Each image was shown on the screen for 200ms, followed by 800ms of interstimulus interval. Pair-wise coherence analysis between the pre-stimulus and post-stimulus periods of broadband gamma

(BBG) extracted from the stereo-electroencephalography (SEEG) signals revealed a face-selective region anterior to the face-responsive region in FFA. To understand the role of this region in holistic and partial processing of faces, the subjects also attended the rapid visual serial presentation of a variety of face images (full face, inverted full faces, full face with and without external features, single facial feature on 2D face contour, certain facial features missing from intact faces), with each image presented on the screen for 500ms, followed by 500ms of interstimulus interval. Our results show a differentiation between pre- and post-stimulus temporal BBG activity in face-selective locations with response to images of intact faces (inverted and non-inverted) with all features presented, while face-responsive locations also show differentiation in their BBG response to pictures of facial parts such as mouth, eyes, or nose in their natural configuration.

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Poster

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Topic: E.06. Vision

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Title: Perturbability of inferior temporal neurons depends on presence of a visual stimulus

Authors: *A. DINH¹, B. KARAMI^{1,2}, R. AZADI¹, A. AFRAZ¹;

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Abstract: We have previously demonstrated that the detectability of stimulation in the inferior temporal (IT) cortex of monkeys varies with the visual stimuli presented. Strikingly, detectability is lowest when no image is shown. We hypothesize that this reflects a network-level constraint, in which artificial activation of a single neuron is less effective when surrounding neurons are at their baseline—an effect we refer to as perturbability. This leads to the prediction that the perturbability of IT neurons is state-dependent, shaped by ongoing cortical activity influenced by visual input. Consequently, identical cortical stimulation would evoke different levels of neuronal activation depending on the presence of visual stimulus. To test this, we employed optogenetic stimulation using the excitatory vector pAAV-CaMKIIa-C1V1(t/t)-TS-EYFP, targeting excitatory neurons. Monkeys were trained to maintain central fixation while a series of images were presented for 1 second each, with 200 ms intervals between images. In 50% of the trials at random, a 200 ms square light pulse was delivered 400 ms after image onset to the transduced tissue. We observe that cortical stimulation during no image condition elicits significantly weaker neural response compared to image presentation condition. This finding

provides an explanation for the animals' reduced performance in detecting cortical stimulation in the absence of visual stimuli, and supports the hypothesis that cortical perturbability is modulated by visual input.

Disclosures: **A. Dinh:** None. **B. Karami:** None. **R. Azadi:** None. **A. Afraz:** None.

Poster

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Title: Predictive context induces flexible and dynamic transformation of neural codes in the face-processing hierarchy

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Abstract: Telling people apart is crucial to thrive in a social environment and depends on visual information derived from faces. Our recognition abilities are thought to rely upon stable sensory neural codes in the visual cortical hierarchy where in the feedforward sweep each stage in the hierarchy gradually extracts features such that the high-level areas show invariance to low-level differences like pose of a face. Nevertheless, our perception is heavily dependent on context, learnt information and predictions. Here, we investigate how learnt predictive and contextual information flexibly affect neural representations in the macaque-monkey face-processing system, a 3-level processing hierarchy in ventral visual cortex, using fMRI-guided simultaneous multi-area electrophysiology from the face areas ML and AL. We find that the neural code in the early feedforward phase exhibits the known representation (i.e. view or shape of the face in ML and mirror symmetry in AL) in a context-independent stable manner. But the late period of the neural response brings about context-dependent flexibility where no inherent coding no longer exists. Instead, when faces are predictable, the lower area, ML shows higher-order invariant representation in the late period of the neural response departing away from its inherent feedforward representations. This indicates that top-down signals dynamically transform lower-order neural representations of faces towards high-order, invariant and separable neural

geometries. On the other hand, when predictions are violated, the late phase period in the low-level area ML exhibits neural representations similar to the high-level areas in a feature specific manner. Interestingly, the higher level area AL exhibits an increase in low-level properties in the late time period of the neural response. Our results provide evidence how predictive context provided through feedback flexibly and dynamically transforms representations towards higher-order properties to optimally use the computational resources provided by cortical processing hierarchies. Whereas, violations in predictions update the predictions by bringing about flexible changes about the features that were violated. This is indicative of a potential bi-directional exchange of information within the ventral visual hierarchy as suggested by various predictive processing theories. Our work on feedback computations and flexibility of sensory neural codes thus has significant implications on how contextual information is utilized in visual perception as well as models of face/object recognition, which are almost entirely built on feedforward architectures.

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Poster

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Program #/Poster #: PSTR281.16/Q2

Topic: E.06. Vision

Support: Simons Foundation
Howard Hughes Medical Institute

Title: Neural representation of dynamic visual experience in the primate brain

Authors: *J. LU^{1,2}, K. MOHAN³, D. Y. TSAO⁴;

¹Neurosci., UC Berkeley, Berkeley, CA; ²Caltech, Pasadena, CA; ³Univ. of California, Berkeley, CA; ⁴UC Berkeley & HHMI, Berkeley, CA

Abstract: Our visual experience is remarkably rich and structured — we naturally parse visual scenes into fundamental elements, intuitively identifying who or what is present, which objects are involved, and how they interact. How does the brain create an abstract, meaningful representation from continuous visual input? We hypothesize that the brain discretizes the continuous visual stream through eye movements, forming a structured, graph-like representation analogous to a sentence diagram. To investigate this hypothesis, we developed a 3D game scene featuring humans and animals interacting randomly, generating a continuous sequence of events that can each be parsed into "subject-verb-object" sentences (e.g., "person A kicks person B"). Neural activity was recorded simultaneously using Neuropixels probes across multiple brain regions while a monkey freely viewed this animated scene. We observed robust neural activation and selective tuning to specific sentence components across a distributed network, including the

inferotemporal cortex, ventrolateral prefrontal cortex, and the hippocampus. The inferotemporal cortex exhibited transient activations aligned with fixations on objects, strongly biased toward the fixated object, consistent with a memory-less, retinotopic coding scheme. In contrast, the prefrontal cortex displayed more sustained responses and selective tuning to specific combinations of identities and actions. Decoding and geometric analyses revealed a high-dimensional representation within the prefrontal cortex, likely emerging from the integration of visual information across multiple saccades. Binding verbs and adjectives to identities also appeared strongly influenced by eye movements. We propose a model wherein visual identities dynamically populate informational slots and interconnect into a graph-like structure enriched by context-specific details. Our findings offer a foundational step toward understanding how the brain extracts semantic meaning from visual scenes.

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Poster

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Topic: E.06. Vision

Support: HHMI

Title: Caudal intraparietal area neurons support reconstruction of 3D shape and pose

Authors: *D. BI¹, D. Y. TSAO²;

¹UC Berkeley, Berkeley, CA; ²UC Berkeley & HHMI, Berkeley, CA

Abstract: Primate caudal intraparietal area (CIP) has been linked to stereoscopic depth processing and 3D orientation encoding. Early functional imaging showed robust activation of this region during stereopsis (Tsao et al., 2003), and single neuron recordings in CIP showed encoding of orientations of 3D planes (Tsutsui et al., 2001; Rosenberg et al., 2013). Here, we recorded the activity of hundreds of CIP neurons with NHP Neuropixels probes while animals passively viewed 1.5k stereo 3D meshes of natural objects rendered at different orientations. To perform object reconstruction, we first trained a variational autoencoder on 24k images of 3D objects to learn a rich latent space of visual features. Then, we trained lightweight neural adapter modules for each experimental session to map CIP population responses to this latent space. Using these models, we show that CIP harbors a visual code for objects that is rich enough to support reconstruction of both an object's 3D shape and its egocentric pose, two key ingredients for an object-centric representation within posterior parietal cortex (PPC). Our results suggest that CIP, and the broader PPC, furnishes a geometric scaffold linking visual object information to spatial frameworks needed to guide intelligent behavior.

Disclosures: D. Bi: None. D.Y. Tsao: None.

Poster

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Topic: E.06. Vision

Support: NIH UF1NS126485
HHMI

Title: Representation of internal models in the primate brain

Authors: *S. CHEN¹, K. MOHAN², J. LU³, D. Y. TSAO⁴;
¹UC Berkeley, Berkeley, CA; ²Neurosci., Univ. of California, Berkeley, CA; ³Neurosci., Caltech, Pasadena, CA; ⁴UC Berkeley & HHMI, Berkeley, CA

Abstract: Primates have a remarkable ability to efficiently extract and store information about the environment. This ability to rapidly learn and generalize is facilitated by the representation of a world model: an internal framework of knowledge about the structure of the world that enables prediction and planning. How are internal models formed and activated during active behavior? Here, we probe the neural implementation of internal models by recording from multiple brain areas in primates as they navigate naturalistic environments. We developed a battery of behavioral tasks that involve passive and active navigation in virtual reality (VR), in which animals search for cued objects by navigating in a complex environment consisting of a set of connected rooms. Monkeys explored the visual environment uniformly and performed the cued active task with high search accuracy, navigating for over tens of seconds on single trials. During active navigation, we conducted high-density, simultaneous *Neuropixels* recordings in IT face patches, entorhinal cortex, and the hippocampus to uncover how the primate brain represents visual information about objects, their locations, and relationships between objects in an internal model. Neuronal responses from IT face cells showed enhanced responses towards cued, target faces during navigation when compared to uncued faces, revealing a detailed representation of objects in the VR environment. Population-level neuronal activity in the hippocampus and the entorhinal cortex shows robust spatial coding that distinguishes between rooms in the VR environment, with data suggesting a potential graph-like representation of the environment. Our findings provide a starting point for understanding how visual object representations are incorporated into an internal model and how it facilitates real-world object recognition during active navigation.

Disclosures: S. Chen: None. K. Mohan: None. J. Lu: None. D.Y. Tsao: None.

Poster

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Topic: E.06. Vision

Support: HFSP LTM LT0055/2022-L

Title: Mapping social perception to social behavior using artificial neural networks

Authors: *N. DOLENSEK^{1,2}, S. CHEN^{1,2}, S. GINOSAR³, D. Y. TSAO^{1,2};

¹UC Berkeley, Berkeley, CA; ²HHMI, Berkeley, CA; ³TTIC, Chicago, IL

Abstract: Primates possess sophisticated social cognitive abilities, interpreting complex social cues and producing adaptive behaviors in real-time. These abilities are thought to rely on distributed circuits spanning sensory, limbic, and prefrontal brain regions. Despite some progress in identifying neural correlates of specific social features, such as gaze and social rank, a comprehensive computational framework linking social perception to behavior remains elusive; at least in part due to the high dimensionality of both the input space (complex social scenes) and the output space (diverse behavioral responses).

To address this, we combine artificial neural networks (ANNs) and high-density Neuropixels electrophysiological recordings in macaques to uncover how the primate brain transforms dynamic social scene perception into social behavior. We first employ whole-brain functional magnetic resonance imaging (fMRI) to locate brain regions selective for social stimuli in macaques. We then target these regions using Neuropixels probes and employ a novel ANN-based approach to map the high-dimensional visual input onto neural population activity and, in turn, to link that activity to behavioral responses.

Our results suggest that identified social frontal brain regions contain a surprisingly rich code for social scenes and play a critical role in transforming social perception into behavior, positioning them as central nodes in social cognition.

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Poster

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Program #/Poster #: PSTR281.20/Q6

Topic: E.06. Vision

Support: ONR
HHMI

Title: Collaboration Across Face Patches Resolves Diverse Visual Ambiguities

Authors: *Y. SHI¹, J. K. HESSE², F. F. LANFRANCHI³, D. Y. TSAO⁴;
¹Caltech, UC Berkeley, Pasadena, Berkeley, CA; ²MCB, UC Berkeley, Berkeley, CA; ³MCB, Caltech/UC Berkeley, Berkeley, CA; ⁴UC Berkeley & HHMI, Berkeley, CA

Abstract: Object recognition in the primate brain is accomplished by the inferior temporal (IT) cortex, where specialized networks encode increasingly invariant representations of object categories. Within this architecture, the face patch system transforms retinotopically variable facial inputs into identity-stable codes, with the anterior face patch AM often exhibiting tolerance to changes such as viewpoint. Yet, how this invariance is constructed—particularly under ambiguous or degraded conditions—remains poorly understood.

A prevailing hypothesis proposes a dichotomy: clear, unambiguous stimuli are processed through feedforward computations, whereas degraded inputs require recurrent computations. Here, we performed simultaneous Neuropixels recordings across multiple face patches to examine how degradations such as occlusion and rotation engage interactions between middle (ML) and anterior (AM) face patches. We found that AM can resolve certain occlusions immediately, while rotation invariance emerges later and appears to depend on reciprocal interactions with ML. Conversely, ML disentangles identity and degradation-related signals in distinct ways depending on the type of degradation. These observations suggest that the face patch network reconfigures its computational strategy based on the nature of visual uncertainty.

Altogether, our results reveal a flexible, context-dependent architecture in high-level visual processing—one that emphasizes inter-patch communication over rigid hierarchy. This framework provides a basis for investigating how recurrent circuits in the primate brain support generative inference and robust perception.

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Topic: E.06. Vision

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Title: Understanding semantic memory representation in the primate anterior temporal lobe

Authors: *R. SUN¹, D. Y. TSAO²;
¹UC Berkeley, Berkeley, CA; ²UC Berkeley & HHMI, Berkeley, CA

Abstract: How is our knowledge of the world stored and represented in the brain? Semantic memory (or conceptual knowledge) refers to the information we possess that enables us to bring meanings to objects, words, sounds, and all other types of stimuli. Early work on a specific type of neurological deficit—characterized by a selective loss of semantic memory but relatively

intact perceptual skills and episodic memory—has shown that focal atrophy in the anterior temporal lobe (ATL) is the single most significant pathological feature, identifying the ATL as a crucial hub for semantic memory. Two core questions underlying semantic memory representation are: (1) How are modality-specific semantic features integrated into a unified conceptual representation? (2) What is the general coding scheme for semantic concepts? To answer these questions, we presented semantically-rich image and video stimuli to non-human primates while using Neuropixels probes to record from different sites within the anterior temporal lobe. First, we found that the cells display different temporal dynamics in response to static image versus video stimuli. Second, we found significant modulation of video responses by concomitant auditory input. Finally, we found that the population representation to visual stimuli in the anterior temporal lobe is better explained by the final categorization layer of vision models (for example fc8 of AlexNet) and the aligned conceptual space of vision-language models (for example CLIP) than by lower-level representations, supporting the role of ATL neural populations in extracting semantic meaning.

Disclosures: R. Sun: None. D.Y. Tsao: None.

Poster

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Title: Is visual cortex purely visual and motor cortex purely motor? Encoding of joint positions and visual features in the monkey inferotemporal and premotor cortex during screen based and naturalistic tasks

Authors: S. SAHA¹, S. SIMON¹, S. MUNDA¹, T. CHERIAN¹, J. DAS¹, G. JACOB¹, D. BHADRA¹, J. JOBY^{2,1}, *S. P. ARUN¹;

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Abstract: While the visual cortex is by definition modulated by visual input and the motor cortex is by definition modulated by motor output, these distinctions have been challenged by recent observations that visual (motor) cortex is modulated by motor (visual) information,

particularly in naturalistic contexts. To what extent do visual or motor features modulate the higher order visual and motor cortex, and does the answer vary between restrained and naturalistic contexts?

To address this fundamental question, we performed wireless brain recordings from inferior temporal (IT) and premotor/prefrontal (PMv/PFC) cortex of two monkeys while they performed two tasks. In the fixation task, monkeys viewed images of natural objects while they voluntarily rested their head on a chin-rest. In the naturalistic task, they moved freely through a naturalistic environment to take food offered by an experimenter across many trials. To measure visual information, we extracted visual features from a pre-trained deep network with input being either the contents of the screen for the fixation task, or the putative visual input from the video feed showing both monkey and experimenter in the naturalistic task. To measure motor information, we extracted joint positions using markerless pose tracking algorithms from the video feed. In each case, we asked whether multiunit activity could be predicted as a linear combination of visual or motor features, while equating models to have the same degrees of freedom.

Our main results are as follows. First, in the fixation task, we found a clear double dissociation between IT and PMv/PFC regions whereby IT neural responses were predicted better by visual (but not motor) features, and PMv/PFC responses were predicted by motor (but not visual features). Importantly the contribution of motor features to IT responses was non-zero whereas that of visual features to PMv/PFC responses was minimal. Next, in the naturalistic task, this double dissociation was far less prominent but observed only during key events in the task (e.g. when the experimenter reached the interaction zone), when the visual and motor inputs are most likely separable and not correlated with each other. During other events, such as when the animal and experimenter were approaching each other, both visual and motor features would contain signatures of these correlated movement, resulting in both types of features predicting neural activity in visual and motor cortex. Taken together, our results reveal a nuanced picture of how visual and motor information modulate higher order visual and motor regions in the brain during controlled and naturalistic behaviour.

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Poster

PSTR281: Higher Visual Areas

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Title: Is neural activity in inferotemporal cortex during real-world vision fundamentally different or more variable than during screen-based tasks?

Authors: *S. MUNDA¹, S. SIMON¹, S. SAHA¹, T. CHERIAN¹, J. DAS¹, G. JACOB¹, D. BHADRA¹, J. JOBY^{1,2}, S. P. ARUN¹;

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Abstract: It is widely believed that neural activity during real-world vision is either fundamentally different or more variable compared to controlled screen-based tasks. This is because each trial in a real-world task might involve variations in when and how an object is viewed and could involve additional influences due to eye, head and body position variations. However, since neural activity has rarely been recorded in both controlled and natural settings, this remains an untested proposition. To address this fundamental gap in our knowledge, we developed a naturalistic environment consisting of a large arena to study real-world vision and a touchscreen workstation to study screen-based vision. We performed wireless brain recordings from inferior temporal (IT) and premotor/prefrontal (PMv/PFC) cortex of two monkeys while they performed two tasks. In the fixation task, monkeys viewed images of natural objects while they voluntarily rested their head on a chin-rest. In the naturalistic task, they moved freely through a naturalistic environment to take different food rewards offered by different experimenters across many trials. To investigate visual information present in each task, we trained linear decoders on the recorded neural population to infer the food & experimenter identity on each trial of the natural task. Our main findings are as follows: (1) Food & experimenter identity during the naturalistic task could be reliably decoded using decoders trained on the fixation task, but only at specific event times; (2) Food was decodable only from IT during the fixation task but from IT & PMv during the natural task, presumably because different foods involved different grasps; experimenter identity could be reliably decoded from IT in both tasks; (3) neural activity during the naturalistic task was systematically different around the times of key events during the trial in both IT & PMv/PFC; (4) When decoders are trained on all available trials, food & experimenter decoding was more accurate during the fixation task compared to the naturalistic task, which supports the hypothesis that natural vision is more variable. However, this advantage reversed on considering matched numbers of trials in the fixation and naturalistic task, i.e. the decoding was more accurate in the naturalistic task. When decoders were trained on matched trials from a limited number of views per object in the fixation task, we obtained similar accuracy in the fixation and naturalistic task. Taken together, our results show that neural activity during real-world vision is less variable than previously thought and can be systematically related to neural activity during controlled fixation tasks.

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Poster

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Title: Encoding of grooming variables & inter-brain synchrony during simultaneous brain recordings from two socially interacting monkeys

Authors: *D. BHADRA¹, S. SIMON¹, S. SAHA¹, S. MUNDA¹, T. CHERIAN¹, J. DAS¹, G. JACOB¹, J. JOBY^{2,1}, S. P. ARUN¹;

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Abstract: Brain activity in monkeys is almost always recorded while they are restrained, which blinds us from understanding brain activity during natural behaviours, particularly social behaviours. Here, we present the first ever simultaneous wireless brain recordings from two socially interacting monkeys. Each monkey was implanted with 256 electrodes targeting the inferior temporal cortex (IT) and the premotor/prefrontal cortex (PMv/vlPFC). Each monkey first performed a fixation task where it viewed images of itself, its social partner and a third unrelated monkey. Subsequently, both monkeys were brought into a single room where they interacted in unrestricted fashion while their brain activity was recorded wirelessly. Over the course of interaction, each monkey performed grooming of its partner, and received grooming from its partner, interspersed with periods where both monkeys did not interact. We asked several fundamental questions about brain activity in both monkeys during this social interaction session. Our main findings are as follows: (1) Grooming state (give groom/receive groom/no groom) could be reliably decoded from IT and PMv/vlPFC in both monkeys, often even reliably predicting impending grooming transitions; (2) Decoders trained on face identity during the fixation task were able to reliably indicate the presence of social partner during the social interaction in each monkey, regardless of grooming state; (3) When neural activity was modelled as a linear combination of self- and partner joint movements, each monkey's own joint movements predicted its brain activity better than its partner's joint movements when it was grooming its partner, but the effect reversed when it was being groomed by its partner. In other words, channels modulated by the monkey's own movements were also modulated by the partner's movements; (4) Inter-brain synchrony was systematically different in each monkey when it was grooming its partner compared to when it was being groomed: multiunit activity of the groomer systematically led the multiunit activity of the partner, and this trend was reflected in some frequency bands of the local field potentials. Overall, our results show stable encoding

of relevant social variables, stable tracking of self and partner movements, and interbrain synchrony in simultaneously recorded brain activity of socially interacting monkeys.

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Poster

PSTR281: Higher Visual Areas

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National Natural Science Foundation of China grant no. U1909205

Title: A fine map of natural scene representation in primate anterior inferotemporal cortex

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Abstract: Anterior inferotemporal cortex (AIT) is known as the final stage of primate ventral visual pathway. How it encodes natural scenes is still unclear. To investigate its functional organization of natural scene representation, we used high-sensitive wide-field and two-photon calcium imaging to collect a massive dataset of AIT cortical and neural responses to more than ten thousands of natural images. A data-driven study revealed a systematic functional map in much details than early estimation, containing rich domains for surface features such as colors or textures, simple shapes such as orientation bar or edges, grades or curves. Traditional defined body and face areas could also be split into multiple sub-regions for different species and view angles. Highly specialized cortical patches even represented unique scenes like animal in cage, face or body with oriented strip. Within those functional domains, individual neurons exhibited additional selectivity and sparseness. This work provided a big-data of primate cortical and neuronal responses to natural scenes, and suggested primate AIT utilizes functional specialization and sparse code from population to single-neuron level for an efficient visual recognition.

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Poster

PSTR282: Spatial and Feature-Based Visual Attention

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Topic: E.06. Vision

Support: NIH Grant EY025648
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Title: Voxel-wise predictive encoding models reveal pre-saccadic remapping in the human visual cortex

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Abstract: Saccadic eye movements cause abrupt shifts in the visual input projected onto neurons' receptive fields (RFs). Electrophysiological studies in non-human primates have shown that the visual system predictively remaps RFs in anticipation of an upcoming saccade, known as *pre-saccadic remapping*, to support trans-saccadic visual processing. However, our understanding of remapping is incomplete, including whether remapping primarily takes the form of an displacement onto the future RF location (future field remapping) and/or a shift toward the saccade target location (convergent remapping). Evidence for distinct types of remapping remains fragmented, with little consensus over how they are implemented and distributed across the brain, particularly in humans. Here, we developed a novel framework combining whole-brain fMRI and a voxel-wise encoding model approach. Human participants (N=8) completed two fMRI sessions: a *population receptive field (pRF) mapping session* to estimate voxel-wise pRFs, and a *main experiment session* to record neural responses to noise-like visual stimulation during saccade preparation (saccade trials) vs stable fixation (no-saccade trials). Importantly, we recorded eye-tracking data to restrict analyses to trials where the stimulus was presented entirely during pre-saccadic window. Based on voxel-wise pRF, we constructed a set of encoding models for that voxel representing different hypothetical forms of remapping—future field remapping, convergent remapping, no remapping, and other variations—and evaluated how well each model predicts the voxel's observed neural response. In most visually-sensitive voxels, models incorporating future field and/or convergent remapping better predicted neural responses in pre-saccade trials than no-saccade trials, providing the first voxel-wise evidence of pRF remapping in the human brain. We identified clusters of voxels favoring future field remapping, convergent remapping, or potentially both. By offering cortex-wide, voxel-level characterization of pre-saccadic remapping in humans, this work bridges the findings from primate electrophysiology and non-invasive human neuroimaging, advancing our understanding of the neural mechanisms supporting visual stability across saccades.

Disclosures: Y. Choi: None. J.D. Golomb: None.

Poster

PSTR282: Spatial and Feature-Based Visual Attention

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Topic: E.06. Vision

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Brain and Behavior Research Foundation

Title: Representations of Ethologically Based Attention in the Frontal Oculomotor Regions

Authors: *W. HU¹, B. SEVAK², X. CHEN³;

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Abstract: Attention is essential for prioritizing sensory input based on behavioral relevance and context. While traditional salience map models emphasize the importance of low-level visual features (e.g., color, contrast) on attention allocation, emerging evidence suggests that the content of the visual scene also plays a critical role in guiding attention deployment. Human studies have shown that visual attention during free viewing of natural scenes is predominantly guided by semantic meaning. Recent findings in nonhuman primates (NHPs) further demonstrate that ethological relevance, defined by behaviorally meaningful stimuli shaped by evolutionary pressures and past experiences (eg., food, predators, and faces), attracts attention. Despite its behavioral importance, the underlying neural mechanisms of ethologically based attention (EBA) remain largely unknown. We hypothesize that EBA is represented in oculomotor regions of the frontal cortex, particularly the frontal eye field (FEF) and supplementary eye field (SEF). To address this knowledge gap, we performed high-density neurophysiology recordings in FEF and SEF of a rhesus macaque while it viewed visual stimuli with different levels of ethological salience. To dissociate attentional modulation driven by ethological relevance from confounding effects of visual feature selectivity or object identity, we employed a novel experimental design in which the stimulus inside each neuron's receptive field (RF) remained constant while the ethological relevance of distractor stimuli outside the RF was manipulated. Behaviorally, we found that fixation durations are significantly longer for both visually and ethologically salient stimuli. In addition, we observed that the fixation durations correlate with the level of ethological salience. Furthermore, we found that some SEF neurons selectively respond to ethologically relevant stimuli presented within the RF, while some others encode overall ethological salience based on the number of ethologically relevant stimuli in the visual array. These findings support our hypothesis that SEF participates in encoding both local and global ethological salience and provide new evidence that EBA engages distinct neural mechanisms in the frontal oculomotor regions.

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Poster

PSTR282: Spatial and Feature-Based Visual Attention

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Program #/Poster #: PSTR282.03/Q14

Topic: E.06. Vision

Title: Classic adaptation paradigm uncovers a novel attention-motion mechanism

Authors: W. WEI^{1,2}, C. HAN², T. OOI², *Z. HE¹;

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Abstract: Feature selective neurons in the visual cortex exhibit attentional modulation, which is thought to be a means of selecting visual information for perception. For example, attention can influence activities of visual motion direction selective neurons in the middle temporal area (MT). However, it remains unclear whether the attention system itself could also be selective with high-level feature properties (e.g., motion). Here, we present evidence for an attention-motion mechanism that responded to attention movements in perceptual space. Our psychophysical experiments measured human observers' perceived motion direction (vertical/horizontal) of an ambiguous 2x2 apparent motion stimuli (*Ramachandran & Anstis*, 1986), to reveal how perceived motion direction was affected after repeated attention movements in the vertical/horizontal direction (motion aftereffect, MAE). Results: (1) When observers viewed an adapting motion display that directed attention away from the test motion stimuli, they perceived element flickering instead of apparent motion. (2) After 75 sec of vertical or horizontal motion adaptation phase, observers perceived more horizontal or vertical motion of the test stimuli, respectively (negative MAE); however, no MAE occurred if observers had directed attention away from the adapting motion display. (3) MAE was not retinal location specific. (4) Similar visual MAE as in (2) occurred when observers repeatedly shifted attention between two horizontally separated auditory motion stimuli, made saccadic eye movements in the dark or shifted covert attention in a static visual target detection task, *without* seeing the adapting visual motion display or experiencing any object motion. (5) The measured magnitude of negative MAE increased with the number of attentional shifts rather than the duration of the adaptation phase. Overall, we found clear evidence of attention-motion adaptation, which indicates the existence of an attention-motion mechanism (Mollon, 1974). The notion of a selective attention-motion mechanism implies: (a) The principle of reducing coding redundancy applies to both the low-level visual pathways and the attention system, and (b) Feature selective attention signals can be externalized as perceptual representations.

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Poster

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Title: Infant gaze toward lower- and higher-order visual features synthesized by deep convolutional neural network

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Abstract: Humans perceive the external world by selectively processing visual information to adapt to complex and dynamic environments. Eye movement is a key behavior to achieve such active selection of visual information. Hence identifying which visual features are selected through eye movement is important for revealing the mechanisms of active visual perception. Recent studies with adult participants have shown that the gaze is attracted to higher-order visual features synthesized by a deep convolutional neural network (DCNN) (Akamatsu, Nishino, and Miyawaki 2023; Watanabe et al. 2019). However, it remains unclear when such gaze bias emerges during development. To address this issue, we recorded eye movements of one-year-old infants (n=8, mean age = 398.5 days, ranging 359 to 436 days) and adults (n=7, mean age = 19.6 years, ranging 19 to 21 years) as they viewed “feature images” — stimuli synthesized with AlexNet (Krizhevsky, Sutskever, and Hinton 2012) that isolated image features characteristic of each convolutional layer (layer 1-5) while suppressing semantic information. The feature images from different convolutional layers were presented side by side, and we determined how often participants fixated on each image over the time course following stimulus onset. Results showed that infants' gaze was attracted to the feature images from the higher layers (layer 4 and 5) of AlexNet, similarly to adults. However, infants also showed a strong preference for the feature images derived from the lowest layer (layer 1). This gaze bias specific to infants was more evident for the trials in which the higher-order feature images were paired with the lowest-order feature images. Adults gazed at the higher-order feature images more frequently than at the lowest-order ones, whereas infants gazed at both images with similar frequency. The time course of gaze frequency also showed differences between infants and adults: infants maintained a high gaze frequency for the lowest-order feature images throughout the stimulus presentation, whereas adults suppressed their gaze toward the lowest-order feature images during either the early or the middle phase of the presentation period. These findings suggest that infants acquire the gaze bias toward higher-order visual features by around one year of age but the dynamic gaze suppression toward the lowest-order visual features, which are observed in adults, may develop later.

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Poster

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Topic: E.06. Vision

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NIH NEI P30 EY001319

Title: Examining the interplay of spatial and feature-based attention in modulating visual neurons

Authors: *A. MARTIN¹, S. SHAH³, S. MAI², I. C. FIEBELKORN², F. BRIGGS⁴;
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⁴Lab. of Sensorimotor Res., Natl. Eye Inst., Bethesda, MD

Abstract: Visual attention refers to the brain's process of enhancing neuronal representations of task-relevant locations or features while suppressing irrelevant locations or features. Spatial attention, which acts as a "spotlight" to enhance neural activity corresponding to a behaviorally important location within the visual field while suppressing all others. Feature-based attention modulates neural activity related to behaviorally important stimulus features, regardless of their position in space. Although spatial and feature-based attention have been extensively studied, it remains unclear whether similar or distinct neuronal mechanisms underlie each. We addressed this open question by investigating how spatial and feature-based attention uniquely modulate neuronal activity. We first assessed neuronal feature tuning independent of attention, utilizing electrophysiological recordings to track activity of neurons in primary visual cortex (V1) in response to drifting gratings and colored bars varying across multiple feature dimensions. We then recorded from the same neurons while monkeys performed three different feature discrimination tasks utilizing a Posner cueing paradigm, whereby spatial attention shifted by block and attended features varied by task. Additionally, we examined patterns of eye movements during these tasks between animals that were head-fixed and animals that voluntarily stabilized their heads with the aid of a mask. In our neurophysiological analysis, we calculated attention index (AI) values per neuron and task and performed Tensor Component Analysis (TCA) to examine how neuronal activity varied across feature attention tasks. We hypothesized that neuronal modulation (facilitation or suppression) would be predicted by the match between a neuron's feature tuning and the attended feature. We show that neuronal activity is facilitated on tasks requiring attention to preferred features and suppressed on tasks requiring attention to non-preferred features. These results confirm that there is an interaction between neuronal feature tuning and feature-specific, attention-related changes in neuronal activity, providing insight into

the mechanisms of attentional modulation during visual processing. We also demonstrate that head-stabilized and head-fixed animals make similar patterns of microsaccades during attention trials, while demonstrating that we can track well-isolated multi-units in V1 across multiple recording sessions.

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Poster

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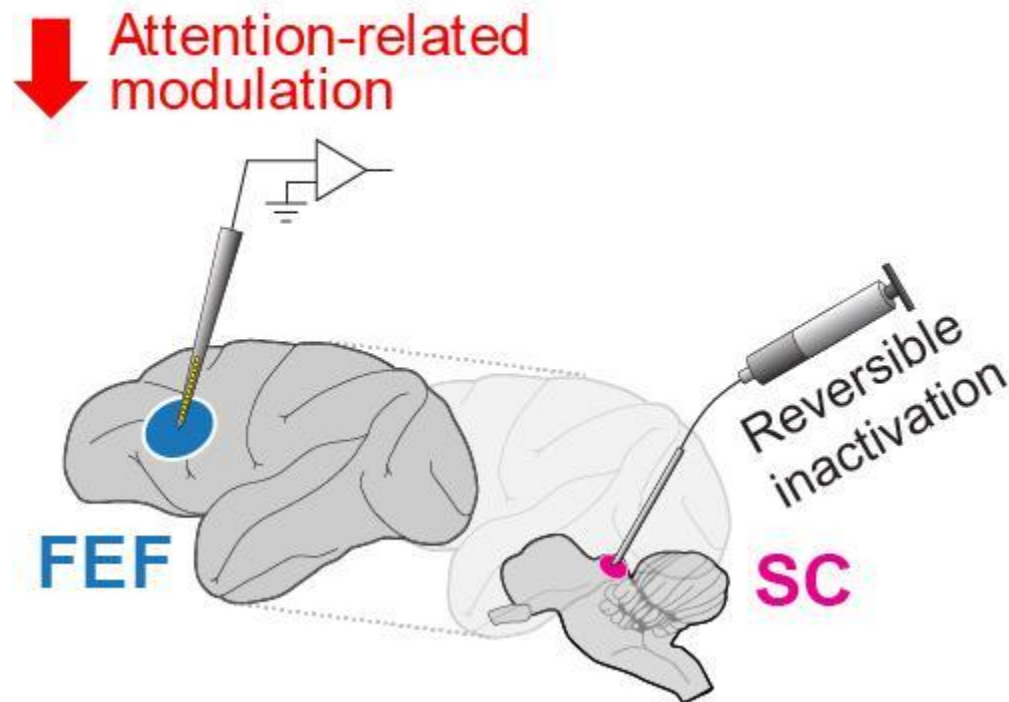
Title: Attention-related activity in the macaque frontal eye fields during inactivation of the superior colliculus

Authors: *G. W. FOLKERT^{1,2}, L. N. KATZ¹, R. J. KRAUZLIS¹;

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Abstract: The cortical frontal eye fields (FEF) and the midbrain superior colliculus (SC) are crucial for visual selective attention in primates, but how these brain regions interact to control attention is unknown. To address this gap, we investigated the effects of inactivating the primate SC on single neurons and local field potential (LFP) activity in FEF. Two monkeys performed a spatial attention task before and during pharmacological inactivation of the SC while we recorded electrophysiological activity from a multichannel probe in FEF. The monkeys maintained central fixation while patches of moving dots were presented in the left and right visual hemifields such that one of the patches was within the response fields of recorded FEF neurons. In *Attend* blocks, monkeys were rewarded for releasing a joystick in response to a change of motion direction in either of the two patches, requiring them to attend to both locations in the periphery. In *Ignore* blocks, monkeys were instead rewarded for reporting a dimming of the fixation point while ignoring the same motion patch stimuli. Before SC inactivation, FEF neurons had higher spiking activity during *Attend* trials than *Ignore* trials. LFP power was also higher during *Attend* trials than *Ignore* trials, most prominently in theta (4-8 Hz) and gamma (>30 Hz) frequency bands. During SC inactivation, deficits in task performance were accompanied by reductions in the attention-related modulation of both FEF spiking and LFP power. Specifically, in the time leading up to the motion-direction change, SC inactivation nearly eliminated the attention-related modulation of a narrow low-gamma frequency band (~30-40 Hz) and halved the attention-related modulation of theta and high gamma power. These results confirm the functional link between the SC and FEF during the performance of covert spatial

attention tasks, and establish a causal link between SC activity and theta-rhythmic and gamma-band signatures of visual selective attention in the primate prefrontal cortex.



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Poster

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Topic: E.06. Vision

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Title: Dynamic read-out of spatial remapping across saccades with time-resolved EEG alpha band decoding

Authors: *T.-Y. CHIU¹, Z. LU², J. D. GOLOMB³;

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Abstract: Our perceptual experience of the world remains remarkably stable even though saccades induce drastic shifts in retinal input. In order to achieve stability across saccades, a remapping process is required, such that spatial information at the previous retinotopic (eye-centered) location has to be removed and updated to reflect the spatiotopic (world-centered)

location. While previous neurophysiological studies found mostly pre-saccadic predictive remapping, behavioral studies on attentional remapping showed mixed evidence for both pre-saccadic remapping of attention and retinotopic attentional trace lingering after saccade completion. Here, we used multivariate decoding of EEG alpha band oscillations to efficiently and more precisely track spatial remapping across the peri-saccadic time window. We recorded eye-tracking and EEG data from human subjects performing a spatial memory task, in which they memorized a cued peripheral location during sustained fixation periods (no-saccade trials) or across a saccade. Subjects participated in two sessions: one in which they memorized the cue's spatiotopic location and another in which they memorized its retinotopic location. We trained linear classifiers to decode the location of the memory cue using alpha band oscillations on no-saccade trials, and then tested the classifiers' preference timepoint-by-timepoint on saccade trials to track the dynamics of spatial attention in the peri-saccadic time window. In both tasks, results in the pre-saccadic time window showed robust decoding of the currently attended location with no reliable decoding of the predictive remapped location. Post-saccadically, in the spatiotopic memory task, decoding of the memory cue's retinotopic location lingered early after the saccade and decoding of the task-relevant spatiotopic location emerged later after saccade completion, whereas results in the retinotopic task showed significant and prolonged decoding of the task-relevant retinotopic location and no spatiotopic decoding. Further analysis showed a significant task difference starting around 120ms after the saccade. Finally, brain-behavior correlation analysis showed that relative decoder preference in the post-saccadic time window correlated with behavioral memory precision, such that subjects with stronger spatiotopic neural decoding had more precise spatiotopic memory reports. Overall, the results provide converging evidence for the gradual remapping of spatial attention after saccades and offer an exciting new approach to study novel aspects of the fine-grain time course of remapping with human non-invasive neural recordings.

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Poster

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NEI EY036078
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Title: Diverse effects of spatial attention on neural activity across the visual cortical hierarchy

Authors: K. JACOBSON¹, A. D. LIEN¹, J. ZHUANG¹, J. DEL ROSARIO², *B. HAIDER¹;

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Abstract: Foundational studies in primates have established that spatial attention improves visual perception and neural responses, particularly in higher visual areas (HVAs) of cortex. These studies suggest that effects of visual attention are strongest in higher order areas that provide feedback to early visual areas (Buffalo et al 2010). However, the strength and timescales of attentional modulation in both excitatory and inhibitory neurons spanning multiple visual cortical areas remain unresolved. Recently, several labs have established tasks to probe spatial attention in mice. Here we performed intrinsic-imaging guided high density neural recordings (Neuropixels) in primary visual cortex (V1) and multiple higher visual areas (HVAs) from head-fixed, water-restricted mice performing a psychometric visual detection task that elicits behavioral signatures of spatial attention (Speed et al. 2020). Mice report detection of small gratings ($\sim 20^\circ$) at one of two spatial locations (either in binocular or monocular visual space); the gratings appear unpredictably in time, but in one fixed location for tens of consecutive trials. Then, without warning, the grating appears in the other location for a block of consecutive trials. This alternating spatial context elicits behavioral improvements in detection speed and accuracy across consecutive trials. Simultaneously, task-irrelevant, low contrast bars (10° wide) appear in random locations throughout the visual field; this allows measurement of neural receptive field responses that are not confounded by motor preparation or reward, while attention is at either grating location. We made acute recordings from V1 and 5 HVAs during the task (10 mice; 105 sessions; $n=1149$ putative excitatory, 325 putative inhibitory neurons). Consistent with our previous work, V1 excitatory and inhibitory neuron responses to task-irrelevant bars showed significant modulation by spatial attention ($p<0.05$). We then asked if this modulation was more prominent in visual areas further along the mouse cortical hierarchy (Siegle et al 2021). HVAs at the top of the hierarchy (preferring monocular visual space) did not show clear or prominent attentional modulation. Instead, the lower order HVAs (preferring binocular visual space) showed significant attentional modulation of excitatory and inhibitory neurons. These preliminary findings suggest that retinotopic preferences and lower rather than higher HVAs play an organizing role for spatial attention in mice. We are currently investigating how the degree of behavioral improvement relates to the strength of neural modulation, and how this varies within retinotopic subregions of HVAs.

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Poster

PSTR282: Spatial and Feature-Based Visual Attention

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR282.09/R4

Topic: E.06. Vision

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Title: Neurostimulation reveals prefrontal cortex plays a central role in endogenous but not exogenous attention

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Abstract: [BACKGROUND] Covert spatial attention improves visual perception. Human neuroimaging studies have shown that partially overlapping brain regions mediate two types of covert spatial attention: exogenous, which is involuntary and transient, and endogenous, which is voluntary and sustained. Human neurostimulation studies, using transcranial magnetic stimulation (TMS) and psychophysics, have revealed that early visual areas (V1/V2) are critical for exogenous -but not for endogenous- attention, and the right frontal eye field (rFEF+, human homologue) plays a central role for endogenous attention (Fernández & Carrasco, CurrBio 2020; Fernández, Hanning & Carrasco, PNAS 2023). Here, we used a psychophysics-TMS protocol to manipulate cortical activity in rFEF+ and investigated whether this area is also critical for exogenous attention.

[METHODS] Human observers performed an orientation discrimination task. Exogenous attention was manipulated using a peripheral cue that was either valid or invalid, preceding one of two oriented Gabor patches, or a neutral cue, which preceded both Gabor patches. We applied MRI-guided double-pulse TMS to each observer's rFEF+ during stimuli presentation, following the cues. To estimate psychometric contrast response functions (CRF), we varied stimulus contrast and analyzed sensitivity (d') in the target-stimulated and distractor-stimulated hemifields, which were contralateral and ipsilateral to the rFEF+ stimulation, respectively.

[RESULTS] When TMS was applied to rFEF+, in the distractor-stimulated hemifield both benefits at the attended location and costs at the unattended location emerged. This result is consistent with the established response gain in psychophysical experiments without TMS. Importantly, in the target-stimulated hemifield, both benefits and costs in response gain remained. This result contrasts the finding when TMS was applied to V1/V2 - the effects of exogenous attention remained at the distractor-stimulated location but were eliminated at the target-stimulated location.

[CONCLUSION] The present results reveal that rFEF+ is not critical for exogenous attention. Thus, rFEF+ and V1/V2 play distinct roles in exogenous attention. Together with our previous findings, the current results help establish a double dissociation between exogenous and endogenous spatial attention and two cortical regions: Whereas V1/V2 play a critical role in exogenous, but not endogenous attention, rFEF+ plays a central role in endogenous, but not exogenous, attention.

Disclosures: Q. Chen: None. H. Lee: None. A. Fernandez: None. N.M. Hanning: None. M. Carrasco: None.

Poster

PSTR282: Spatial and Feature-Based Visual Attention

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Program #/Poster #: PSTR282.10/R5

Topic: E.06. Vision

Title: Direct thalamic recordings reveal evoked responses are associated with improved visual search performance and parietal alpha desynchronization

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Abstract: Visual attention enables selective processing of relevant information and suppression of distractions to support efficient perception and goal-directed behaviour. The thalamus is posited to play a role in filtering and routing visual information to higher order cortex, but evidence in humans remains extremely limited. We leveraged direct intracranial thalamic recordings in tandem with eye-tracking data from children with epilepsy undergoing clinical monitoring ($n = 6$) to investigate thalamocortical interactions during a cluttered scene visual search task. We report a visually evoked response in the human medial thalamus, emerging ~200ms after stimulus onset - following the canonical visual cortex evoked response (Fig. 1A). Using linear mixed effects models, we showed that higher amplitude evoked responses were associated with faster time to first fixation and reduced overall search time ($p = 0.003$ and $p = 0.001$, respectively; Fig 1B), linking thalamic activation to both overt orienting and task efficiency. The magnitude of the thalamic response was also associated with greater alpha-band desynchronization in the superior parietal cortex ($p_{\text{bonferroni}} = 0.02$; Fig. 1C), a pattern linked to increased attentional flexibility. These findings suggest that the thalamus may facilitate cortical engagement of task-relevant regions to support goal-oriented behaviour. This work advances emerging models of thalamocortical integration in cognition and informs future neuromodulation strategies to improve attentional function in clinical populations.

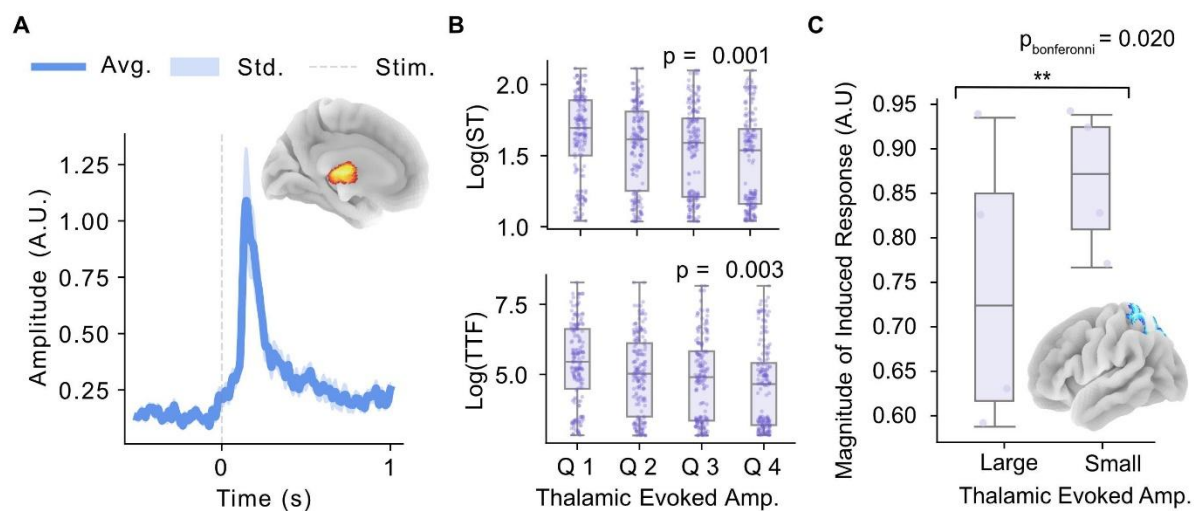


Figure 1. (A) Average evoked response in medial thalamus across subjects (solid blue line), with shaded area representing standard deviation. Evoked activity peaks ~200ms after stimulus onset. **(B)** Greater evoked response amplitudes associated with faster time to first fixation (TTF) and shorter overall search time (ST). **(C)** Larger evoked responses associated with stronger alpha band desynchronization in the superior parietal cortex.

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Poster

PSTR282: Spatial and Feature-Based Visual Attention

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Program #/Poster #: PSTR282.11/R6

Topic: E.06. Vision

Support: Whitehall Foundation
Research to Prevent Blindness

Title: Spatiotemporal fluctuations in visual detection sensitivity following a visual stimulus or an eye movement

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Abstract: Sensitivity for faint visual targets is not fixed but varies from moment to moment. One factor that has been proposed to influence visual sensitivity is the state of intrinsic fluctuations in cortical activity that can travel as waves across the cortical surface (Davis, Muller, et al., Nature 2020). Sensory and motor events such as visual stimuli and saccades generate traveling waves of cortical activity (Muller et al., Nat. Communi. 2014; Zanos, et al., Neuron 2015) but it is unknown whether these evoked waves also modulate sensitivity like intrinsically generated traveling waves. To test this, we asked healthy adult human subjects to report their detection of faint visual targets while triggering evoked responses (a visual stimulus or an eye movement) that putatively form traveling waves along a foveal-peripheral axis in the visual cortex. The targets appeared randomly at various eccentricities and delays with respect to the timing of the wave-triggering event and were titrated in size and contrast to achieve similar levels of performance. We tracked eye movements as subjects reported their detection of faint visual targets with a mouse button press. Detection performance followed a rhythmic time course with respect to trigger onsets consistent with previous reports (Fiebelkorn et al., Current Biology 2013; Hogendoorn, J. Cog. Neurosci. 2016). When we separated detection performance based on target eccentricity, we found that the modulation time course shifted following a progression consistent with the rhythmic fluctuation traveling as a wave. These results were consistent with a model that generated similar rhythmic performance modulations by activity propagation in the

human visual cortex across the retinotopic map. This data supports the view that spatiotemporal fluctuations in cortical activity modulate sensory processing and perceptual sensitivity and suggest that responses to stimuli or eye movements generate secondary fluctuations that can impact visual function.

Disclosures: **Z.W. Davis:** None. **D. Jensen:** None. **A. Royston:** None.

Poster

PSTR282: Spatial and Feature-Based Visual Attention

Location: SDCC Halls B-H

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Program #/Poster #: PSTR282.12/R7

Topic: E.06. Vision

Support: T32 EY024234
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Research to Prevent Blindness
F32 EY02259

Title: Cortical Traveling Waves Persist Under States of Attention

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Abstract: Intrinsic fluctuations in cortical activity often exhibit coherent spatiotemporal structure within a cortical area, which can be described as a traveling wave. These intrinsic traveling waves (iTWs) reflect fluctuations in the relative excitability of neuronal populations and are predictive of performance on visual behavioral tasks (Davis, Muller et al., 2020; Davis, Busch, et al., 2024). The enhancement of sensory information through shifts in intrinsic excitability resembles the effects of the allocation of attention on cortical populations. Yet, one of the benefits of attention is the reduction of low frequency correlated variability (Cohen and Maunsell, 2009; Mitchell and Reynolds, 2009), which can reduce the sensitivity of cortical populations (Nandy et al., 2019). Traveling waves have been hypothesized to be a source of correlated variability (Huang et al., 2019), raising the possibility that attention attenuates iTWs or reduces their impact on cortical function. Alternatively, it has been proposed that iTWs need not be a source of correlated variability (Davis, Benigno et al., 2021), leaving room for a functional interaction with states of attention. To test between these alternatives, we analyzed a previously published dataset of bilateral Utah array recordings in V4 of a macaque monkey performing a spatially cued orientation change detection task (Mayo and Maunsell, 2016; Parkash, et al., 2021). We found that attention reduced spike-LFP phase coupling and increased the spatial heterogeneity of phase patterns across the array contralateral to the attended hemifield. These results are consistent with attention desynchronizing the neuronal population. Despite this, iTWs were still present in the attended array, and the spatial patterns were

informative of trial outcome. These results argue against the view that iTWs are attenuated by the allocation of attention and suggest an interaction between the state of iTWs and states of attention.

Disclosures: D. Jensen: None. J. Mayo: None. Z.W. Davis: None.

Poster

PSTR282: Spatial and Feature-Based Visual Attention

Location: SDCC Halls B-H

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Program #/Poster #: PSTR282.13/R8

Topic: I.05. Working Memory

Support: NHMRC Grant APP1156733

Title: The influence of NMDAR antagonists on prefrontal cortex neuron activity during a touchscreen task of working memory in mice

Authors: *G. JIN¹, M. HUDSON¹, R. A. HILL², L. INGER¹, Y. LIU³, C. VINNAKOTA², N. C. JONES¹;

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Abstract: NMDAR antagonists impair working memory (WM), but their effects on underlying neuronal activity remain unclear. Here we examined multi-unit neuronal activity in the medial prefrontal cortex (mPFC) of C57BL/6 mice during the Trial-Unique Nonmatch to Location (TUNL) task of working memory. We aimed to establish differences in firing rates between correct and incorrect trials during the maintenance phase, and to investigate how this changed after exposure to the NMDAR antagonists ketamine and MK-801.

Male C57BL/6 mice (n=6) were trained to criterion on the TUNL task of working memory in automated touchscreen operant chambers, and then were implanted with a tetrode in the left mPFC. After recovery, mice completed TUNL sessions under saline, 30 mg/kg ketamine, 0.1 mg/kg MK-801 or 0.3 mg/kg MK-801 treatment conditions. Simultaneous tetrode recordings were carried out across all sessions. Electrophysiological data was processed through Kilosort 2 and Phy 2 to identify spikes, and time-stamped to specific behavioural task stages. Multi-unit firing rate was calculated across the baseline period and during the maintenance phase of each trial. Mice repeated sessions with the tetrode lowered where possible for a total of 88 sessions. Both ketamine and MK-801 significantly reduced TUNL task accuracy, reflecting WM impairment (1-way ANOVA $p = 0.0014$, saline VS ketamine; $p = 0.031$, saline VS low dose MK; $p = 0.0041$, saline VS high dose MK). Comparing baseline activity across drug conditions, we also found ketamine and MK-801 to significantly increase MUA firing rate (Kruskal-Wallis $p < 0.0001$, saline VS ketamine; $p = 0.0095$, saline VS low dose MK; $p < 0.0001$, saline VS high dose MK). However, in our preliminary data we have found no changes in maintenance phase firing rate or relative changes from baseline to maintenance periods when comparing correct and

incorrect trials across each treatment.

We conclude there are overall no MUA differences between correct and incorrect trials during the maintenance phase of WM. While NMDAR antagonists increase mPFC activity during WM, most likely via disinhibiting neuronal firing, WM impairments induced by NMDAR antagonists may involve more complex mechanisms.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Program #/Poster #: PSTR283.01/R9

Topic: F.02. Cerebellum

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Title: Altered cerebellar Purkinje cell excitability and synaptic transmission in the IB2 KO mouse model of autism

Authors: *E. PALI¹, S. MASOLI¹, G. PELLAVIO¹, P. TUPPUTI¹, L. MAPELLI¹, F. PRESTORI¹, E. D'ANGELO^{1,2};

¹Univ. of Pavia, Pavia, Italy; ²IRCCS Mondino Fndn., Pavia, Italy

Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental condition characterized by impairments in social interaction, restricted interests, and repetitive behaviors. Growing evidence implicates the cerebellum in ASD pathophysiology. Deletion of the IB2 gene, commonly associated with Phelan-McDermid syndrome, leads to severe motor impairments and core autistic traits. IB2 knockout (KO) mice recapitulate these phenotypes, displaying motor, social and cognitive deficits. Furthermore, previous studies using the IB2 KO mouse model have reported altered synaptic plasticity in the cerebellar granular layer and morphological abnormalities in Purkinje cells (PCs). To investigate how IB2 loss affects PC function, we performed whole-cell patch-clamp recordings in acute cerebellar slices from 18- to 24-day-old IB2 KO and wild-type (WT) mice of either sex. While PCs from WT mice exhibited typical electrophysiological properties (n= 14), those from IB2 KO mice segregated into two distinct groups. The first group (IB2 KO-1) showed reduced firing frequency and excitability (n= 10). The second group (IB2 KO-2) displayed burst-like spontaneous activity, increased excitability (n= 8), and complex bursts in response to depolarizing currents. Interestingly, blocking inhibition during depolarizing current steps reduced both the firing rate and complex bursts in IB2 KO-2, aligning its activity with that of the IB2 WT PCs. This suggests that inhibitory

mechanisms contribute to the observed electrophysiological differences. Furthermore, in response to five-pulse parallel fiber stimulation at varying frequencies (4-200 Hz), IB2 KO-2 PCs exhibited an abnormal response with an initial burst followed by a prolonged pause, suggesting impaired synaptic transmission at the parallel fiber-PC synapse. To further investigate these alterations, we are developing a computational model of PCs based on morphological reconstructions from recorded cells, designed to replicate the properties of WT, KO-1, and KO-2 PCs. Overall, our findings strengthen the evidence for cerebellar involvement in ASD, revealing two distinct, dysregulated PC subtypes and disrupted inhibitory modulation in the IB2 KO mouse model. These alterations suggest complex physio-pathogenetic processes, potentially involving compensatory mechanisms and dysfunction in inhibitory circuits, which need further investigation.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.02/R10

Topic: F.02. Cerebellum

Support: VBT The Virtual Brain Twin Project has received funding from the European Union's Research and Innovation Program Horizon Europe under grant agreement No 101137289.

Title: In-vivo characterization of cerebellar modulation of prefrontal cortex activity in anesthetized mice

Authors: *M. CONFORTI¹, D. DI DOMENICO¹, C. CIAPPONI¹, I. MONTAGNA¹, P. TUPPUTI¹, S. TRITTO¹, L. MAPELLI¹, E. D'ANGELO^{1,2};

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Abstract: The functional connectivity between the cerebellum and neocortical regions, like the medial prefrontal cortex (mPFC), strongly supports the cerebellum role in cognitive processes. Alterations in cerebello-prefrontal cortex (mPFC) connections characterize several cognitive dysfunctions (such as schizophrenia and autism spectrum disorders), suggesting that the cerebellum might play a critical role in the proper functioning of the mPFC. Cerebellar activity might impact on mPFC neuronal activity through two different projection pathways: the dopaminergic one, through the ventral tegmental area, and the glutamatergic one, through the mediodorsal and ventrolateral nuclei of the thalamus. Despite these findings, the impact of cerebellar activity on mPFC requires clarification. To get a deeper insight into the process of cerebellum-mPFC communication, we performed single-unit recordings *in vivo* from the prelimbic area (PrL) of mPFC in anesthetized C57BL/6 adult mice of either sex during the

stimulation of the contralateral cerebellar dentate nucleus (DN). The DN electrical stimulation elicited a pause in the spontaneous firing of most PrL neurons, sometimes followed by an excitatory rebound (n=86, N=45). Similar responses were obtained by DN optogenetic stimulation after viral injection causing the expression of ChannelRhodopsin (ChR2) (n=5, N=3). To investigate the nature of PrL responses, we locally superfused the PrL with antagonists of GABA-A receptors (gabazine), D1-like and D2-like dopamine receptors (SCH23390 and Sulpiride, respectively), and NMDA and AMPA glutamate receptors (NBQX, D-APV and 7Cl-kynurenate). Gabazine perfusion confirmed the inhibitory nature of the pause, while the blockade of dopaminergic transmission modulated PrL neurons spontaneous firing without abolishing pause responses. Perfusion of glutamate receptor antagonists almost completely abolished the basal discharge of PrL neurons along with any response to DN stimulation. Our data show that cerebellar activation transiently decreases PrL activity engaging local inhibitory circuits most likely through thalamic projections, while dopaminergic transmission regulates PrL basal discharge. *In vivo* calcium imaging of multiple neurons is undergoing to obtain further insights on the nature of these electrophysiological results. Overall, these findings provide evidence for a complex cerebellar modulation of PrL activity.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Program #/Poster #: PSTR283.03/R11

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Title: Cerebellar Dopamine Signaling Shapes Network Activity: Receptor-Specific Mechanisms and Antipsychotic Modulation

Authors: *P. S. FARIS¹, I. HEREDIA ORTIZ¹, A. VERGANI², E. D'ANGELO¹;

¹Dept. of Brain and Behavioral Sci., Univ. of Pavia, Pavia, Italy; ²Dept. of Brain and Behavioral Sci., Univ. of Pavia, Italy, Italy

Abstract: The cerebellum integrates sensorimotor and cognitive inputs and maintains reciprocal communication with dopaminergic systems, which are altered in schizophrenia (Faris et al 2024). Cerebellar neurons synthesize dopamine (DA), express all the DA receptor types, receive DAergic inputs, in turn, regulate midbrain DAergic nuclei. Despite this, the physiological role of DA signaling and the effect of antipsychotic drugs in the cerebellar network remains unclear. Here, we performed in vitro high-density microelectrode array (HD-MEA) recordings of acute

sagittal slices of the cerebellar vermis (lobule VI) from juvenile (P18-24) C57BL/6J mice of both sexes, focusing on spontaneous Purkinje cell (PC) firing as the sole cerebellar cortical output and mossy fiber-evoked granule cell local field potentials (LFPs, including N2a/N2b) as the main regulatory input. The modulatory effects of DA and clozapine (atypical antipsychotic) were assessed. Statistical analysis was performed using one-way ANOVA; $p < 0.05$. DA perfusion (0.5-100 μM) induced a dose-dependent increase in PC firing rate ($N=3$ slices, $n=181$ channels) demonstrating excitatory modulation. In contrast, DA mostly reduced N2a and N2b peak amplitudes ($N=3$, $n=121$) in the granule layer evoked by single-pulse mossy fiber stimulation (0.1Hz), indicating decreased synaptic responsiveness. The D1-like receptor antagonist SCH23390 (10 μM) strongly suppressed PC firing and blocked DA excitatory effect ($N=2$, $n=73$). This suppression reversed after washout, confirming a D1 receptor-dependent mechanism. Clozapine (15 μM) suppressed PC firing rate. Conversely, sulpiride (10 μM) did not affect basal PC activity but partially reversed the effect of clozapine ($N=2$, $n=104$), implicating D2 receptors in clozapine modulation of PC firing. In the granule layer, clozapine produced heterogeneous effects on N2a/N2b peaks, with either increases or decreases in different recording sites ($N=3$, $n=151$). Co-application of sulpiride attenuated these changes, partially restoring N2a/N2b amplitudes ($N=2$, $n=80$), supporting D2 receptor pathways involvement in granule cell network modulation. In summary, these preliminary findings reveal receptor- and layer-specific dopamine modulation in cerebellum, with D1 receptors controlling the PC output and D2 receptors the granule cell input. These observations support cerebellum as a potent target for antipsychotic drugs in schizophrenia. Ongoing work is examining how the DA system influences plasticity. Moreover, these observations are being incorporated into computational models to predict the impact of DA and DAergic drugs on network dynamics in schizophrenia.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

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Program #/Poster #: PSTR283.04/R12

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Integrated, multi-scale computational models of patient patho-physiology ('virtual twins') for personalized disease management #101137289 — VIRTUAL BRAIN TWIN — HORIZON-HLTH-2023-TOOL-05

Title: Personalized multiscale brain modeling unveils disruption of brain dynamics in Parkinson's disease patients: Toward the identification of neural markers and the development of brain digital twins

Authors: *D. PISCHEDDA¹, M. VITACCA¹, F. PALESI¹, R. M. LORENZI¹, A. MONTEVERDI², A. TESSITORE³, R. DE MICCO³, M. SICILIANO³, F. ESPOSITO³, C. A. M. GANDINI WHEELER-KINGSHOTT^{4,1,2}, E. D'ANGELO^{1,2};

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Abstract: Parkinson’s disease (PD) is a heterogeneous progressive neurodegenerative disorder that primarily affects motor function. The spread of PD worldwide is increasing more rapidly than other disorders of the nervous system, thus producing a significant social and economic impact on society. PD is characterized by the degeneration of dopaminergic neurons in the substantia nigra, leading to a cascade of motor (e.g., slowness of movement, tremor, rigidity) and non-motor (e.g., cognitive decline, sleep disturbances) impairments. Although experimental research has significantly advanced our understanding of PD, its underlying causes are not completely understood, and no cure exists. Current treatments primarily target symptom management rather than disease modification, and the response to therapy varies widely among patients, underscoring the need for tailored approaches. We aim to develop Brain Digital Twins (BDT), namely computational personalized models that simulate brain dynamics (D’Angelo & Jirsa, 2022), to unravel the complex mechanisms of PD and to optimize therapeutic interventions. Therefore, we built subject-specific models to reveal biomarkers of disease severity. We developed a multiscale model of brain circuits to simulate neural dynamics in early-stage PD patients (N = 32, 17 women, age 64.3 +/- 8.7 years) and healthy controls (HC, N = 15, 7 women, age 60.6 +/- 5.3 years). We exploited structural and functional magnetic resonance imaging data in the framework of The Virtual Brain (TVB) modeling (e.g., Monteverdi et al., 2023) to derive parameters of connectivity and excitation/inhibition reflecting macroscale properties. These parameters were extracted for both whole-brain and resting-state networks. We used independent sample tests to compare TVB parameters of PD patients and HCs and identify neural markers of the disease. Compared to HCs, PD patients showed significantly reduced inhibitory local coupling at the whole-brain level and decreased long-range connectivity and inhibitory local coupling within the sensorimotor network, but not within other resting-state networks. These results reflect the predominantly motor impairment in early-stage PD patients and confirm previous findings that emerged using other analysis approaches (e.g., Tolosa et al., 2021), thus proving the effectiveness of the method in simulating brain dynamics in PD populations. This is a first step toward creating BDTs, which will serve as valuable tools to gain a more integrated view of the disease and provide a platform for improving treatment selection and evaluation. Thus, BDT could transform our approach to the diagnosis, monitoring, and treatment of PD.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Title: The impact of cerebellar zebrin stripes on large-scale brain dynamics

Authors: ***R. M. LORENZI**¹, M. DE GRAZIA¹, S. TARTABINI¹, A. GEMINIANI^{1,2}, C. GANDINI WHEELER-KINGSHOTT^{3,1,4}, C. CASELLATO¹, F. PALESI¹, E. D'ANGELO^{1,4};
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Abstract: The cerebellar cortex is organized in parasagittal stripes of Purkinje cells (PC) differing in Zebrin II expression, separating zebrin-negative (PCz⁻) and zebrin-positive (PCz⁺) subpopulations with distinct properties. The zebrin specialization has been described at cellular level but whether it impacts on brain recordings, e.g. the Blood Oxygenation Level Dependent (BOLD) signals, remains unexplored, due to the challenge of linking cellular to macroscale brain physiology. We address this gap by developing a multiscale modeling framework that embeds zebrin specificity into mesoscale mean field models of the cerebellar cortex and integrates them with subject-specific structural connectivity to construct virtual brains able to explore in-silico how neuronal properties shape large-scale signals (Sanz-Leon et al 2013). Using a cerebellar spiking neural network with differentiated PCz⁻ and PCz⁺ dynamics as microscale functional reference, we derived zebrin-specific transfer functions that are the input/output relations of PCz⁻ and PCz⁺ at population (mesoscale) level while retaining single-cell features. These transfer functions were used to define a zebrin-specific cerebellar mean field model (zCRBL MF). Virtual brains of 9 healthy volunteers (Human Connectome Project, 6 women, 34±4 years) were constructed using individual structural connectivity, computed from diffusion weighted MRI, and associating each cerebellar cortical node with a zCRBL MF, while deep cerebellar nuclei (DCN) and brainstem nodes were represented with generic mesoscale models. Specifically, PCz⁻ and PCz⁺ regions were identified by grouping micro-zones based on the majority of PCz⁻ (or PCz⁺) that belongs to a specific anatomical macro-region, according to literature. Subject-specific resting-state simulations of a cerebellar-brainstem network were performed using these models to generate synthetic BOLD signals (Lorenzi et al 2025). At microscale, PCz⁻ fired ~50 Hz more than PCz⁺ across the input frequency range (4-200 Hz), yielding distinct population transfer functions: quasi-sigmoidal for PCz⁻, sharp on-off for PCz⁺. For each subject, virtual brain simulation showed a higher PCz⁻ firing rate than PCz⁺ for all cerebellar cortical regions,

confirming zebrin functional identity is preserved at macroscale. Spectral analysis of simulated BOLD signals in DCN revealed statistically significant zebrin-driven frequency shifts (Wilcoxon test, $p < 0.05$). Our framework, by enabling non-invasive assessments of cerebellar contributions to stripe-dependent mechanisms, opens the way to study Z- and Z+ involvement in compensatory mechanisms in neurodegeneration.

Disclosures: R.M. Lorenzi: None. M. De Grazia: None. S. Tartabini: None. A. Geminiani: None. C. Gandini Wheeler-Kingshott: None. C. Casellato: None. F. Palesi: None. E. D'Angelo: None.

Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.06/R14

Topic: F.02. Cerebellum

Support: Ebrains 2.0
Ebrains Italy
Virtual Brain Twins

Title: Synaptic modulation of Purkinje cell responses in a cerebellar circuit model

Authors: *M. RIZZA¹, D. BENOZZO¹, M. DE GRAZIA¹, S. MASOLI¹, F. MARCHETTI¹, F. PRESTORI¹, C. CASELLATO¹, E. D'ANGELO^{1,2};

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Abstract: Purkinje cells (PCs), located in the cerebellar cortex, are known for their extensive and planar dendritic arbor that performs specific computations for motor control, coordination, and learning. This arborization allows to receive a high volume of synaptic inputs from parallel fibers (pfs) that form thousands of excitatory synapses onto PC spines and from molecular layer interneurons (MLIs) - stellate cells (SCs) and basket cells (BCs) - that inhibit PCs. The dendritic architecture contributes to spatial and temporal signals summation: PCs integrate excitatory and inhibitory inputs over space (different parts of the dendritic tree) and time, allowing to modulate their response based on the input timing and spatial distribution. This dendritic processing it is still not clear. Starting from experimental patch-clamp recordings, we reconstructed multi-compartmental biophysically realistic PC, SC and BC models in Python-NEURON. For the circuit simulations, we used the Brain Scaffold Builder framework using cell placement data and connectivity rules based on experimental data to build the entire cerebellar network. We investigated the intrinsic and synaptic electrophysiological properties of the neurons involved in the circuit and their impact on PC model responses. PCs generated the characteristic burst-pause responses in reaction to pf input bursts, with the pause reinforced by MLI inhibition. SCs modulated PCs activity through dendritic GABAergic synaptic inhibition, instead BCs through somatic GABAergic synaptic inhibition. Simulations predicted BC and SC filtering of PC

responses, highlighting the different role of SCs and BCs in frequency modulation of PC discharges. Simulations showed that the intensity and bandwidth of molecular layer filtering were modulated by the number of active synapses between pfs-SCs-PCs and pfs-BCs-PCs. SCs and BCs emerged as critical elements controlling cerebellar processing in time and frequency domains. Tuning of transmission bandwidth and delay through specific membrane and synaptic mechanisms contribute to explain the role of SCs and BCs in motor learning and control.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Program #/Poster #: PSTR283.07/R15

Topic: F.02. Cerebellum

Support: MNESYS PE0000006 (DN. 1553 11.10.2022)

Title: Computational modeling of NMDA receptor alterations in the cerebellar granule cells of the IB2 autism mouse model

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Abstract: Autistic spectrum disorders (ASD) pervasively affect development and highly impact social interaction. Mice engineered with mutations in neurexin/neuroligin genes, Shank proteins and the tightly linked IB2 gene exhibit ASD-like behavioural deficits, like delayed motor skill acquisition. The role of the cerebellar in ASD has been well documented, showing a dependent association between cerebellar damage and increased ASD risk (Bauman, Kemper 2005). In this study, we focused on the cerebellar granule cells (GrCs). Experimental data from acute cerebellar slices of IB2 knock-out (KO) mice revealed enhanced intrinsic excitability, increased voltage-dependent Na and K currents, larger NMDA receptor-mediated currents, increased tonic release and ambient glutamate, and a higher release probability (Giza et al., 2010, Soda et al., 2019). Based on this empirical evidence, we re-parameterized a wild-type (WT) granule cell (GrC) multi-compartment model (Masoli et al., 2017) to reflect the observed changes. Specifically, Na and K maximum ionic conductances were increased by a factor of ~1.6, and a tonic glutamate level was included into the mossy fibers - GrC synaptic model to match the observed shift of the input-output (I-f) relationship. Since ambient glutamate increases baseline NMDA receptor occupancy, the resulting tonic activation reduces the available receptor resources for responding to presynaptic signals. Therefore, NMDA maximum conductance was optimised with the ambient glutamate level to match both the observed I-f and NMDA current changes. The source of ambient glutamate is due to spontaneous and spike-independent

exocytosis from the mossy fibers terminals (Balmer, Borges-Merjane, and Trussell 2021). Our model predicted a baseline ambient glutamate of $\sim 11 \mu\text{M}$, and a NMDA conductance change of ~ 4 times. These alterations in IB2-KO GrCs affected the microcircuit, specifically modifying the E/I balance in the granular layer (Soda et al., 2019). To model this, we used a bottom-up framework to build a representative cerebellar microcircuit. This multiscale approach enables predictions of circuit-level dynamics from single-cell modifications. Our results reproduced the broader and higher E/I balance around IB2-KO GrCs. Ongoing validation compares model predictions with in vitro MEA recordings from both WT and IB2-KO mice under both spontaneous and stimulus-driven conditions. Each cell type was reproduced through a multi-compartment model interfaced with the NEURON simulator (Carnevale, Hines 2006). The entire process was managed by the Brain Scaffold Builder framework (De Schepper et al., 2022).

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.08/R16

Topic: F.02. Cerebellum

Title: Immunohistochemical profiling of neuronal cell types in the human cerebellar cortex

Authors: *G. SENGUL¹, V. TURGUT², E. CANDAR², I. DEMIRCUBUK³;

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Abstract: The human cerebellar cortex exhibits a highly organized neural architecture critical for motor coordination and balance. This study aimed to delineate the cellular composition of the cerebellar cortex through immunohistochemical analysis. Six formalin-fixed cerebellar tissue samples from the vermis were processed using antibodies against NeuN, CART, vGLUT1, parvalbumin (Pv), and calbindin (Cb). NeuN labeling demonstrated a widespread distribution of neurons, with pronounced nuclear staining in the granule cell layer. Cb displayed strong immunoreactivity in the Purkinje cell layer and molecular layer. Pv immunoreactivity was observed in Purkinje cells, as well as in stellate and basket cells of the molecular layer—interneurons that play key inhibitory roles in cerebellar circuitry. vGluT1 staining highlighted mossy fiber terminals within the granule cell layer and parallel fiber synapses in the molecular layer, indicating active glutamatergic transmission in these regions. CART-positive neurons were identified in the molecular layer, with weaker staining in Purkinje cells. Partial labeling of climbing fibers—likely originating from the inferior olivary nucleus—was observed in the molecular layer. This study provides the first comprehensive immunohistochemical characterization of cell types in the human cerebellar cortex, underscoring their distinct neurochemical phenotypes. The findings enhance our understanding of the cerebellum's cellular heterogeneity and its involvement in complex neural networks.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

Support: NINDS Grant NS116893
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Title: Cerebellar cortical alternating current (AC) stimulation induces changes in Purkinje cell complex spike activity

Authors: B. AVLAR¹, E. CETINKAYA¹, R. RAHMAN¹, M. SAHIN², *E. J. LANG¹;
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Abstract: Transcranial stimulation of the cerebellum has shown potential as a therapeutic modality. Yet, its effects on cerebellar activity are relatively unknown. In particular, how it might induce long-term changes in brain function that underlie its therapeutic actions is not known and such knowledge would be useful to optimize its efficacy. To begin to address this issue, extracellular single unit recordings were made from Purkinje cells (PCs) in ketamine-xylazine anesthetized rats. Recordings were made from the apices and walls of several hemispheric lobules. In particular, we sought to determine the effect of alternating current (AC) stimulation (0.5 Hz) applied to the cerebellar surface on complex spike (CS) activity, as CSs are thought to modulate plasticity in the cerebellar cortex that could underlie persistent changes in cerebellar functioning. Changes in CS waveform and in CS firing rate were observed. The waveform changes reflected both a steady-state reduction in CS size (measured as the absolute area of the CS), and a phase-dependent modulation in CS size. Most often CSs that occurred during the negative phase of the AC cycle were smaller in size, and this decrease was generally due to the loss or reduction of spikelets at the end of the CS. The magnitude of the phasic changes in CS waveform often correlated with the strength of the induced simple spike modulation of the same cell. In terms of CS firing rate changes, approximately half of the CS population showed significant firing rate modulation during AC stimulation, as determined by significant circular mean vectors ($p < 0.05$). As CSs gate plasticity at the parallel fiber-PC synapse, these changes in CS timing and waveform have potential implications for transcranial stimulation's ability to induce long term changes in cerebellar function. For example, controlling the timing of CSs could allow one to time when plasticity occurs. Moreover, there is evidence that the number of spikelets in a CS controls the strength and type of plasticity (e.g., LTP vs LTD; Mathy et al. 2009 Neuron 62:388-99) that is induced. Thus, control of CS waveform by AC stimulation raises the possibility of biasing plasticity towards a particular form of plasticity, one which may be better for treating a specific disorder.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

Support: NIH Grant R35-NS116854

Title: Projection and functional role of trigeminocerebellar mossy fibers in whisker-related sensorimotor integration by crus I/II

Authors: T. ROSSI, *I. M. RAMAN;
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Abstract: Whisker-related sensory signals in mice are conveyed to the cerebellum by mossy fibers (MFs) primarily originating in the principal (Pr5) and spinal interpolaris (Sp5i) trigeminal nuclei, in which action potentials carry sensory information, including tactile input to the face. In previous work, recording in vivo from crus I/II Purkinje cells, we observed well-timed inhibition in response to tactile whisker input, and broadly timed excitation in association with whisks. Previous studies from several labs have shown that granule cells receive convergent information from multiple sensory modalities. These observations raise the question of the extent to which sensory trigeminocerebellar streams converge to permit specific computations relevant to whisker-related sensorimotor integration. Here, we investigate the anatomical overlap and/or segregation of trigeminocerebellar MFs carrying sensory signals to whisk-related regions of the cerebellar cortex, and their contribution to well-timed firing of Purkinje cells. Trigemino-cerebellar MFs were identified by injection of AAV1.hSyn.hChR2(H134R)-eYFP in Pr5 or Sp5i. The results show that 79.9% of Pr5-MFs terminate in the ipsilateral cerebellar hemisphere, with 15.6% in crus I and 64.3% in crus II (6 mice). Similarly, 78.5% of Sp5i-MFs end in the ipsilateral cerebellar hemisphere, with 22.5% in crus I and 56% in crus II (1 mouse). Results from a single Gabra6-cre mouse, which received an injection of AAVDJ.EF1a.DIO.iRFP670 in crus I/II and AAV1.CAG.tdTomato in Sp5i as well as the injection in Pr5, show that trigeminocerebellar MFs lie in close proximity to their postsynaptic target, making convergence at the level of some granule cells appear possible. At the same time, regions of segregation were evident, consistent with the idea that crus I/II Purkinje cells may receive at least some information from separate populations of sensory-related granule cells. Optogenetic activation of both types of MFs in awake head-fixed mice engaged strong suppression of Purkinje cell simple spikes with similar onset and duration, without eliciting whisker movement. Specifically, during extracellular recordings from crus I/II Purkinje cells, optogenetically activating Pr5-MFs evoked simple spike suppression (N=48/86), acceleration (N=10/86) or no response (N=28/86, 7 mice). Similarly, activation of Sp5i-MFs most often

elicited suppression (N=17/27), with rarer acceleration (1/27) or no change (9/27, 2 mice). The preferential recruitment of feedforward inhibition suggests that sensory-specific microcircuits shape spiking patterns of Purkinje cells to integrate whisker-related sensory information.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

Support: Whitehall Foundation
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Chan Zuckerberg Initiative

Title: Temporal Expectation Signals during Adaptive Motor Timing Behavior

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Abstract: Temporal expectation—the ability to anticipate when events will occur based on prior experience—is fundamental to learning, perception, and action. It relies on a distributed brain network including the frontal and parietal cortices, basal ganglia, hippocampus, and cerebellum. Both the cerebellum and posterior parietal cortex (PPC) are implicated in generating internal models that predict future events, from sensorimotor to reward outcomes. These models are refined by comparing predictions to actual sensory feedback, generating prediction errors that drive plasticity. Although evidence supports the presence of such internal models in both cerebellum and PPC, it remains unclear whether they function independently or interact to form a unified internal representation of the external world. We address this question in the context of a novel behavioral paradigm in which mice are required to perform a temporally precise motor task, involving two lever pushes which are either self or externally timed. The interval timing between pushes changes unexpectedly to investigate prediction errors and adaptation. In this task, animals use a mixed strategy, integrating contextual knowledge and real-time prediction errors to shape future movements and update internal models. To investigate the role of the cerebellum in this adaptation process, we used two-photon calcium imaging of Purkinje cell dendritic activity during behavior. We found that information about outcome and context can be decoded from the population activity of these areas. Using optogenetic manipulation of cerebellar and PPC neurons, we examined the role of these areas in adaptive behavior and observed that such adaptation may be impaired by this manipulation. These results reveal that

both cerebellar and cortical regions contribute to temporal expectation and adaptive behavior. Ongoing work will identify functionally distinct neuronal subpopulations in each area and clarify how they interact to generate predictions and correct errors.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

Support: NIH Grant R01 DC018061-06

Title: Identifying complex spike activities from multi-channel electrode recording in vestibular cerebellum using a deep learning-based sorting technique

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Abstract: The Vestibular cerebellum integrates multi-sensory signals of self-motion for movement coordination and balance control. This computation takes place in cerebellar cortex circuitry and Purkinje cells play a critical role as the sole outputs from the cerebellar cortex to the deep cerebellar nuclei and vestibular nuclei. While parallel fiber inputs from mossy fibers via granule cells drive Purkinje cell simple spike (SS) activities, powerful climbing fiber inputs drive another unique spike activity patterns called complex spike (CS), which are infrequent and have highly variable spikelet waveforms. This variability, coupled with electrode drift in long-term recordings, makes automated CS detection difficult. The lack of trustworthy and automatic high-throughput complex spike sorters severely hinders our understanding of the Purkinje cell complex spike activities, especially with large data scale from high-density multichannel electrodes. To address the challenge of CS sorting and the limitations in current spike sorters, we present a deep learning-based approach for CS sorting in high-density multi-channel electrode recording data. Our model uses a U-Net architecture as an information bottleneck to condense and extract CS waveform features, augmented with hybrid inception blocks using both self-attention mechanisms and multi-level convolution kernels to integrate both long-term and short-term local field potential and action potential signals from the multi-channel recordings for CS detection. The algorithm outputs CS event probabilities in a sequence-to-sequence manner; probabilistic detections are clustered to find each CS clusters and then paired with concurrently detected SS clusters to reconstruct the full Purkinje cell firing activity. We benchmarked our method on rhesus macaque cerebellar vermis recording datasets and found our method achieves human expert consensus-level performance (leave-one-out cross validation F1 score = 0.82 ± 0.03 , mean $\pm$ standard error) and significantly outperforms other existing CS sorting methods

and state-of-the-art SS sorting algorithms. CS detection results from our model are robust to CS spikelet waveform variability and electrode drift, ensuring consistent sorting across long-term and high-throughput recordings. Overall, we conclude that our deep learning-based CS sorting method facilitates our further understanding of Purkinje cell plasticity, internal model updating, and cerebellar-dependent learning.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

Support: NIH R37-NS128416

Title: Population coding in the fastigial nucleus of the cerebellum

Authors: *A. SHOUP, P. HAGE, M. FAKHARIAN, R. SHADMEHR;
Biomed. Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: The cerebellum plays a critical role in motor control. During saccadic eye movements, primates can stop their eyes precisely on a target due to neuronal activity in the caudal fastigial nucleus (cFN). Behavioral deficits following cerebellar damage suggest that cFN neuron firing rates should encode saccade parameters. Yet, activity of individual cFN neurons often persists beyond the end of movement, raising the question: how does the cerebellum control saccades if individual neuron activity appears poorly related to movement parameters?

A similar puzzle exists in the cerebellar cortex. Purkinje cells (P-cells) in the oculomotor vermis display simple spike patterns that show weak relationships to saccade direction or velocity, and their modulation also outlasts the movement. However, we recently showed that each P-cell can be assigned a "potent vector", a weight vector representing its net behavioral effect, defined by climbing fiber input. These potent vectors revealed that population activity lies in structured "null spaces," where certain spikes cancel out to prevent unintended effects on movement. Here, we extend this approach to the fastigial nucleus. Using high-density silicon probes, we recorded 677 well-isolated neurons from the cFN of head-fixed common marmosets performing a visually guided saccade task, including sessions with over 49 simultaneously recorded neurons. Based on waveform shape and timing, we identified two putative classes of outward-projecting neurons ("large" and "wide," n=175, 164), named for their soma size and action potential duration. These neurons exhibited strong within-class synchrony but weak synchrony with other cell types.

To assign a potent vector to each neuron, we searched for a movement direction that maximized directional symmetry in firing rates—an approach inspired by the symmetry seen in P-cell activity. This allowed us to hypothesize that fastigial potent vectors may be inherited from P-

cells.

We then grouped these neurons into populations. “Large” cells fired early, pushing the eyes in the direction of movement; “wide” cells fired later as the saccade decelerated. Firing rates exhibited cancellation orthogonal to movement direction, demonstrating push-pull dynamics consistent with known fastigial projections to brainstem oculomotor centers.

In sum, we classified fastigial neurons by size and synchrony, assigned potent vectors, and uncovered a population code that mirrors dynamics of saccade acceleration and deceleration. These findings suggest that fastigial neuron output can be understood via potent vectors inherited from cerebellar cortical inputs.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

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Title: Organization of extra-cerebellar inhibitory inputs to the inferior olive

Authors: *B. ZINGG¹, H. DONG²;

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Abstract: The inferior olive plays an important role in refining motor movements via its climbing fiber projections to the cerebellar cortex. Signaling through this pathway is shaped by inhibitory feedback that is thought to primarily originate from GABAergic neurons in the cerebellar nuclei. However, feedback from the fastigial cerebellar nucleus to the caudal medial accessory olive (cMAO) appears to be largely absent, suggesting other brain regions may account for the inhibitory input to cMAO. Previous studies have identified several brainstem sources of GABAergic inhibition to specific subnuclei of the cMAO, though a complete description of all inputs is still lacking. Here we use monosynaptic rabies tracing in transgenic mice to first report all presynaptic input to the olive from across the entire brain and spinal cord. We then examined labeled neurons in all regions for the co-expression of VGAT, a marker for inhibitory neurons, to identify novel sources of olivary inhibition. Besides the well characterized GABAergic projection from cerebellar nuclei, we identified numerous sources of long-range inhibition to the olive from mid- and hindbrain structures, many of which, to our knowledge, have yet to be described. We then cross-examined these input sources using dual-color viral co-injections in Vglut2-Cre x VGAT-Flp mice to reveal excitatory and inhibitory axon targeting to specific olivary subnuclei. Each inhibitory projection targeted a specific subnucleus and collectively they innervated the entire cMAO. In addition, we report specific lateralization differences for each inhibitory source and provide evidence for the direct targeting of many of

these inhibitory cell populations by excitatory fastigial nucleus output, suggesting a potential feedback role within the context of olivocerebellar loops. Together these data close a gap in our understanding of the distribution of inputs to the inferior olive, and define all potential sources of inhibition that originate from outside the cerebellar nuclei.

Disclosures: **B. Zingg:** None. **H. Dong:** None.

Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

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Topic: F.02. Cerebellum

Support: NINDS R01NS122741

Title: Cerebellar transcranial AC stimulation produces frequency-dependent modulation of the cerebello-thalamic pathway.

Authors: ***D. MOURRA**¹, M. SAHIN², E. J. LANG¹;

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Abstract: Electrical neuromodulation techniques are increasingly being used to treat a range of neurological and psychiatric disorders. In particular, cerebellar transcranial alternating current stimulation (ctACS) is an attractive technique because it is non-invasive, low-cost, and given the cerebellum's widespread projections to the forebrain, it has the potential to treat a wide range of disorders. Indeed, several lines of evidence suggest that ctACS not only can improve motor learning but can also affect cognitive and emotional processes in normal and brain-injured individuals. However, ctACS's full potential has not been realized in part because we have little knowledge of how it alters cerebellar output. Previously, we found evidence that ctACS modulates Purkinje cell (PC) activity, which then drives activity of cells in the cerebellar nuclei (CN), the main output station of the cerebellum. However, the CN contains multiple classes of cells with distinct properties that give rise to the various cerebellar efferent pathways. This suggests that different cerebellar output pathways may have distinct response profiles to ctACS. Here, we characterize the response of CN-thalamic (CN-Th) neurons, the major origin of cerebellar pathways to the forebrain. Single-unit recordings of CN-Th cells were obtained in anesthetized adult Sprague-Dawley rats. Antidromic responses to stimulation of the contralateral ascending superior cerebellar peduncle fibers at the mesodiencephalic junction were used to identify CN-Th cells. The ctACS electrode was positioned on the skull above the ipsilateral crus 1, medial to the recording site. The return electrode was placed under the skin of the shoulder ipsilateral to the recorded cell. ctACS was found to robustly modulate CN-Th activity at either the stimulus frequency or double that frequency, resulting in uni- or bi-modal response patterns in the spike histograms, respectively. To quantify and classify these patterns, we applied model-

based clustering for circular data to fit a mixture of two von Mises populations to the bimodal distribution and to separate them into two unimodal responses that could be analyzed individually. The strength of CN-Th modulation increased, and the bimodal response pattern became predominant, as the stimulation frequency was increased from 0.5 Hz to 80 Hz. Our results show that ctACS can strongly modulate the first stage in the cerebello-thalamo-cortical pathway, which has potential implications for its use in treating motor and cognitive disorders.

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Poster

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Topic: F.02. Cerebellum

Support: NIH NIDCD R00 DC016905
National Ataxia Foundation
Hearing Health Foundation

Title: The role of NMDA receptors in developing cerebellar unipolar brush cells

Authors: ***H. N. HARIANI**, T. S. BALMER;
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Abstract: Unipolar brush cells (UBCs) are excitatory interneurons that have a characteristic single dendritic brush that amplifies and extends incoming sensory signals in the cerebellum. UBCs transform synaptic input through their ionotropic and metabotropic glutamate receptors. Differential regulation of receptors is a critical cell-type specific developmental process. For example, another excitatory cerebellar interneuron, the granule cell, expresses the NMDA receptor NR2B/A subunits in early postnatal life that are replaced by NR2C subunits in adulthood. NR2C subunits are less sensitive to Mg^{2+} block and have different kinetic properties, resulting in markedly different granule cell synaptic responses across development, which is necessary for normal development and function. How the expression of glutamatergic receptor types change in UBCs as they develop is not understood. Here we examine the expression of NMDA receptor subunits during maturation and whether signaling through these receptors is necessary for normal structure and function of UBCs. We conducted whole-cell patch clamp electrophysiology recordings from UBCs in acute brain slices to characterize NMDA receptor-mediated currents and consequent firing patterns in young (P10-14) and electrophysiologically mature (P26-32) mice. We also used fluorescence in situ hybridization (RNAscope) to confirm the presence of NR2C/D RNA in young UBCs. Our results show the presence of NMDA receptor-mediated currents at hyperpolarized potentials (-75 mV) in young, but not mature, UBCs, which are abolished by NR2C/D specific antagonist (PPDA), suggesting that young UBCs express these subunits, which are lost later in development. RNAscope corroborated our

electrophysiology data and also revealed the differential developmental regulation of NR2C and D subunits which could not be observed using electrophysiology owing to the lack of specific pharmacological agents capable of differentiating between the two. We show that NR2D mRNA expression is highest at P7, and that for NR2C subunit peaks at P12, while the expression of mRNA for both subunits is significantly lower at P30. Despite considerable variation in firing patterns following 50 Hz 200 ms synaptic stimulation we consistently observed a paradoxical increase in excitability after blocking NMDA receptors, including decreased latency of the first spike, increased number of spikes, and increased burst duration. Further experiments will investigate the role of NMDA receptors in UBC migration and dendritic brush development. This study will provide novel insights into the role of NMDA receptors in UBC development and cerebellar circuit processing.

Disclosures: H.N. Hariani: None. T.S. Balmer: None.

Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.17/S9

Topic: F.02. Cerebellum

Support: Chan Zuckerberg Initiative
Whitehall Foundation
Research Corporation for Science Advancement

Title: Cerebellar Mechanisms of Adaptive Motor Timing in a Flexible Eyelid Conditioning Task

Authors: S. MALEKPOUR¹, Z. SALAMTABRIZI¹, O. AMIRATASHANI¹, H. NGUYEN², M. CHEN¹, T. STAMM¹, S. BITA¹, M. BIBIDAKIS³, S. KUMAR¹, S. MEDICO¹, S. AMINNAJI¹, *F. NAJAFI¹;

¹Sch. of Biol. Sci., ²Wallace H. Coulter Dept. of Biomed. Engin., ³Sch. of Electrical and Computer Engin., Georgia Inst. of Technol., Atlanta, GA

Abstract: Accurate timing of motor responses is critical for adaptive behavior. The cerebellum is thought to play a key role in predicting and adjusting the timing of movements, especially when task demands change. However, how cerebellar circuits support rapid adaptation of motor timing remains poorly understood. To address this, we developed a variant of eyelid conditioning in head-fixed mice, in which animals must learn to time their blinks to match the expected onset of an air puff following a visual cue. Each session includes two types of blocks with different inter-stimulus intervals, requiring mice to flexibly adjust the timing of their conditioned responses. This design allows us to probe temporal expectation, prediction error, and adaptive motor learning within a controlled behavioral framework. We used two-photon calcium imaging to record Purkinje cell dendritic activity in the cerebellar cortex of mice during learning. Trial-by-trial analyses revealed that specific populations of neurons show activity patterns aligned with

sensory events and prediction errors. Ongoing work investigates how Purkinje cells update their representations across distinct blocks of trials, and how sleep deprivation affects this adaptive process. Our findings will illuminate how cerebellar circuits form and update internal timing models necessary for flexible motor behavior.

Disclosures: **S. Malekpour:** None. **Z. Salamtabrizi:** None. **O. Amiratashani:** None. **H. Nguyen:** None. **M. Chen:** None. **T. Stamm:** None. **S. Bitá:** None. **M. Bibidakis:** None. **S. Kumar:** None. **S. Medico:** None. **S. Aminnaji:** None. **F. Najafi:** None.

Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.18/S10

Topic: F.02. Cerebellum

Support: KAKENHI 22H03456
KAKENHI 23K24714

Title: Disruption of cerebellar circuit development occurs due to impaired microglia-mediated synaptic pruning in diffuse White Matter Injury

Authors: **S. TOMINAGA**, C.-G. JUNG, M. SHINDO, N. TAJIRI, M. SUZUKI, *H. HIDA;
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Abstract: While the majority of research has focused on the cerebrum in the context of diffuse white matter injury (DWMI), the cerebellum, which is integral to motor coordination, also warrants scholarly investigation. In normal rats, immature climbing fibers (CFs) initially form multiple synapses with Purkinje cells (PCs). These synapses undergo pruning in two distinct phases, facilitated by microglia: an early phase around postnatal day 10 (P10) and a late phase around P15, ultimately resulting in a single CF-PC synapse by P28. However, the specific mechanism through which DWMI affects the formation of cerebellar circuits has yet to be clarified. To investigate this, diffuse white matter injury (DWMI) was induced in postnatal day 3 (P3) male Wistar rats by occluding the right common carotid artery, followed by exposure to 6% hypoxia for one hour, resulting in dysfunction on the left side. Immunohistochemical analyses were performed on Calbindin D-28k, a marker for Purkinje cells, and vGluT2, a marker for climbing fibers, within the hindlimb-associated cerebellar cortex at developmental stages P10, P15, and P28. Western blot analysis was conducted at P28, and microglial morphology was assessed through Iba1 staining. Our findings reveal no significant hemispheric differences in CF-PC synapses at P10 and P15. However, the left (affected) cerebellum demonstrated a reduction in CF translocation, along with an increase in synaptic density at P28. Notably, there was no correlation between the number of CF synapses and the number of Purkinje cells. Moreover, at P10, a reduction in the number of microglia was observed on the left side. By P15, there was a marked decrease in amoeboid microglia, suggesting a decline in pruning activity. The findings

suggest that inadequate microglia-mediated synaptic pruning during critical developmental periods may explain the atypical cerebellar morphology observed in DWMI. This disruption in circuit refinement could potentially contribute to the motor impairments associated with neonatal hypoxic-ischemic injury. However, further research is necessary to elucidate the role of microglial cytokines in this process.

Disclosures: **S. Tominaga:** None. **C. Jung:** None. **M. Shindo:** None. **N. Tajiri:** None. **M. Suzuki:** None. **H. Hida:** None.

Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.19/S11

Topic: H.05. Mood Disorders

Title: Eeg-based neurodynamic model for drug effect prediction in individuals

Authors: *Y. YAN;
Gnosis Neurodynamics Co., Beijing, China

Abstract: Large-scale modeling techniques have been broadly used in pretraining neuroelectrophysiological or neuroimaging data. However, traditional pre-trained models are primarily suited for descriptive tasks. Structural and parametric lack of physiological interpretability makes these models difficult to reveal pathological mechanisms and predict individual intervention effects. We designed a neural dynamics model based on EEG observations. By integrating local neural dynamics modeling and whole-brain functional connectivity, we formulated the local neural activities into interpretable biophysical differential equations, while leveraging functional connectivity maps to model interactions across brain regions. The primary goal of this modeling approach is to decode EEG signals into clinically actionable brain function parameters with real biophysical meanings. In model validation, we applied this framework to EEG data from 706 clinically diagnosed depression patients. By computing dynamic parameters of brain functional activity for each patient and comparing them against healthy controls, we identified individualized abnormal dynamic signatures. Further, we simulated the effects of three antidepressant classes (SSRIs, SNRIs, NMDA receptor antagonists) by perturbing these parameters according to their known pharmacological mechanisms. Predictions of treatment outcomes were derived from the resultant changes in dynamic parameters. Our results demonstrate that the neural dynamics model, when combined with drug mechanism knowledge, can effectively predict individualized treatment responses in depression. This approach may hold significant promise for precision medicine in psychiatric disorders with heterogeneous etiologies and subtypes.

Disclosures: **Y. Yan:** A. Employment/Salary (full or part-time): Gnosis Neurodynamics Co..

Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR283.20/S12

Topic: F.02. Cerebellum

Support: NIH R37-NS128416

Title: Millisecond precision of spike timing in cerebellar Purkinje cells and interneurons

Authors: ***M. FAKHARIAN**¹, E. TAECKENS¹, A. SHOUP², A. VASSERMAN¹, H. Y. ELSEWEIFI⁴, P. HAGE⁵, R. SHADMEHR³;

²Biomed. Engin., ³Dept Biomed. Eng, ¹Johns Hopkins Univ., Baltimore, MD; ⁴Johns Hopkins Univ., ⁵Biomed. Engin., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: In the cerebellum, neurons influence each other's spike timing with sub-millisecond precision. Purkinje cells (P-cells) exhibit oscillatory spiking with rate-dependent inter-spike intervals (ISI), while molecular layer interneurons (MLIs), particularly type 1s, display bursty, irregular spiking at similar rates. What determines whether a neuron spikes rhythmically or stochastically, intrinsic dynamics or network architecture, and what is the functional relevance? To explore this, we recorded high-density electrophysiological data from two marmosets performing a saccade task. Cell types were identified using layer localization via complex spike waveform changes across P-cell dendritic and axonal compartments, and sub-millisecond spike interactions. We asked whether MLI-P-cell spike interactions are static or dynamically modulated during behavior.

Sub-millisecond interactions, mediated by gap junctions and ephaptic coupling, challenge synchrony metrics due to behaviorally driven rate fluctuations. We used interval jittering to generate surrogate spike trains and assessed synchrony at 1 ms resolution. In both MLIs and P-cells, joint spiking rate scaled linearly with the jittered rate with a slope > 1. That is, as firing rates increased, the number of synchronous spikes was always greater than chance, growing multiplicatively as a function of rates. Despite large behavioral rate changes, synchrony between two neurons remained a constant multiple of chance, not a constant offset.

What is the purpose of this synchrony? We examined MLI-P-cell interactions. P-cell spiking was suppressed via two inhibitory mechanisms: a rapid initial suppression at ~0.4 ms latency, likely due to ephaptic coupling via the pinceau, then a slower suppression at ~1.3 ms, likely due to GABA. Synchronous MLI spikes produced additive P-cell suppression, consistent with a superposition model. This implies that P-cells integrate MLI input over a very short window: multiple MLIs must fire within 2 milliseconds to suppress spiking beyond a single cell's refractory period.

A conductance-based leaky integrate-and-fire (LIF) model with gap junction input reproduced MLI synchrony and predicted that oscillatory P-cell output requires correlated excitatory (parallel fiber) and inhibitory (MLI) input, consistent with feedforward inhibition. Conditional probability analyses further indicated shared input to MLIs and P-cells.

These findings show that sub-millisecond spike timing coordination, shaped by network structure and cell-type specific connectivity, supports cerebellar computation beyond rate coding.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

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Program #/Poster #: PSTR283.21/T1

Topic: F.02. Cerebellum

Support: NSTC 112-2628-B-002-011
NSTC 113-2628-B-002-002

Title: Molecular Layer Interneurons Encode Velocity Sensing in the Cerebellum with Subsecond Precision

Authors: *J.-Y. HO¹, M.-K. PAN²;

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Abstract: Sensory feedback is critical for sensorimotor coordination, a key function of the cerebellum. However, the cerebellar coding mechanisms of sensory feedback remain unclear. In this study, we demonstrate that molecular layer interneurons (MLIs) in the cerebellum encode the sensory feedback of moving velocities with remarkable precision. Utilizing two-photon calcium imaging in awake, behaving mice within a virtual reality (VR) system on a rotary platform, we observed that the intensity of cerebellar MLI activity is reported through populational coding. These populational MLIs accurately encode moving velocity with 99.9% precision over 90 seconds of walking, maintaining a temporal resolution of 30 frames per second in each trial for every tested mouse, despite the trial-by-trial variations of self-paced walking. This precision is achieved using a simple random forest regression that accounts for the biological domain knowledge of synaptic weights and threshold effects, without requiring optimization through machine learning. Cross-correlogram analysis confirmed that the velocity codes of MLIs exhibited approximately a 300 ms lag relative to the motion, indicating a sensory feedback rather than a motor output mechanism. Experiments involving VR-motion dissociation and dark-field conditions further revealed that the velocity codes are independent of visual inputs and are instead encoded through the secondary integration of somatosensory information. Moreover, silencing the GABAergic synaptic outputs of MLIs using bicuculline led to a significant reduction in the accuracy of velocity feedback, which in turn elicited compensatory increases in moving velocity in mice. Our findings provide compelling evidence of a cerebellar mechanism for sensorimotor coordination with exceptional mathematical precision by encoding moving velocity.

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Poster

PSTR283: Cell Types and Circuit Physiology in the Cerebellum

Location: SDCC Halls B-H

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Program #/Poster #: PSTR283.22/T2

Topic: F.02. Cerebellum

Support: Seed fund at Mount Sinai #02482323
Barrow Neurological Foundation #3033227

Title: A Novel Early Onset Spinocerebellar Ataxia 13 BAC Mouse Model with Cerebellar atrophy, Tremor, and Ataxic Gait

Authors: *J. YIN¹, J. NICK², S. KHARE², H. KLOEFKORN⁵, M. GAO⁶, M. WU⁷, J. WHITE⁷, J. RESNICK³, K. ALLEN², H. NICK⁴, M. WATERS¹;

¹Icahn Sch. of Med. at Mount Sinai, New York, NY; ³Dept. of Mol. Genet. and Microbiology,,

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Engin., Oregon State Univ., Corvallis, OR; ⁶Dept. of Neurobio., Barrow Neurolog. Institute, St. Joseph's Hosp. and Med. Ctr., Phoenix, AZ; ⁷Barrow Neurolog. Inst., Phoenix, AZ

Abstract: Background: Spinocerebellar ataxia 13 (SCA13) is an autosomal dominant neurological disorder caused by mutations in *KCNC3*. Our previous studies revealed that *KCNC3* (Potassium Voltage-Gated Channel Subfamily C Member 3) mutation R423H results in an early-onset form of SCA13. Previous biological models of SCA13 include zebrafish and *Drosophila* but no mammalian systems. More recently, mouse models with *Kcnc3* mutations presented behavioral abnormalities but without obvious pathological changes in the cerebellum, a hallmark of patients with SCA13. **Method:** Here, we present a novel transgenic mouse model by bacterial artificial chromosome (BAC) recombineering to express the full-length mouse *Kcnc3* expressing the R424H mutation. **Results:** This BAC-R424H mice exhibited behavioral and pathological changes mimicking the clinical phenotype of the disease. The BAC-R424H mice (homologous to R423H in human) developed early onset clinical symptoms with aberrant gait, tremor, and cerebellar atrophy. Histopathological analysis of the cerebellum in BAC-R424H mice showed progressive Purkinje cell loss and thinning of the molecular cell layer. Additionally, Purkinje cells of BAC-R424H mice showed significantly lower spontaneous firing frequency with a corresponding increase in inter-spike interval compared to that of wild-type mice. **Conclusion:** Our SCA13 transgenic mice recapitulate both neuropathological and behavioral changes manifested in human SCA13 R423H patients and provide an advantageous approach to understanding the role of voltage-gated potassium channel in cerebellar morphogenesis and function. This mammalian *in vivo* model will lead to further understanding of the R423H allelic form of SCA13 from the molecular to the behavioral level and serve as a platform for testing potential therapeutic compounds.

Disclosures: J. Yin: None. J. Nick: None. S. Khare: None. H. Kloefkorn: None. M. Gao: None. M. Wu: None. J. White: None. J. Resnick: None. K. Allen: None. H. Nick: None. M. Waters: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR284.01/T3

Topic: F.04. Voluntary Movements

Support: Department of Science and Technology, Government of India

Title: Reactivation strengthens human motor memories against interference from competing inputs

Authors: *P. K. MUTHA¹, T. SURESH², A. KUMAR³;

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Abstract: Memory consolidation theory states that memories are labile upon acquisition but stabilize and become resistant to interference from competing inputs after a period of consolidation. However, the reconsolidation theory posits that “reactivation”, or active retrieval, of a consolidated memory destabilizes it and makes it susceptible to degradation from another memory acquired by learning a new task. We investigated the fate of a consolidated motor memory when it is reactivated and challenged with a competing memory. Based on the reconsolidation hypothesis, we predicted that performance on an initially consolidated task would be much more impaired when a competing task is learned following its reactivation, compared to when it is not reactivated. In our first experiment, we used a 3-day A-B-A visuomotor adaptation paradigm, wherein following baseline practice, subjects learned a 30° counter-clockwise rotation A on day 1 followed by a competing 30° clockwise rotation B on day 2, and were retested on A on day 3. Subjects were divided into two groups, depending on whether A was reactivated on day 2 just before B learning (A-AB-A) or not (A-B-A). A third, control group (A-A) did not adapt to B. Contrary to our prediction, we did not observe any worsening in A performance upon re-exposure on day 3 in the A-AB-A group. Specifically, relearning of A was no different from naïve A learning in either group, suggesting that while interference was clearly present, it was, in all likelihood, independent of reactivation. This led us to consider an alternative view that reactivation might *protect* against interference rather than destabilize the memory trace. In our second and third experiments, we modified our design to probe a putative benefit from active retrieval. We introduced no rotation (N) trials prior to A relearning on day 3 (A-AB-NA and A-B-NA groups), or immediately after B learning on day 2 (A-ABN-A and A-BN-A groups). These null trials served to wash out potential anterograde effects of B learning. Here, we found a retrieval-mediated benefit, but only when learning from the interfering task was washed out immediately thereby preventing its consolidation. Collectively, our results show that reactivation of a consolidated motor memory does not necessarily destabilize it in a way that makes it more vulnerable to interference, as predicted by the reconsolidation model. Instead, reactivation may serve a protective function, shielding the

original memory from degradation when challenged by competing learning. Our results hold implications for strategizing protocols aimed at improving motor memory retention in neurorehabilitation and sports.

Disclosures: P.K. Mutha: None. T. Suresh: None. A. Kumar: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

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Program #/Poster #: PSTR284.02/T4

Topic: F.04. Voluntary Movements

Title: Jumping the cursor, not the target: causal inference affects implicit motor learning

Authors: *Z. NIE¹, H. WANG¹, T. ZHANG¹, E. COHEN², K. WEI¹;

¹Sch. of Psychological and Cognitive Sci., Peking Univ., Beijing, China; ²Univ. of Florence, Florence, Italy

Abstract: Motor learning requires continuous corrections of motor errors, whose inferred causality affect how errors are used. Previous studies have shown that errors attributed to sensorimotor imperfections are more likely to be corrected, whereas those attributed to external environmental changes are more likely to be exempted from being used for sensorimotor recalibration. Hence, most adaptation paradigms perturb the effector (e.g., via a force field) or the representation of the effector (e.g., via a visuomotor rotation) to elicit sensorimotor adaptation. A notable exception is the target jump paradigm, where the target is abruptly displaced during movement to create a target error and to induce subsequent learning. Recent studies have found that target jump fails to induce implicit adaptation and attributed this null effect to the ineligibility of target error as the error signal for implicit learning. We hypothesize that the jumped target is inferred as an environmental change and thus not used for learning. To test our hypothesis, we manipulated the hand cursor, as opposed to the target, during a reaching task. Under identical reaching contexts (e.g., even with the same target error), cursor jump, but not target jump, robustly elicits implicit adaptation. We further show that implicit adaptation remains even if the hand cursor jumps onto the target to nullify target error, as long as the hand cursor deviates from the intended movement direction. Cursor jump, but not target jump, leads to proprioceptive changes, as measured by localization tests of a passively placed hand. The distinct learning patterns from target jump and cursor jump can be well predicted by a state-space model which integrates with the causal inference idea. Our findings support our recent theorization that implicit adaptation is driven by perceptual prediction error, i.e., the error between intended and perceived movement. Furthermore, combining with previous findings on target jump in saccadic and reaching adaptation, our findings suggest that causal attribution of movement feedback to environmental or sensorimotor changes determines implicit motor learning.

Disclosures: Z. Nie: None. H. Wang: None. T. Zhang: None. E. Cohen: None. K. Wei: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

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Program #/Poster #: PSTR284.03/T5

Topic: F.04. Voluntary Movements

Support: NIH NS112367

Title: Extrinsic and intrinsic intermanual transfer in a complex zigzag-maze task via reinforcement and error-based learning

Authors: *M. WANG, H. J. BLOCK;
Dept. of Kinesiology, Indiana Univ. Bloomington, Bloomington, IN

Abstract: Intermanual transfer—where motor learning acquired with one hand transfers to the other—has been well characterized in simple tasks but remains poorly understood for complex skills. Transfer may be extrinsic, intrinsic, or both. Extrinsic transfer encodes visual information about movement and target in a world-centered reference frame, whereas intrinsic transfer relies on a body-centered reference frame to compute kinesthesia and intersegmental relationships. Understanding intermanual transfer could have important clinical implications: in stroke rehabilitation, maximizing transfer from the unaffected limb to the affected limb could accelerate functional gains. Here, we addressed the underlying basic science question in healthy young adults, examining both extrinsic and intrinsic transfer in a complex zigzag-maze tracing task. We tested two learning processes in separate groups: error-based and reinforcement learning. Error-based learning involves progressive corrections based on error signals induced by the sensory discrepancy between the expected and actual movement. In contrast, reinforcement learning entails repeating past actions that led to success and avoiding those that led to failure, requiring binary success/failure feedback. Twenty right-hand-dominant participants trained on the maze-tracing task with their right hand, receiving error-based feedback ($n = 10$, 26.2 ± 3.8 yr) or reinforcement feedback only ($n = 10$, 24.6 ± 4.3 yr). All participants completed pre- and post-tests across six movement time (MT) bins, using the right hand (RH) on the standard maze to assess acquisition, the left hand (LH) on the standard maze to assess extrinsic intermanual transfer, and the LH on the mirror-reversed maze to probe intrinsic intermanual transfer. The error-based group received continuous cursor feedback and earned graded accuracy points; the reinforcement group received only binary (0/1) feedback based on percentage-correct thresholds. We evaluated speed-accuracy and speed-variability trade-offs by calculating post-pre differences at each MT. Preliminary results suggest improved movement variability in both extrinsic (LH standard) and intrinsic (LH mirror) transfer conditions following training in both groups. No learning effects were observed for the trained right hand, which could indicate a performance ceiling—participants had already reached near-maximal accuracy in the pre-test. It is possible that differences in intermanual transfer are too subtle to observe in this preliminary sample. It is also possible that both training methods engage widespread interhemispheric pathways, both intrinsic and extrinsic.

Disclosures: M. Wang: None. H.J. Block: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR284.04/T6

Topic: F.04. Voluntary Movements

Title: Aging enhances implicit visuomotor adaptation in a manner related to increased sensitivity to task success and independent from cognitive impairment

Authors: *O. A. KIM¹, Y. ZHOU²;

¹Neurosci., ²Psychology, Bates Col., Lewiston, ME

Abstract: Older adults (59+ years old) exhibit enhanced cerebellar implicit visuomotor reach adaptation, despite showing impaired learning in classical versions of the task (Vandevorode & De Xivry, 2019; Neurobiol Aging). During this task, the experimenter disrupts the relationship between visual feedback and hand position to generate an error signal that drives compensatory learning. Although there are age-related declines in proprioception that may affect older adults' ability to detect the visual-proprioceptive mismatch, these sensory changes do not appear to be responsible for the enhancement of implicit adaptation (Vandevorode & DeXvry, 2021; J Neurophys). Because aging is accompanied by cognitive changes and changes in reward processing, we investigated whether cognitive impairment or alterations in sensitivity to task failure related to age-related enhancements in implicit reach adaptation (Harada et al., 2014; Clin Geriatr Med; Dreher et al., 2008, PNAS). To investigate these possibilities, we assessed healthy older and younger adults' implicit reach adaptation and their responses to perceived task success by shifting the location of the target relative to the visual feedback (Moorehead et al., 2017, J Cog Neuro; Al-Fawakhiri et al., 2023, J Neurophys). Participants also completed the Self-Administered Gerocognitive Examination (SAGE) to evaluate aging-related cognitive decline. Contrary to our initial hypothesis, the extent of enhanced implicit adaptation among older adults was not correlated with scores on the SAGE cognitive assessment. However, we found that, compared to younger adults, older adults exhibited greater suppression of reach adaptation when the target was shifted to the location of visual feedback. Interestingly, the magnitude of this suppression was inversely related to the extent of their enhanced implicit adaptation for specific individuals. These findings suggest that the processes governing the suppression of adaptation, potentially involving altered sensitivity to task success or changes in visual error processing when the target and cursor are co-localized, may underpin the age-related enhancement in implicit visuomotor adaptation.

Disclosures: O.A. Kim: None. Y. Zhou: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR284.05/T7

Topic: F.04. Voluntary Movements

Support: NIH Grant R25NS114097

Title: Effects of Solution Redundancy in a VR Reaching Control Task

Authors: ***B. BORCHELT**¹, V. PIERCE², L. R. SCHOLL², A. L. ORSBORN²;

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Abstract: Human-machine interfaces (HMIs) are often redundant, meaning they have more dimensions of input than dimensions of feedback. This mismatch creates a credit assignment problem that complicates the user's ability to discern which aspects of their motor commands contribute to successful task outcomes. HMIs can be argued to have two types of redundancy: goal redundancy (multiple inputs achieve the same endpoint) and sensory redundancy (multiple inputs yield identical sensory feedback). Sensorimotor error and reward feedback are known to drive learning in different ways, which could mean that these types of redundancy may also have dissociable effects on learning. To investigate the specific contribution of sensory redundancy, we designed a novel redundant HMI using a virtual reality (VR) visuomotor perturbation task. Participants control a cursor using their hand movements in 3D space to perform a 2D point-to-point reaching task. One dimension of this task is defined as 'goal-irrelevant,' such that reaching any endpoint along this dimension, above or below the target on the goal-relevant plane, resulted in task success. We manipulated sensory feedback between groups to isolate the effect of sensory redundancy. In a pilot experiment with six participants assigned to two groups, one group received full 3D visual feedback of their cursor position, eliminating sensory redundancy, while the other group's cursor feedback was confined exclusively to the goal-relevant plane, preserving sensory redundancy. In both groups, goal redundancy is still present. Although significant differences in learning were not observed in the small pilot cohort, there was some indication that sensory redundancy may lead to more variation in the solution space, which will be investigated in a continuation of the study with more participants. Understanding the specific effects of sensory redundancy will guide engineers on what type of additional feedback about task performance — beyond what is minimally required to achieve the goal — is beneficial or detrimental for user learning. These insights are becoming increasingly more important as HMIs expand into applications requiring robust skill acquisition, such as rehabilitation robotics and immersive virtual environments.

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Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR284.06/T8

Topic: F.04. Voluntary Movements

Support: De Luca Foundation Seed for Science Award 2024

Title: A home-based VR-EMG platform for rehabilitation of finger-individuation in stroke survivors

Authors: ***A. B. SOLANO**¹, L. ALEJANDRO², L. NEIRA², P. MARTINEZ VIADEMONTE¹, D. BLUSTEIN³, V. DELLA-MAGGIORE¹;

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Abstract: Fine motor skill impairment, particularly in finger individuation, remains a significant neurorehabilitation challenge post-stroke. Conventional approaches often fail to address maladaptive compensatory strategies, such as co-activation of adjacent fingers or wrist deviations, which obstruct true recovery of motor function. In recent years, motor re-learning approaches grounded in use-dependent neuroplasticity have shown promise in improving finger individuation through focused, repetitive practice, even long after injury (Mawase et al., 2020). However, this approach typically requires close in-clinic supervision and the use of mechanical constraints to prevent compensatory strategies, which limits its accessibility for underserved populations. To address this, we implemented a home-based Virtual Reality (VR) training system integrated with wireless surface electromyography (sEMG) and machine learning (ML) algorithms for advanced, personalized finger motion analysis. The system features an engaging VR game on a low-cost headset, where users steer a spaceship with discrete thumb-to-finger oppositions to avoid asteroids. Critically, sEMG data from forearm muscles is processed by ML algorithms, which can be personalized to individual patient motor profiles and evolving recovery stages. This allows for highly refined detection of true, subtle finger activations and differentiation from compensatory muscular synergies—details often imperceptible to visual tracking alone. Such an approach aims to ensure game control is driven by desired individuated movements, while simultaneously offering insights into underlying muscle activity patterns. Preliminary results in healthy participants demonstrate the system's feasibility and the algorithm's efficacy. The ML model effectively discriminated targeted gestures from compensatory movements. Usability assessments confirmed the system's high feasibility, while participants demonstrated consistent improvements in gesture execution speed and precision. A pilot study with chronic (>6 months) stroke survivors is currently underway. This sEMG-VR-ML system offers a pathway to more precise and adaptive home-based neurorehabilitation. If successful, this approach will underscore a unique opportunity to further enhance stroke rehabilitation by combining compensation-sensitive, home-based finger training with the beneficial effects of sleep timing (Solano et al., 2022; 2024), aligning motor re-learning with biologically sensitive windows of plasticity.

Disclosures: **A.B. Solano:** None. **L. Alejandro:** None. **L. Neira:** None. **P. Martinez Viademonte:** None. **D. Blustein:** None. **V. Della-Maggiore:** None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR284.07/T9

Topic: F.04. Voluntary Movements

Support: De Lucas Foundation Seed for science award 2024

Title: A VR-based immersive rehabilitation paradigm for finger individuation at home

Authors: ***P. MARTINEZ VIADEMONTE**¹, A. B. SOLANO¹, D. BLUSTEIN², V. M. DELLA-MAGGIORE¹;

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Abstract: Stroke-related impairments in fine motor skills are a major challenge in neurorehabilitation. Current clinical interventions are resource-intensive, often unaffordable and inaccessible for many patients. Furthermore, conventional methods often fail to correct maladaptive compensatory movements, hindering true recovery. While lab-based devices, such as piano-like finger trainers, have demonstrated effectiveness in promoting finger individuation (Mawase et al., 2020), they often lack engagement and necessitate frequent clinic visits. Emerging home-based VR systems using data gloves offer better engagement but can suffer from imprecision, unreliable detection of small movements, and interference with tactile feedback. To overcome these limitations, we developed a VR training system for accessible home-based finger individuation therapy. Our solution utilizes a low-cost, standalone VR headset, with hand and finger movements tracked directly by its integrated cameras. The immersive Unity-based game requires users to steer a spacecraft, avoiding asteroids, using discrete thumb-to-finger opposition gestures. A custom computer vision algorithm analyzes hand movements to specifically detect these target gestures while actively ignoring common compensatory strategies, such as wrist deviation or co-activation of non-target fingers. This ensures that only precise, isolated movements result in successful game actions. We evaluated the system's feasibility, usability, and adherence in healthy participants. The algorithm effectively discriminated target gestures from compensatory movements. Results demonstrated high tolerability and engagement, with learning curves showing consistent improvement in gesture execution speed and precision. These findings support the feasibility of our camera-based approach for home use. A pilot study with chronic (>6 months) stroke survivors is currently underway. This work introduces a scalable and engaging platform for targeted, home-based motor rehabilitation, with the potential to significantly improve outcomes and reach underserved populations.

Disclosures: **P. Martinez Viademonte:** None. **A.B. Solano:** None. **D. Blustein:** None. **V.M. Della-Maggiore:** None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

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Topic: F.04. Voluntary Movements

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Mitacs FR130899

Title: Implicit sensorimotor adaptation is preserved in multiple sclerosis

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Abstract: Multiple sclerosis (MS) disrupts the integration of sensory and motor information, leading to a myriad of motor symptoms that can vary considerably from one patient to another. In addition to impairments in motor control, motor learning deficits have also been reported, in particular the more explicit components. Still, it remains unclear whether implicit sensorimotor adaptation, the process through which motor commands are automatically adjusted in response to sensory prediction errors, is affected by MS. Sensorimotor adaptation is critical to practice-related functional reorganization and recovery, hence a better understanding of how this process is altered by MS could help refine rehabilitation interventions. The aim of this study was therefore to examine the impact of MS on both motor performance and implicit sensorimotor adaptation. To this end, we used a visuomotor adaptation task with random perturbations (rotations of $\pm 30^\circ$), a protocol known to isolate the implicit component of adaptation, and compared performance between MS participants ($n = 21$, 14 women, 55 ± 11 years) and aged-matched healthy controls ($n = 21$, 12 women, 55 ± 13 years). The post-rotation bias in hand direction at peak velocity (PRB-HDPV) was the main dependent variable for implicit adaptation. Results revealed clear motor deficits in MS participants, with increased reaction time ($p = .006$), increased movement time ($p = .005$), and greater variability of hand trajectories at peak velocity ($p = .014$), suggestive of noisier motor output. Strikingly, however, there was no difference between MS patients and healthy controls in terms of implicit adaptation (PRB-HDPV: $p = .979$ and $p = .994$ for $+30^\circ$ and -30° perturbations, respectively). These results indicate that, in spite of impaired motor performance, the mechanisms that subtend implicit adaptation may be preserved in MS.

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Poster

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Topic: F.04. Voluntary Movements

Support: ANPCyT, FONTCyT: PICT-2019-02156

Title: Ultra-fast gains: one-second rests drive hippocampal-dependent motor sequence learning

Authors: *G. PEREZ ALSINA, C. SMITH, G. DE PINO, A. YEFFAL, V. DELLA-MAGGIORE;
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Abstract: Interleaving waking rest with practice has emerged as a key determinant of skill memory consolidation, possibly by allowing memory traces to be reactivated and strengthened in real time. In motor skill learning, recent work has shown that performance gains occur during rest pauses between practice blocks rather than during movement execution, a phenomenon known as micro-offline gains (MOGs). The existence of MOGs suggests that consolidation mechanisms take place on a much shorter timescale than previously thought. Growing neuropsychological and neuroimaging evidence suggests that MOGs result from neural replay in the hippocampus. However, in humans, MOGs have been observed over 10-30 second rest windows, whereas replay events in rodents occur on a millisecond scale. Given that humans can perform a 5-item sequence in under a second and considering that neuronal activity during replay is compressed, here we tested whether inserting 1-sec rest periods within practice blocks would improve motor skill learning. To this end, 42 right-handed healthy adults (aged 18-30) performed a 5-item motor skill learning task (41324) with their non-dominant hand inside a 3T MRI scanner. Participants were randomly assigned to two groups: a control group executing continuous sequences within task blocks, and an experimental group in which each motor sequence was followed by a 1-second pause. Both groups followed a 10-sec on/10-sec off block design. We hypothesized that introducing 1-sec rests would shift both behavioral and neural markers of early consolidation. We found that learning was faster in the experimental group, consistent with more frequent opportunities for memory reactivation. In fact, improvements in performance in this group took place during the 1-sec intervals embedded within task blocks (micro-MOGs), whereas in the control group they occurred during the 10-sec rest periods (MOGs). Consistent with the behavioral pattern of results, preliminary fMRI analyses showed that the left hippocampus was more active during task periods in the experimental group, and during the rest periods in the control group ($p < 0.001$, uncorrected), suggesting that hippocampal engagement may be temporally coupled with effective motor memory reactivation. Our findings provide compelling evidence that even 1 second is sufficient to trigger reactivation of motor memories, accelerating learning. This challenges conventional assumptions about minimal temporal requirements for consolidation and opens new avenues for optimizing training protocols across learning domains.

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Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

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Program #/Poster #: PSTR284.10/T12

Topic: F.04. Voluntary Movements

Title: Hd-tdcs demonstrates a lateralized role of the posterior parietal cortex in dominant-hand speed-accuracy relationships during precision drawing

Authors: *N. YAGHOUBI, S. SHARP, J. MANNING, C. SELB, P. E. PIDCOE, B. DEXHEIMER;
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Abstract: 90% of individuals are right-hand dominant for precision tasks like writing, drawing, etc. After losing dominant hand (DH) function due to stroke, peripheral nerve injury, or amputation, an individual may attempt to compensate with the non-dominant hand (NDH). Increased functional connectivity within a left parietal-prefrontal network has been associated with more effective NDH compensation for precision tasks. Additionally, high-definition transcranial direct current stimulation (HD-tDCS) applied to the left, but not right, posterior parietal cortex (PPC) improves gross motor learning transfer to the NDH. In this study, we hypothesized that the left PPC is actively engaged in precision drawing using the NDH and predicted that applying HD-tDCS to the left, but not right PPC would facilitate DH to NDH motor learning transfer. Our preliminary dataset includes 31 right-handed young adults who received HD-tDCS (15 min., 2 mA) over left PPC (n = 10), right PPC (n = 11), or under sham conditions (n = 10). During HD-tDCS, participants used a stylus on a touchscreen to trace shapes with their DH. Following 60 trials, participants immediately performed the same task with their NDH, with no stimulation applied. To assess possible training transfer effects, an additional five participants performed the task exclusively with their NDH under sham conditions. We computed the mean stylus tip velocity and tracing error, defined as the Euclidean distance between the stylus tip and the shape outline at each sampling time point. According to mixed-effects modeling, all groups experienced a significant increase in tracing error ($p < .0001$) and a significant decrease in velocity ($p = .0016$) when switching from the DH to the NDH. The greatest reduction in velocity occurred in the left PPC group ($P < .0001$). When normalizing each participant's tracing error to their velocity, we found significant differences between the stimulation groups depending on hand, as reflected by a significant group by hand interaction ($P = .0002$), with the largest differences in the right PPC group ($P < 0.0001$). To assess training transfer effects, we compared tracing error between the subgroup of five sham participants who completed the task with only their NDH with the sham participants who performed with their DH, then NDH. Individuals who trained initially with their DH exhibited lower tracing errors ($P < .0001$) and higher velocities ($P < .0001$) when using their NDH, compared to those performing with only their NDH. Our findings suggest different effects of left and right PPC HD-tDCS on different aspects of precision drawing. Data collection remains ongoing.

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Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

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Topic: F.04. Voluntary Movements

Support: Natural Sciences and Engineering Research Council of Canada Discovery Grant RGPIN-2025-04104

Title: Dissociating implicit versus explicit motor decision-making during sensorimotor adaptation

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Abstract: The human motor system must determine which errors to learn from and which to ignore. Recent reaching studies show that the motor system is adept at implicitly parsing movement errors into externally-generated error (EGE), caused by environmental disturbances, and internally-generated error (IGE), caused by intrinsic motor variability (Ranjan & Smith, 2018, 2020; Kim et al., 2025). These studies have shown that implicit motor adaptation — the automatic recalibration of sensorimotor mappings — is driven primarily by EGE, while IGE is largely ignored. Specifically, these studies showed that participants adapted to small randomized visuomotor rotations (EGE) while ignoring size-matched errors caused by motor noise (IGE). While these studies show that humans robustly adapt to EGEs as small as 2°, another study (Gaffin-Cahn et al., 2019) showed that people are only aware of EGEs once they are 1.5 times their baseline IGE (i.e., > 3°). However, no prior studies have directly examined this relationship between implicit adaptation and conscious error detection during motor learning. Here, we aimed to unify findings across implicit adaptation and perturbation-detection studies. Given previous results showing that robust adaptation occurs with EGEs as small as 2° and that perturbation detection thresholds require errors that exceed this magnitude, we hypothesized that participants will implicitly discriminate between IGE and EGE and adapt to small EGE magnitudes well below conscious detection thresholds. We tested 8 neurotypical participants across 2 experiments, using identical EGE magnitudes spanning ±1-10° for both sessions. Experiment 1 assessed implicit adaptation and parsing of errors into IGE-EGE components. Experiment 2 assessed conscious perturbation detection based on participants' binary reports of the presence of a perturbation (yes/no) following each reach. Consistent with the previous studies, our participants showed clear discrimination between IGE and EGE: sensitivity to EGE, quantified by adaptive responses, swamped sensitivity to size-matched IGEs. Specifically, we observed robust implicit adaptation and clear parsing of errors in response to EGE as small as 1°, whereas the conscious detection threshold was markedly higher ($d' = 1$ at 4.0° EGE). This work supports the hypothesis that automatic sensorimotor recalibration occurs independently of, and below the threshold for, conscious perturbation detection. This separation suggests distinct functional roles for automatic control versus conscious detection of environmental perturbations.

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Poster

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Topic: F.04. Voluntary Movements

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Dorothy Foehr Huck and J. Lloyd Huck Distinguished Chair of
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Title: The role of left and right posterior parietal cortex during dominant arm visuomotor adaptation and subsequent interlimb transfer: A cathodal HD-tDCS study

Authors: *M. WALI¹, N. M. KITCHEN³, P. R. RUELOS⁵, R. F. DOXTADER², M. YAROSSE⁶, E. TUNIK⁷, R. L. SAINBURG⁴;

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Abstract: Prior research from our laboratory has demonstrated that lesions to the left, but not right, posterior parietal cortex (PPC) impair visuomotor adaptation in both arms. Similarly, in neurotypical adults, cathodal high-definition transcranial direct current stimulation (HD-tDCS) to the left PPC disrupts both adaptation and interlimb transfer (Kumar et al. 2020). In contrast, our recent findings show that anodal HD-tDCS to the left PPC enhances interlimb transfer without affecting initial adaptation. This apparent discrepancy between lesion effects and anodal stimulation may reflect differences in the polarity of the neural disruption. Lesions produce permanent loss of neural function, while anodal HD-tDCS induces a transient increase in cortical excitability. To investigate whether these differences are attributable to stimulation polarity, we applied cathodal HD-tDCS to the left PPC, right PPC, or used a sham condition during right-hand adaptation in right-handed, healthy adults. Participants first completed a baseline phase with veridical visual feedback, followed by a 30° visuomotor rotation applied to the dominant (right) hand (adaptation phase- stimulation applied). Immediately afterward, the stimulation was removed, and the same rotation was applied to the non-dominant (left) hand to assess interlimb transfer, followed by a final veridical phase with the left hand to evaluate after-effects. This design allowed us to test whether contralateral (left PPC), but not ipsilateral (right PPC), cathodal stimulation impairs adaptation, consistent with prior tDCS and lesion findings. If so, these results would suggest that stimulation polarity determines the stage of motor learning affected: cathodal stimulation primarily disrupts initial learning, while anodal stimulation facilitates consolidation and transfer. These findings aim to clarify the lateralized contributions

of the PPC to motor learning and inform the development of targeted neuromodulation strategies for rehabilitation.

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Poster

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Topic: F.04. Voluntary Movements

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Dorothy Foehr Huck and J. Lloyd Huck Distinguished Chair of Kinesiology and Neurology awarded to R.L. Sainburg

Title: The effect of continuous theta burst non-invasive brain stimulation to the posterior parietal cortex in a visuomotor rotation adaptation task

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Abstract: The posterior parietal cortex (PPC) integrates sensory inputs to guide motor actions, particularly during visuomotor tasks. Our previous lesion studies have shown that damage to the left, but not right, PPC impairs visuomotor adaptation in both arms, implicating the left PPC in motor learning. Supporting this, cathodal high-definition transcranial direct current stimulation (HD-tDCS) to the left PPC in neurotypical adults has been shown to disrupt both adaptation and interlimb transfer (Kumar et al. 2020). However, our prior findings using left, but not right, anodal HD-tDCS revealed facilitation of interlimb transfer, but not of adaptation itself. This apparent discrepancy may reflect differences in the polarity, strength, and/or timing of the neural disruption—lesions produce permanent loss of neural function, whereas tDCS modulates excitability transiently. To probe this further, we used continuous theta burst stimulation (cTBS)—a form of repetitive transcranial magnetic stimulation (rTMS) that induces strong but temporary inhibition of cortical activity—to test whether transient suppression of PPC activity mimics lesion effects on visuomotor adaptation. Healthy, right-handed young adults received 40 seconds of cTBS to the left or right PPC (three pulses at 50 Hz repeated at 5 Hz; Huang et al., 2005) at 70% of resting motor threshold (RMT). RMT was defined as the minimum intensity at the motor hotspot that elicited ≥ 50 μ V muscle responses in $\geq 50\%$ of trials. Following a baseline

phase without stimulation or visual perturbation, participants received cTBS or sham stimulation and then completed reaching movements under a 30° visuomotor rotation with their dominant (right) hand. This was followed by reaching with the non-dominant (left) hand under rotated feedback, and a final non-dominant hand phase with veridical feedback to assess after-effects. This study tests whether cTBS to the left PPC replicates the effects of our previous lesion data in inhibiting adaptation. The findings aim to clarify the role of the PPC in motor learning and thus to inform targeted neuromodulation strategies for rehabilitation.

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Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

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Program #/Poster #: PSTR284.14/T16

Topic: I.09. Spatial Navigation

Title: Motor Imagery-Based Brain-Computer Interface for Prosthetic Arm Control Using EEG

Authors: *A. HOVHANNISYAN¹, B. WILBUR², A. GILL², H. KULHANDJIAN²;
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Abstract: Brain-Computer Interfaces (BCIs) based on motor imagery decode user intention from non-invasive EEG signals, offering new ways to control assistive technologies such as prosthetic limbs. This study explores the use of OpenBCI hardware and deep learning to classify EEG signals corresponding to imagined and executed wrist movements. We focused on signals from sensorimotor regions (C3 and C4) and assessed additional encoding triggered by eye closure. Two male participants completed repeated trials involving four distinct wrist movements (e.g., left/right flicks). EEG signals were recorded using the OpenBCI Cyton Board with a 16-channel setup and processed in OpenVibe and MATLAB. We used two convolutional neural networks, ResNet-50 and Inception-V3, fine-tuned through transfer learning for classifying motor imagery. Clear alpha wave patterns and event-related desynchronization (ERD) were observed during tasks. Both models achieved 100% accuracy in distinguishing between wrist movement types, supported by confusion matrices. PCA revealed strong separability between left and right movement data. Eye closure events were also detected and encoded, offering a secondary signal pathway. These results confirm the feasibility of using EEG-based BCIs for high-accuracy motor intention classification. Our framework supports future development of real-time neuroprosthetic control systems. It also highlights the potential of combining motor imagery with eye-blink detection to expand functional control.

Disclosures: A. Hovhannisyan: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

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Program #/Poster #: PSTR284.15/U1

Topic: F.04. Voluntary Movements

Title: Application of HD-tDCS to enhance motor learning: Neural mechanisms and kinematics underlying acquisition of complex motor tasks.

Authors: J. L. SCHOLL, L. A. BAUGH, *T. J. BOSCH;
Univ. of South Dakota, Vermillion, SD

Abstract: This study extends our previous research on the neurological adaptations associated with learning to use chopsticks in right-handed adults. In this previous work, we observed increased functional activity and connectivity changes within the anterior supramarginal gyrus (aSMG), a brain region previously implicated in novel tool use. In the present study, we investigated the effects of high-definition transcranial direct current stimulation (HD-tDCS) on motor learning by applying anodal stimulation to the aSMG in a double-blinded, sham-controlled session, 24 participants (12 active, 12 sham). Those in the active condition received 3mA of HD-tDCS focused over the aSMG while watching a 20-minute video of the task to be performed - picking up a marble with chopsticks and dropping it into a cylindrical container. In comparison, those in the sham condition watched the same video while being exposed to sham stimulation consisting of a 30s ramp-up and ramp-down at both the start and end of the 20-minute video. Immediately following the video task, participants performed 15 one-minute trials where they performed the task modeled in the video while electroencephalography (EEG) recordings were measured. Performance was assessed by the average number of successful marble drops per minute (MDPM) across trials. Additionally, video-based motion analysis was employed to quantify and compare kinematic parameters across groups. Recordings were analyzed using DeepLabCut to compare key kinematic metrics, providing insights into subtle variations in movement patterns associated with the marble task. Results showed an overall significant increase in MDPM from the first to the last trial (10.5 vs. 17.5 MDPM; $p < .001$), with participants in the active stimulation group demonstrating an overall marked performance improvement across all trials compared to the sham group (17.3 vs. 14.1 MDPM; $p < .05$). Preliminary EEG results also show participants receiving active stimulation exhibited enhanced event-related gamma synchronization at Cz, suggesting improved motor refinement and sensory integration. These findings suggest that HD-tDCS enhances the rate of motor learning in novel tool use and underscore the potential of aSMG-targeted stimulation in facilitating complex motor tasks. Further studies are warranted to explore the broader applicability of HD-tDCS in skill acquisition and rehabilitation.

Disclosures: J.L. Scholl: None. L.A. Baugh: None. T.J. Bosch: None.

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

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Topic: F.04. Voluntary Movements

Support: CIHR PJT 165987
CIHR PJT 183970

Title: Cortical motor learning areas in humans outside of primary motor cortex

Authors: *S. EBRAHIMI¹, M. DARAINY¹, M. TIMOTHY², E. LEWIS-HOEBER², D. J. OSTRY¹;

¹McGill Univ., Montreal, QC, Canada; ²McGill Univ., MONTREAL, QC, Canada

Abstract: This paper reports the presence of cortical motor learning areas in humans that do not involve primary motor cortex in learning. The work stems from findings in sensorimotor adaptation, in which direct disruption of motor cortex using magnetic brain stimulation fails to produce an impairment in the retention of newly learned movements, which means the new learning is initially retained elsewhere. These tasks provide a window onto the cortical basis of motor learning, outside of motor cortex. In the present studies, adaptation to altered visual feedback is used as an experimental model of learning. Following learning, continuous theta burst magnetic stimulation (cTBS) is applied to either primary motor cortex, to the rostral parietal cortex, overlying areas 2 and 5, or to dorsal premotor, to test for their causal contribution to learning. In tests of retention, 24h later, disruption of rostral parietal cortex or dorsal premotor cortex each lead to an impairment, whereas disruption of primary motor cortex does not. In further tests, rostral parietal cortex and dorsal premotor cortex are both disrupted in the same participants, to test for independent / additive effects on retention. Here the impairment is no greater than when either area is disrupted alone. This is consistent with the hypothesis that these areas are interconnected in a motor learning circuit such that disruption of either results in a similar impairment when the circuit is broken. Overall, this work shows there is adaptation and learning in humans which does not depend on primary motor cortex.

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Poster

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Program #/Poster #: PSTR284.17/Web Only

Topic: F.04. Voluntary Movements

Title: Engaging reach: comparing classic and adaptive practice for skill retention in consumer head-tracked VR

Authors: *P. PATEL¹, J. CASTILLO¹, H. PANDYA²;

¹Kinesiology, California State Univ. at Fullerton, Fullerton, CA; ²Woven by Toyota, Palo Alto, CA

Abstract: Motor (re)learning typically hinges on repetitive drills that can reduce motivation and curb long-term retention. Wireless virtual-reality (VR) headsets such as the widely available Meta Quest offer immersive, game-like tasks that may keep users engaged while capturing high frequency kinematic data through built-in hand tracking. We will test how different practice schedules grounded in classic motor-learning theory shape acquisition and retention of a seated upper-limb reaching task delivered entirely in VR. Thirty healthy adults (18-25 yr) will wear the headset and, while seated at a table, place virtual puzzle pieces at varying spatial locations. After a short baseline block, participants will complete ten practice blocks in one of two randomized schedules: (1) classic random practice, in which easy, medium, and hard trials are presented in a mixed order, or (2) difficulty-adaptive practice, in which difficulty is adjusted every third trial to maintain $\approx 70\%$ success. Performance will be assessed immediately and after 24 hours in a standardized no-feedback block. Primary outcomes include endpoint accuracy, completion time, and hand-path smoothness extracted from headset tracking data; secondary outcomes include self-reported engagement, comfort, and simulator-sickness scores. We hypothesize that both schedules will yield significant within-session performance gains, but that difficulty-adaptive practice will produce higher next-day retention and greater perceived engagement without increasing cybersickness. We further predict that improvements in hand-path smoothness will correlate with retention accuracy across individuals, suggesting that headset-captured kinematics can serve as sensitive markers of motor learning in VR. These findings will provide one of the first systematic demonstrations of how theory-based practice scheduling influences motor learning in a marker-less VR headset such as the Meta Quest and may guide the design of engaging, data-rich rehabilitation protocols.

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Poster

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Topic: F.04. Voluntary Movements

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Title: Exploration-dependent generalization in de novo motor learning of a redundant visuomotor mapping

Authors: *T. KAWANO¹, S. HAGIO²;

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Abstract: Humans have a remarkable ability to learn arbitrary visuomotor mappings, a process known as *de novo* learning (Krakauer et al., 2019, *Compr. Physiol.*). One key phenomenon observed during this process is generalization (Kawano et al., 2024, *Front. Sports Act. Living.*). To achieve generalization, the motor system must learn a function that enables prediction of outputs for unknown inputs (Poggio and Bizzi, 2004, *Nature*). Previous studies suggested that two types of functions coexist: one based on target position and the other based on movement direction (Wu and Smith, 2013, *J. Neurosci.*). However, it remains unclear how these two types of functions contribute to generalization in *de novo* learning. We address this question by examining how experience with target position and movement direction generalize to untrained movements. Twenty-five healthy participants manipulated two three-axis joysticks to move an on-screen cursor to a series of targets. To induce *de novo* motor learning, we introduced a non-intuitive mapping from 6-dimensional joystick input to 2-dimensional cursor position (Haith et al., 2022, *J. Neurophysiol.*). The task included 900 training trials and 160 interleaved test trials. During training, four distinct target conditions were designed to elicit learning from experience with specific components: one target emphasized target position (P), another emphasized movement direction (D), a third encouraged learning of both elements separately (P+D), and a fourth involved learning both position and direction in combination (PD). In the test trials, we assessed generalization for the P, D, and P+D conditions by evaluating trajectories in both cursor space and joystick control space on novel movements to the target. As training progressed, participants learned to move the cursor accurately to targets. In test trials probing directional experience (D and P+D), initial direction errors were smaller than in trials probing positional experience (P), indicating that direction learning facilitated accurate movement initiation. While movement initiation benefited from directional experience, final position errors in the P+D condition varied across individuals. Because this variability could not be explained by trajectories in cursor space, we examined the 6-dimensional joystick control space. The analysis revealed that participants whose learned trajectories for position and direction overlapped within the control space showed smaller final position errors. Together, these findings suggest that generalization in *de novo* learning depends on how exploration organizes and integrates position- and direction-based control strategies.

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Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

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Program #/Poster #: PSTR284.19/U4

Topic: F.04. Voluntary Movements

Support: NIH R01DC019134-01A1

Title: Disrupting both movement preparation and execution with tms impairs implicit sensorimotor adaptation

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Abstract: Sensorimotor adaptation, driven by a mismatch between predicted and observed sensory outcomes, allows us to maintain accurate movements in response to changes in environmental or body dynamics. A recent study found nonhuman primates adapted less to a 45° visuomotor rotation when movement preparation was disrupted by microstimulation at both dorsal premotor (PMd) and motor (M1) cortexes, suggesting that movement preparation plays a causal role in adaptation, perhaps through seeding the correct neuronal state in M1. However, learning in response to large visuomotor rotations (in humans) involves both implicit sensorimotor adaptation and explicit aiming; thus it is unclear whether stimulation during preparation affected adaptation per se. Here, we test whether cortical stimulation (single-pulse TMS) during movement preparation reduces implicit adaptation in humans (n = 15). To ensure consistent delivery of TMS pulses, we first targeted left M1, using motor-evoked potentials to verify the timing of TMS. Participants reached with their unseen right hand to one of two visual targets when that target turned green. In perturbed trials, participants received continuous visual feedback of their reach, rotated 15° from the target. To isolate implicit adaptation, participants were instructed to ignore the error-clamped feedback. Each perturbed trial was flanked by trials without visual feedback, with single-trial learning (STL) computed as the difference in hand angle between these flanking trials. To disrupt movement preparation, TMS was delivered ~30 ms after a target turned green in perturbed trials. As controls, TMS delivered at reach midpoint (positive control) and no TMS stimulation (negative control). A linear mixed-effect model found differences in STL between stimulation conditions (p = .003): compared to no stimulation (M = 3.92, SE = 0.14), adaptation was reduced when TMS was delivered either in movement preparation (M = 3.34, SE = 0.14, p = .01) or execution (M = 3.32, SE = 0.14, p = .01), with no difference between the stimulation conditions (p = .78). These results show that disrupting the motor cortical state during movement preparation impairs implicit adaptation to an equivalent degree to stimulation during the movement itself, suggesting a critical role of the motor state prior to sensory error processing in adaptation and reinforcing a causal role of movement preparation in this process above and beyond any effects on strategy or aiming. Ongoing work (n = 15) will stimulate PMd in the same paradigm; larger impairments of stimulation than in M1 during preparation, coupled with no impairments during execution, would further support this conclusion.

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Poster

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Topic: F.04. Voluntary Movements

Support: JSPS Grant-in-Aid for Research Activity Start-up #24K23826

Title: Lower-limb-specific visuomotor adaptation processes in goal-directed movements

Authors: *M. MORIYAMA, M. KOUZAKI, S. HAGIO;
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Abstract: Lower-limb movements play an essential role not only in postural control and locomotion, but also in precise, goal-directed actions, such as kicking a ball or climbing stairs. However, how the visuomotor system learns such precise goal-directed lower-limb movements remains poorly understood (Moriyama et al., 2022). While studies on upper-limb reaching have shown that visuomotor adaptation involves both explicit aiming strategies and implicit movement adjustments (Taylor et al., 2014), it is unclear whether these mechanisms operate similarly in the lower limbs. This uncertainty arises from the limited visual feedback available during lower-limb movements, which could lead to less precise visuo-proprioceptive mapping compared to the upper limbs. In this study, we aimed to clarify the adaptation mechanisms of goal-directed lower-limb movements by focusing on explicit and implicit learning.

Sixteen healthy participants performed a forward right-foot swing while seated. A monitor placed in front of the participants showed a target and a cursor representing the foot position in the horizontal plane. After a 30-trial baseline phase, a 20° visuomotor rotation was applied for 160 trials, altering the cursor trajectories relative to the actual foot movement. To quantify explicit learning, participants indicated on the display the direction they intended to move their foot to bring the cursor to the target during the rotation trials. To assess implicit learning, two catch trials without visual feedback of the cursor were conducted every eight rotation trials. Participants were instructed to aim at the target. After the 160 rotation trials, 10 catch trials and 40 washout trials were performed.

Consistent with findings in upper-limb learning, both explicit and implicit processes contributed to visuomotor adaptation of the lower-limb. However, unlike upper-limb learning, the magnitude of implicit learning was greater than that of explicit learning during the early phase. Moreover, the relative contributions of explicit and implicit learning varied substantially across participants in the late phase. Notably, participants who exhibited greater baseline movement variability tended to demonstrate larger amounts of implicit learning. These results suggest that limitations in visuo-proprioceptive integration, possibly less accurate in the lower limbs than in the upper limbs, may reduce the efficacy of visually guided explicit strategies, thereby leading to stronger reliance on implicit learning. Taken together, these findings reveal limb-specific visuomotor adaptation with distinct contributions of explicit and implicit learning.

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Poster

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Location: SDCC Halls B-H

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Program #/Poster #: PSTR284.21/U6

Topic: F.04. Voluntary Movements

Title: Implicit visuomotor adaptation in adults who stutter

Authors: K. LOUDERMILK, J. LIU, H. MARGULIS, *K. S. KIM;
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Abstract: Developmental stuttering is a neurodevelopmental disorder that causes disruptions in speech fluency. It has been demonstrated that people who stutter exhibit motor control difficulties not only in speech tasks but also movements in the non-speech effector system, such as finger or arm motion. Notably, studies have reported that people who stutter show limited sensorimotor adaptation (i.e., updating subsequent movements in response to sensory errors) in both speech auditory-motor (i.e., updating speech movements in response to altered auditory feedback) and upper limb visuo-motor (i.e., updating arm movements in response to altered visual feedback) tasks. Previous research established that sensorimotor learning can consist of implicit and explicit learning components. Given that speech auditory-motor adaptation is mostly, if not entirely, implicit (i.e., participants are unaware of the learning), it is thought that people who stutter have limited implicit adaptation in the speech effector system. It remains unclear, however, whether such limited implicit adaptation also extends to the upper limb effector system. In the current study, we recruited adult participants who stutter and non-stuttering sex-, age-, and handedness-matched control individuals to perform arm reaching movements towards 8 different targets placed around the workspace in the KINARM system (KINARM End-Point, BKIN Technologies), and applied clamped visual feedback (8° counterclockwise to targets) to examine implicit adaptation. During clamped visual feedback, a visual cursor maintained a fixed direction counterclockwise from targets relative to the center of the workspace, regardless of the participant's actual arm motion. Even though participants were informed that their cursor direction was independent of their actual reaching movement, they gradually adjusted their reach towards clockwise direction across trials in response to clamped visual feedback, achieving implicit adaptation. The cursor was also invisible towards the end of reaching (i.e., near targets), preventing any potential explicit adaptation. Data collection is underway, but preliminary analyses from 11 stuttering and 8 non-stuttering participants revealed a subtle group difference in the rate of adaptation. Our preliminary finding suggests that implicit adaptation difficulties in adults who stutter may generalize across different effector systems. Implications regarding sensorimotor mechanisms underlying stuttering will be discussed.

Disclosures: K. Loudermilk: None. J. Liu: None. H. Margulis: None. K.S. Kim: None.

Poster

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Location: SDCC Halls B-H

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Program #/Poster #: PSTR284.22/U7

Topic: F.04. Voluntary Movements

Support: NIH 1 R21 HD114233-01

Title: Mapping infant brain activity during goal-directed reaching movements using functional ultrasound imaging (fUSI)

Authors: *A. E. STEPANIAN¹, S. SAKELLARIDI³, I. SAHIN², M. ALANIZ⁴, Y. HARRIS⁴, C. LIU⁵, E. KOKKONI^{1,2}, V. N. CHRISTOPOULOS^{6,1,2};

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Abstract: The first year of life is marked by rapid brain development, critical for lifelong motor and cognitive skills. Yet, most infant motor studies rely primarily on behavioral observation, providing limited insight into neural mechanisms. Existing functional neuroimaging faces limitations with awake infants due to motion artifacts and compliance issues, resulting in high exclusion rates and few studies on early motor systems. Functional ultrasound imaging (fUSI) is an emerging neuroimaging technology that addresses these limitations. It offers high sensitivity, broad spatial coverage, and exceptional spatiotemporal resolution (~100 μ m at <1 Hz). Importantly, fUSI can leverage the infant fontanelle as a natural acoustic window, enabling noninvasive brain imaging, more tolerant to motion than existing techniques. Here, we report the first simultaneous acquisition of fUSI recordings, electromyography (EMG), and kinematic data in awake infants while performing upper limb motor tasks. Infants (6-8 months old) participated in a single experimental session structured into alternating blocks: 30 seconds of resting state followed by 60 seconds of goal-directed reaching. Infants were seated either in a high chair or on a parent's lap to ensure trunk stability and comfort. During resting state blocks, infants were presented with an age-appropriate video to maintain engagement while minimizing movement. During reaching blocks, objects were presented at the infants' body midline to elicit reaching actions. A fUSI probe was placed on the anterior fontanelle using a custom-designed headset, aligned with the superior sagittal sinus, to collect functional neuroimaging data. EMG and inertial sensors were attached to the limbs to record muscle activity and limb acceleration, respectively. Multi-view video recordings were collected to annotate events of interest, such as reaching actions and segments of no movement. This pilot study marks a significant step in infant neuroimaging, demonstrating the simultaneous acquisition of fUSI recordings, EMG, and kinematic data in awake, behaving infants. This integrated approach not only overcomes critical methodological limitations of prior neuroimaging techniques in this population but also proves feasible, yielding robust data streams from the participants. Ongoing efforts, including further analysis of these data and collection from additional participants, aim to identify localized changes in cerebral blood volume within expected sensorimotor cortical areas during movement activities, and to potentially map their temporal alignment with observed reaching actions, EMG activity, and kinematic changes.

Disclosures: **A.E. Stepanian:** A. Employment/Salary (full or part-time): Neuroscience Graduate Program, Univ. of California, Riverside, Riverside, CA. **S. Sakellaridi:** A. Employment/Salary (full or part-time): Research Institute, Casa Colina Hospital and Centers for Healthcare, Pomona, CA. **I. Sahin:** A. Employment/Salary (full or part-time): Department of

Bioengineering, Univ. of California, Riverside, Riverside, CA. **M. Alaniz:** A. Employment/Salary (full or part-time): Department of Pediatrics, Casa Colina Hospital and Centers for Healthcare, Pomona, CA. **Y. Harris:** A. Employment/Salary (full or part-time): Department of Pediatrics, Casa Colina Hospital and Centers for Healthcare, Pomona, CA. **C. Liu:** A. Employment/Salary (full or part-time): Neurorestoration Center and Department of Neurological Surgery, Univ. of Southern California Keck School of Medicine, Los Angeles, CA. **E. Kokkoni:** A. Employment/Salary (full or part-time): Neuroscience Graduate Program, Univ. of California, Riverside, Riverside, CA., Department of Bioengineering, Univ. of California, Riverside, Riverside, CA. **V.N. Christopoulos:** A. Employment/Salary (full or part-time): Neuroscience Graduate Program, Univ. of California, Riverside, Riverside, CA., Department of Bioengineering, Univ. of California, Riverside, Riverside, CA., Department of Biomedical Engineering, Univ. of Southern California, Los Angeles, CA..

Poster

PSTR284: Reaching Control: Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR284.23/U8

Topic: F.04. Voluntary Movements

Title: Motor learning history constrains optimal variability patterns for novel precision constraints

Authors: ***J. WIJFFELS**¹, R. RANGANATHAN²;

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Abstract: In tasks requiring high precision (such as surgery), humans are able to adapt movements to task-specific precision demands, enhancing accuracy when necessary while exploiting redundancy to buffer noise. Optimal Feedback Control (OFC) explains this via the minimum intervention principle, predicting that variability is minimized only in task-relevant dimensions. Yet, empirical findings often show an overall reduction in variability with learning, suggesting convergence onto a stable nominal trajectory. One potential source for this discrepancy is that classical OFC does not account for learning history or persistent coordination tendencies acquired during training. We sought to determine whether prior motor learning influences variability when precision demands are altered after learning. If variability patterns purely reflect optimal responses to current task demands, then prior motor learning should have no effect on how variability is organized. Participants (n = 12) trained in virtual reality on a sinusoidal via-point task with five 3D cylindrical targets. We manipulated the location of task-relevant precision demands while keeping the spatial path identical. The target diameter (task-relevant dimension) varied between groups, while the target depth (task-redundant dimension) remained constant. One group trained with a precision requirement early in the sequence, and a second group trained with the requirement late. After training, both groups were transferred to a common via-point task that preserved the same average path but altered precision requirements. Allowing assessment of whether post-learning reduction in variability reflects a fixed nominal

trajectory or remains adaptable. In the task-relevant dimension, variability was consistently higher between via-points than at the via-points, consistent with OFC's prediction of strategic variability modulation via the minimum intervention principle. With learning, variability decreased in both relevant and redundant dimensions, in line with prior findings. Crucially, in the transfer task, performance differed by training history, on both dimensions, suggesting that participants had adopted a feedforward strategy that persisted despite altered precision constraints. The motor system appears to prioritize retaining previously learned coordination strategies over reorganizing behavior to fit new task demands. This challenges classical OFC, which does not consider learning history. Future models should integrate such history, recognizing that system-level optimality may diverge from that of the task at hand.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Program #/Poster #: PSTR285.01/U9

Topic: F.06. Posture and Gait

Support: AHA 16BGIA27250047

Title: Cognitive Decline Outweighs Adiposity in Influencing Lower Limb Gait Kinematics in Older Adults

Authors: *S. L. GORNI¹, H. MENG²;

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Abstract: Increased adiposity amounts have been demonstrated to negatively impact gait and postural control in older adults. Specifically, increased adiposity has been found to reduce peak angular displacement and range of joint motion of the lower extremities during gait in individuals over the age of 65 years. Recently, the influence of cognitive impairment on gait function has emerged as a potential predictor of gait abnormalities. However, the combined influence of increased adiposity and cognitive impairment in older adults during gait-based activities has not been specifically investigated. In this study, thirty older adults (age 70 ± 7 years; 15 men and 15 women) with body mass indices ranging from 18.5 - 40.0 kg/m² completed a series of anthropomorphic measurements, walking tasks, and cognitive evaluations. Anthropomorphic measurements consisted of body mass index evaluation and body composition, including assessment of adipose tissue amounts (via dual x-ray absorptiometry, DXA). Joint angle values for the major joints of the lower extremities (hip, knee, and ankle) were measured bilaterally over a straight-line walking distance of 200m while participants walked at their preferred natural walking speed. Potential cognitive impairment was assessed using the Montreal Cognitive Assessment (MoCA). Working memory via the N-back task was evaluated while

participants were standing quietly on a force platform. Joint angle values during walking, including average values and range of motion, were significantly impacted by cognitive performance across all evaluated joints of the lower extremity ($p < 0.05$). Adiposity measures only impacted range of motion of the ankle joint ($p < 0.005$). These data indicate that impairments in cognitive function, specifically in the domain of working memory, underlie gait aberrations in a stronger fashion than increased adiposity with aging. Changes gait function with aging could be used as a potential marker of impaired cognitive function, independent of documented adiposity influences on gait in aged populations.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

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Program #/Poster #: PSTR285.02/U10

Topic: F.06. Posture and Gait

Support: Minnesota Office of Higher Education #232836
Minnesota Office of Higher Education #232568

Title: Opioid and endocrine contributions to contralateral and postural effects after brain and spinal cord injury: toward targeted pharmacological therapies

Authors: ***I. LAVROV**¹, **H. WATANABE**², **M. ZHANG**³, **G. Y. BAKALKIN**²;
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Abstract: Lateralized brain and spinal cord injuries can induce asymmetric postural and motor deficits, although, the underlying mechanisms remain unclear. We investigated the neural and neuroendocrine pathways mediating contralateral effects of traumatic brain injury (TBI) and lateral spinal cord injury (SCI), with a particular focus on the role of the opioid system. The unilateral TBI in rodents induced hindlimb postural asymmetry (HL-PA), characterized by contralateral limb flexion, even in animals with completely transected spinal cords, implicating a neurohormonal mechanism acting via the humoral pathway. These effects were modulated in a side-specific manner, i.e. delta- and kappa-opioid antagonists selectively inhibited HL-PA following left- and right-sided TBI, respectively, indicating a lateralized opioid-mediated neuromodulation system. Gene co-expression analyses revealed ipsilateral coordination of opioid gene networks between the hypothalamus and lumbar spinal cord, which was disrupted by TBI. Similarly, high lumbar lateral hemisection (LHS) produced HL-PA marked by ipsilateral hindlimb flexion, which persisted after complete spinal cord transection at the level of injury.

Naloxone treatment abolished this asymmetry, suggesting that postural changes were mediated by lumbar opioid circuits exhibiting neuroplasticity. Conversely, cervical LHS performed after lumbar transection induced contralateral HL-PA that was resistant to naloxone, pointing to a non-opioid, likely supraspinal neuroendocrine mechanism. Together, these findings highlight a complex interplay between spinal and supraspinal opioid and non-opioid pathways in generating lateralized postural deficits after TBI and SCI. The discovery of opposing, side-specific mechanisms open new avenues for targeted pharmacological interventions aimed at restoring motor symmetry in patients with hemiparesis or hemiplegia following neurotrauma.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Program #/Poster #: PSTR285.03/U11

Topic: F.06. Posture and Gait

Support: RS-2021-KD000003
RS-2023-00248118

Title: Efficacy of Exoskeletal Wearable Robot for Gait Recovery in Subacute Stroke Patients: A Multicenter, Parallel-group Randomized Controlled Trial

Authors: *H.-B. CHOI, W. CHANG;

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Abstract: Introduction: Overground exoskeletons, designed to assist gait by providing torque at lower limb joints, offer a potential means to enhance active participation, balance, and muscle activation. However, robust evidence from large, multicenter trials is lacking regarding their efficacy in subacute stroke patients. The aim of this international, multicenter, randomized, controlled trial was to investigate the effect of overground gait training using a torque-assisted exoskeleton in subacute stroke patients. **Methods:** A total of 151 subacute stroke patients were enrolled in this study. Participants were randomized to either the robotic-assisted gait training (RAGT) group, receiving 30 minutes of conventional gait training plus 30 minutes of exoskeleton-assisted walking (ANGEL LEGS M20), or the control group, receiving 60 minutes of conventional gait training, five times per week for four weeks (total 20 sessions). Functional assessments including FAC (primary outcome), Fugl-Meyer Assessment-Lower Extremity (FMA-LE), Motricity Index for lower limb (MI-LL), Trunk Control Test (TCT), and Berg Balance Scale (BBS) were performed before and immediately after the intervention by blinded raters. Independent ambulation was reassessed by telephone interview three months post-intervention. Statistical analyses included intention-to-treat comparisons, with non-parametric

and ANCOVA methods as appropriate. **Results:** There were no serious adverse events related to the interventions, and dropout rates tended to be higher in the RAGT group without statistical significance. Both groups showed significant improvement in FAC after the intervention; however, no significant difference between groups in the primary outcome. Independent ambulation was achieved by 77.6% (RAGT) and 75.4% (control). Both groups exhibited significant gains in balance (TCT, BBS) and lower limb motor function (FMA-LE, MI-LL); however, the RAGT group demonstrated a significantly greater improvement in lower limb strength (MI-LL, $p=0.034$). **Conclusion:** Overground gait training using a torque-assisted exoskeleton was found to be as effective as conventional gait rehabilitation in improving ambulatory function in subacute stroke patients, with the added benefit of greater gains in lower limb strength. These findings support the potential of exoskeletal RAGT as a viable intervention for gait recovery in this population, especially for those with preserved balance function.

Disclosures: H. Choi: None. W. Chang: None.

Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR285.04/U12

Topic: F.06. Posture and Gait

Support: P2CHD086844
NIH/NICHD R25HD106896

Title: Contribution of Altered Bilateral Functional Brain Connectivity to Poor Balance Control in Chronic Stroke Survivors.

Authors: *K. KUKKAR¹, S. LI², M. MIYAKOSHI³, P. J. PARIKH¹;

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Abstract: Balance control is a key indicator of independence in activities of daily living, and its impairment contributes to falls in stroke patients. Stroke causes brain reorganization, which may alter brain network connectivity. Such alterations can deleteriously affect balance control and the generation of effective responses to prevent a fall when balance is challenged. Remarkably, how changes in brain network dynamics following stroke contribute to impaired balance control remains unknown. We have previously identified a network of fronto-parietal brain regions related to balance control during a challenging balance task in neurotypical adults. In this study, we investigated stroke-related changes in brain network dynamics for balance control during a similar task. We recruited fifteen stroke participants with mild-to-moderate severity and an equal number of age- and gender-matched healthy adults. Participants maintained upright stance in response to balance perturbations on a computerized platform. Whole-head brain activity was simultaneously recorded using electroencephalography (EEG). A second objective was to

develop a custom MATLAB-based application to streamline EEG study creation and enable MRI-based visualization of brain activations and dipole density distributions. Stroke participants showed poor performance on the clinical Berg Balance test and the lab-based balance task compared to healthy controls. EEG source activity was localized to central posterior parietal cortices (PPC), supplementary motor area (SMA), and bilateral dorsolateral prefrontal cortices (dlPFC) in both groups. Stroke participants showed higher SMA-PPC connectivity, which was negatively associated with balance, while this association was positive in controls. This suggests maladaptive sensorimotor integration post-stroke. Additionally, stroke participants showed lower SMA-dlPFC connectivity on the affected side and lower PPC-dlPFC connectivity on the less affected side. Notably, higher SMA-dlPFC connectivity on the affected side correlated with better balance. Our findings suggest prefrontal regions are recruited to compensate for impaired sensory processing. These altered connectivity patterns may serve as neurophysiological markers of balance recovery and inform development of cortically targeted interventions to reduce falls after stroke.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Topic: F.06. Posture and Gait

Support: Research Council of Finland (#350528) to SW
Research Council of Finland (#361732) to HP

Title: Sensorimotor inhibition differs during quiet standing and anticipation of balance perturbation between young and older adults.

Authors: *J. JUURAKKO¹, H. PIITULAINEN¹, S. VEKKI¹, W. TAUBE², J. M. PIIRAINEN¹, S. WALKER¹;

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Abstract: Aging is accompanied by impairment of balance performance and deterioration of the sensorimotor system, where cortical inhibitory circuits may play a key role. However, there is still uncertainty about their functional causality. We aimed to reveal how aging affects neural strategies at the anticipation phase and during balance perturbation and whether inhibitory GABAergic activity predicts balance performance. We measured 28 young (27.4 ± 4.2 y), 35 middle-aged (57.7 ± 4.7 y), and 27 older (73.6 ± 2.6 y) healthy participants. A custom-built, sliding force platform was used to measure the center of pressure (CoP) of the subject during quiet standing with eyes open, and during perturbations mimicking a backward-falling situation (300 mm displacement at a velocity of 170 mm/s with an acceleration of 2000 mm/s^2). Sway was

determined as the average velocity of CoP during platform displacement. GABAergic activity was estimated with sensorimotor beta power (13-30 Hz) during quiet standing measured with a 9-channel electroencephalography and short-interval intracortical inhibition (SICI). SICI (80% / 120% of active motor threshold (aMT), interval of 2 ms) and corticospinal excitability (single pulse motor evoked potential (MEP), 120% aMT) were assessed during the anticipation phase and 90 ms after the start of perturbation (20 trials each). Stimulations were delivered with a neuronavigated transcranial magnetic stimulator to the M1 cortex. SICI and MEP were measured with electromyography from m. tibialis anterior with a pseudo-monopolar arrangement. Young adults had the lowest sway velocity (97 ± 13 mm/s) and relative beta power ($33.7 \pm 10.1\%$) compared to middle-aged (113 ± 20 mm/s, $p < 0.01$, $52.5 \pm 12.0\%$, $p < 0.001$) and older (141 ± 27 mm/s, $p < 0.001$, $56.0 \pm 10.7\%$, $p < 0.001$) adults. Middle-aged adults had less sway velocity ($p > 0.001$) than older adults, but no difference in relative beta power ($p > 0.05$). SICI showed a main effect of age during the anticipation phase (young: $40 \pm 25\%$, middle-aged: $38 \pm 23\%$, older: $25 \pm 21\%$ inhibition, $p < 0.05$). SICI diminished during perturbation, with no between-group differences. In older adults, higher SICI during anticipation was associated with higher sway velocity ($r = 0.46$, $p < 0.05$), while young adults showed the opposite ($r = -0.45$, $p < 0.05$). Older adults with higher relative beta power had lower sway velocity ($r = -0.49$, $p < 0.05$). In conclusion, sensorimotor inhibition differs between young and older adults during the anticipation phase before perturbation. Furthermore, the correlations between estimated GABAergic activity and dynamic balance performance suggest that age-modulated inhibition has functional relevance.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

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Program #/Poster #: PSTR285.06/U14

Topic: F.06. Posture and Gait

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Foundation for Physical Therapy Research Promotion of Doctoral Studies
I

Title: Pain Impairs Locomotor Learning Differently Across the Lifespan

Authors: *S. JACKSON¹, R. T. POHLIG², S. M. MORTON³;
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Abstract: Older adults experience chronic pain and its negative health-related outcomes at a much higher rate than young adults. Physical therapy is considered a first-line treatment to reduce pain and promote functional recovery in older adults, and many active interventions rely

on motor learning principles, e.g., retraining lifting mechanics in a patient with chronic low back pain. However, emerging evidence suggests that pain may interfere with certain aspects of motor learning, particularly the ability to retain newly acquired motor skills across days. Yet, no study to date has assessed whether motor learning or retention is specifically impaired in older adults with pain. To test this hypothesis, we recruited a cohort of healthy young adults and a cohort of healthy older adults, and randomized each into one of two groups: a control group that experienced no pain during the study, and a pain group that experienced acute pain via our experimental model of topical capsaicin (0.1%) and heat applied to the anterior lower leg. On Day 1, all participants learned a new asymmetric stepping pattern using visual feedback. A monitor displayed real-time feedback of step lengths, represented as bars growing vertically on the screen. During learning, the feedback was distorted, making one leg appear to take longer steps and the other to take shorter steps. To hit the visual step length targets, participants had to acquire a novel step asymmetry of 9%. The pain group experienced pain only during this period. Twenty-four hours later, retention was tested with the 9% distorted visual feedback. A motion capture system recorded step lengths during baseline and learning periods on Day 1 and the retention period on Day 2. Preliminary results in older adults suggest that learning of the locomotor pattern is impaired in older adults who experience pain during learning ($4.7\% \pm 3.4\%$; $n = 11$) compared to older adults who learn pain-free ($8.7\% \pm 3.5\%$; $n = 11$). This contrasts with young adults, who appear to learn similarly whether they experience pain during learning ($8.2\% \pm 1.9\%$; $n = 13$) or learn pain-free ($8.8\% \pm 1.7\%$; $n = 11$). Taken together, these findings suggest that pain's effect on motor learning is moderated by age.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

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Program #/Poster #: PSTR285.07/U15

Topic: F.06. Posture and Gait

Title: Early and prolonged cortical electrical stimulation preserves motor functions and nigrostriatal dopaminergic neurons in the mitopark model of parkinson's disease

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Abstract: Parkinson's disease (PD) is the second most prevalent neurodegenerative disorder among the elderly. While pharmacological treatments alleviate symptoms for many individuals with PD, they often fail to provide complete symptom control, highlighting the need for alternative therapeutic strategies. Neuromodulation techniques, particularly cortical electrical stimulation (CES), have emerged as potential interventions by modulating cortical plasticity. However, the therapeutic efficacy of CES in PD remains uncertain. This study aimed to evaluate the therapeutic potential of a novel CES protocol using a transgenic MitoPark PD mouse model.

The MitoPark PD mouse model was applied to identify the therapeutic effects of long-term (starting 8 weeks old, 1 session/day, 2 days/week, for a duration of 12 weeks) on motor function and neuroprotection. After the CES intervention, detailed functional behavioral tests including open-field locomotor activity, beam walking test, gait analysis, as well as nigrostriatal dopaminergic neuron degeneration level. The results demonstrated that early and prolonged CES significantly ameliorated the motor deficits in locomotor activity, gait patterns, and beam-walking performance in the MitoPark mice. Furthermore, immunohistochemical analysis revealed a higher survival rate of nigrostriatal dopaminergic neurons in the CES-treated group compared to the sham treatment group. These findings suggest that long-term CES exerts neuroprotective effects on nigrostriatal pathways and mitigates motor deficits in the MitoPark PD model. This study provides compelling evidence for the potential clinical application of CES as an adjunctive treatment for PD.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Topic: F.06. Posture and Gait

Support: NIH Grant R01HL102457
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Title: Associations of physical balance assessments with brain white matter free water content measured with MRI diffusion tensor imaging (DTI)

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Abstract: Falls can lead to brain injury, bone fractures, and increase risks of disability and mortality in older adults. Cognitive disability, the leading cause for disability in the United States, increases older adults' fall risk by ninefold. Recent findings show that 360° turn and function reach tests (FRT), both brief balance assessments, are effective at identifying individuals with cognitive impairment at elevated risk for falls and mobility disability. Disruptions in brain structural connectivity, particularly in the prefrontal cortex, adversely affects motor coordination and balance in older adults. Diffusion tensor imaging (DTI) quantifies brain free water (FW), a marker of white matter microstructural integrity that may reveal neuronal edema, atrophy, and demyelination with aging. This study examined the associations of 360° turn and FRT with FW measured using DTI. A total of 167 cognitively normal (MMSE > 25) adults (20-81 yrs) completed the balance and MRI testing. Balance was assessed through: 1) 360° turn which is the average of 2 turns (measured in seconds) without spinning on one foot

(one clockwise turn and one counterclockwise turn) averaged together; and 2) FRT which measured the greatest distance (cm) a participant could reach forward with both arms stretched out without altering foot position. Brain FW was measured using DTI on a 3-Tesla scanner (Phillips Medical Systems, Best, The Netherlands). Both voxel-wise and region-of-interest analyses of associations of FW with balance assessments were performed through multiple linear regressions. Older (> 65 yrs) participants had a lower FRT (32.4 ± 6.6 cm) compared to young (20-45 yrs, 44.0 ± 7.2 cm; $p < 0.001$) and middle-aged (45-65 yrs, 40.5 ± 8.3 cm; $p = 0.041$) participants. Older participants also had a longer 360° time (2.9 ± 0.5 sec) compared to younger (2.3 ± 0.4 sec, $p < 0.001$) and middle-aged (2.6 ± 0.5 , $p = 0.034$). For the total sample, when controlled for age, sex, and education, a longer 360° was associated with a higher FW ($R^2 = 0.245$, $p < 0.001$) content in the bilateral anterior corona radiata and the bilateral body of the corpus callosum. When controlled for age, sex, and education, higher FRT was associated with lower FW ($R^2 = 0.298$, $p < 0.001$) in the right superior corona radiata, the right anterior limb of the internal capsule, and the fornix. Poor balance performance (i.e., lower FRT and longer 360° turn) was associated with higher brain white matter FW content in the regions which are involved in the integration of cognitive and motor function, suggesting a central mechanism of decline in balance performance with age.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Topic: F.06. Posture and Gait

Support: NIDA Grant R01DA047637

Title: Exploring Motor Deficits and Brain Oscillation Alterations at the Early Stage of Alzheimer's Disease

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Abstract: Alzheimer's disease (AD) is a progressive neurodegenerative disorder and the leading cause of dementia worldwide. Although memory decline and cognitive impairment are widely recognized as the hallmarks of AD, growing clinical evidence suggests that motor deficits may emerge earlier in the disease progression, before cognitive decline, and serve as sensitive biomarkers for clinical detection. To investigate locomotor activity changes at an earlier stage in AD model animals, we explored behavioral performance and brain oscillation patterns in Alzheimer's disease model mice (AD; App^{NL-F/NL-F}; RIKEN Brain Science Institute, Japan) and

age-matched wild-type mice (WT; C57BL/6J, JAX:000664; Jackson Laboratory, USA). A total of 37 AD and 16 WT mice across three age groups (3-4, 6-8, and 10-12 months) were used. We employed behavioral testing (grid-walking, elevated beam, and rope pulling) and DeepLabCut-based video tracking to analyze locomotor behaviors, in vivo electrophysiological recordings to assess brain oscillations, and morphological staining for amyloid- β plaques. We identified age-related locomotor behavior abnormalities in AD mice compared to WT mice. In the grid-walking task, 3-4 months old AD mice exhibited significantly increased travel distance (AD: 28.72 ± 4.32 m vs WT: 16.48 ± 2.15 m; mean $\pm$ SD, $p = 0.045$) and movement percentage (AD: $46.6 \% \pm 3.4$ vs WT: $31.3 \% \pm 2.6$; mean $\pm$ SD, $p = 0.014$) along with a relatively shorter tail tip swing distance. In the elevated beam task, 3-4-month-old AD mice showed significantly reduced travel speed on both rough (AD: 34.05 ± 4.63 mm/s vs WT: 61.71 ± 5.53 mm/s; mean $\pm$ SD, $p = 0.0013$) and smooth (AD: 35.87 ± 4.67 mm/s vs WT: 64.79 ± 4.41 mm/s; mean $\pm$ SD, $p = 0.0007$) surfaces. In the rope-pulling task, 6-8-month-old AD mice demonstrated less rope length pulled (AD: 23.65 ± 7.35 cm vs WT: 66.25 ± 14.80 cm; mean $\pm$ SD, $p = 0.042$). Meanwhile, in AD mice, two-channel intracranial EEG recordings from ipsilateral dorsal hippocampus and lateral vestibular nuclei revealed disrupted theta-gamma coupling starting from 6-8 months, and morphological staining showed sparse amyloid- β plaque formation beginning from 8 months. Our findings indicate that specific locomotor behavioral features could serve as a non-invasive and sensitive biomarker for early-stage AD screening and guide potential therapeutic strategies.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Topic: F.06. Posture and Gait

Support: JSPS KAKENHI Grant Number JP25K14494

Title: Biphasic control of unidirectional sway underlies body sway variability during quiet standing in middle-aged and older women

Authors: *S. SASAGAWA;
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Abstract: Previous studies have shown that age-related declines in standing balance ability are associated with a higher risk of falls in older adults. Therefore, investigating the physiological mechanisms underlying this age-related decline in standing balance ability is essential for preventing falls and maintaining quality of life in older adults. This study aimed to investigate the physiological mechanisms underlying individual differences in body sway variability among

middle-aged and older women during quiet standing. Forty-eight women aged 54-89 years were asked to stand barefoot and quietly on a force platform for 30 seconds. Based on the measured ground reaction forces, the position of the body's center of mass (CoM) and its acceleration (ACC) in the anterior-posterior direction were obtained. The CoM sway trajectory was decomposed into unidirectional (i.e., forward or backward) sway segments, from which the total number, mean size, mean duration, and mean speed of the individual sways were calculated. The results indicated that body sway variability over a longer timescale (~30 seconds) was primarily determined by the mean size of the unidirectional sways, rather than by their total number. Furthermore, the mean size of these sways was more closely associated with their mean speed than with their mean duration. A time-locked averaging technique revealed a biphasic pattern of ACC that regulates the unidirectional CoM movements. This averaged pattern showed that an increased amplitude of ACC from the braking phase of the previous sway to the acceleration phase of the current sway was responsible for the increased speed of the CoM. These findings support the idea that reduced accuracy in the biphasic, ballistic-like ACC pattern increases both the speed and displacement of the CoM during unidirectional movement, thereby contributing to greater sway variability over longer timescales during quiet standing.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR285.11/V3

Topic: F.06. Posture and Gait

Title: Assessing resting and maximal trunk compliance in participants with and without chronic low back pain

Authors: *F. ABTAHI¹, P. E. PIDCOE¹, A. STAMENKOVIC¹, S. M. VAN DER VEEN², J. S. THOMAS¹;

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Abstract: Many studies have reported altered motor control in people with chronic low back pain (CLBP). Some studies suggest that pain leads to increased trunk muscle co-contraction (general stiffening occurs in multiple directions), while other research indicates that CLBP reduces muscle co-activation. Methods for assessing trunk stiffness include biomechanical spine modeling, but this approach has limitations due to generalized anatomical features and complexity. The Trunk Compliance Index (TCI) is a biomarker that measures trunk compliance (i.e., 1/stiffness) by trunk displacements relative to normalized force on the trunk across frontal, sagittal, and transverse planes using direct trunk measurements. We aimed to determine if TCI varies across six directions during unexpected and maximally resisted multi-directional trunk perturbations in participants with CLBP and healthy (H) controls. Also, we assumed that sex

would impact TCIs in both expected and unexpected trials in CLBP. The study included 40 participants: 22 with CLBP (mean age 34.27, 50% female) and 18 in the H group (mean age 29.55, 50% female). A set of 30 sudden unexpected multi-directional perturbations was applied to the trunk using a puller motor system in six directions across sagittal, frontal, and horizontal planes, randomized, and each direction was repeated five times. Afterwards, a second set of 30 sudden, expected (subjects knew the timing and direction of impulses) multi-directional perturbations were applied, with subjects instructed to maximally stiffen their trunk before perturbation onset. Six separate three-way mixed-model ANOVAs were conducted; interactions were analyzed via pairwise comparisons. As expected, there were main effects of perturbation types (expected, unexpected) across all TCIs for both genders and groups, except for LSB. However, unexpectedly, there was no main effect of the group (LBP, H) on TCIs. There were no significant sex differences in TCI measurements across six directions, except for LSB in CLBP, where females exhibited higher stiffness than males. Results showed no significant difference in TCI at resting states between CLBP and H groups, as well as no significant difference at maximally stiffened states between both groups in all six directions. This study did not support previous findings of different TCIs at resting state between CLBP and H but confirmed higher stiffness in females with CLBP compared to H in LSB. The results support previous research that CLBP doesn't increase trunk stiffness and offer insights into spinal stiffness, potentially enhancing CLBP interventions.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Program #/Poster #: PSTR285.12/V4

Topic: F.06. Posture and Gait

Title: Assessing effects of age, level of resistance, and direction of step practice on gait performance

Authors: K. RODRIGUEZ, *J. M. HONDZINSKI;
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Abstract: Stepping strategies change across the lifespan to assist humans with mobility. When individuals lack the ability to effectively control stepping, rehabilitation specialists can intervene. Successful strategies for improvements in gait often involve increasing strength and improving quality of stepping. Unidirectional and multidirectional step practice reveals improved gait, like greater weight shifting, balance, and intralimb coordination compared to baseline in patients or people with age-related declines. Less is known about step practice effects in healthy young adults (YA) or middle-aged adults (MA), the latter of which may already show evidence of aging declines. We reasoned that introduction of resistance during step practice might also cause

aftereffects revealing gait improvements. We investigated the effect of age on comfortable walking gait to learn more about the age-related effects on gait in our participants and to determine if acute bouts of step practice with and without resistance would influence gait across different age groups. We hypothesized that individuals will produce greater step length and gait speed after acute bouts of resistance step practice compared to baseline with the greatest changes observed in older adults (OA). Healthy YA (18-25 years), MA (45-55 years), and OA (65-75 years) performed three comfortably paced walking trials on an instrumented GaitRite mat before (baseline) and after three 45 second bouts of practice in each step condition in a single session. Step conditions included two stepping directions (unidirectional (forward and return)—UDS, multidirectional (forward and return, lateral and return, backward and return)—MDS) with three levels of resistance (none-N, 10% of body weight on the trunk-T, or ankles-A). Randomization of the 6 practice conditions (UDS-N, MDS-N, UDS-T, MDS-T, UDS-A, MDS-A) and 5 minutes of rest between conditions helped negate potential order effects and practice accumulation, respectively, on spatiotemporal measures of gait, along with heart rate (HR) and ratings of perceived exertion (RPE) just prior to walking trials. As expected, HR and RPE increased, especially after weight application, compared to baseline values with RPE highest for the weighted ankle step conditions. Gait velocity was highest in YA, while OA often demonstrated the shortest step lengths and highest cadence. MA revealed similarities to YA and OA that varied within this group and across gait variables. Although we observed age-related differences in gait parameters, a single session of step practice with resistance did not consistently influence selected gait parameters, possibly due to limited practice.

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Poster

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Topic: F.06. Posture and Gait

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Title: Aging and cognitive-motor challenge differentially modulate cortical motor contributions to standing balance control

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Abstract: Older adults (OA) engage more cortical resources than younger adults (YA) to control balance, which may contribute to falls. Competing cognitive demands may prevent cortical resource allocation towards balance, yet how cortical engagement is modulated during balance is unclear. We hypothesized that difficult balance tasks engage motor cortical substrates that are shared during difficult cognitive tasks. Transcranial magnetic stimulation (TMS) can index motor cortical engagement by measuring cortical spinal excitability (CSE), which is not often assessed in OA during cognitive-balance challenge. We used TMS during standing cognitive-balance dual tasks to measure CSE as a function of balance and cognitive challenge in able-bodied YA and OA. We predicted CSE would increase with balance challenge, but cognitive and balance performance would decrease, particularly in OA, due to increased competition for cortical resources. Data were collected in 18 YA (18-35 years) and 14 OA (60-85 years). TMS was delivered during seated, easy (quiet stance; QS) and difficult (narrow stance over foam; NS) standing balance tasks, performed with and without a difficult (2back) cognitive task. CSE was assessed as the peak-to-peak amplitude of 20 motor evoked potentials (MEPs) elicited by TMS pulses targeted to the soleus hotspot. Motor and cognitive task performance were assessed with center of pressure (COP) sway and n-back accuracy, respectively. Consistent with our predictions, our results suggest that OA may recruit more corticomotor resources during balance challenge. Compared to QS, MEPs increased during NS in OA ($\beta = -50.7 \pm 13.6$, $p < 0.001$) but not YA ($\beta = 23.2 \pm 12.2$, $p = 0.16$). Contrary to our predictions, in NS, MEPs decreased with cognitive challenge in YA ($\beta = 60.5 \pm 20.5$, $p = 0.0096$) but not OA ($\beta = -33.2 \pm 23.1$, $p = 0.39$). Behaviorally, we found no dual task differences in balance performance, indicating balance was maintained. However, cognitive dual task cost was reduced during QS compared to NS in OA ($p = 0.033$) but not YA ($p = 0.97$). Our results suggest cognitive resources may differentially modulate CSE across age and cognitive-balance challenge. We found YA flexibly shifted to automatic balance control by reducing cortical engagement during cognitive-balance challenge. On the contrary, OA maintained reliance on cortical motor resources and were unable to shift attention away from balance and engage an automatic balance strategy, as age impaired subcortical motor systems likely could not support balance alone. This work will inform the development of precision (p)rehabilitation strategies to maximize balance function and reduce falls.

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Poster

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Topic: F.06. Posture and Gait

Support: CHNF 1177074

Title: Task-specific gait adaptations in the Yucatan mini pig and the impact of SCI

Authors: *L. KONAN^{1,2,3}, W. OSTEEN⁵, M. BOCZEK⁶, C. OVERLEY⁷, R. ALREFAI⁶, S. DAVISON⁸, D. R. HOWLAND^{9,10,4};

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Abstract: Community ambulation requires more than regular, continuous stepping; it demands complex coordination to navigate uneven surfaces and environmental obstacles. Such challenges place greater demands on weight-bearing, balance, and the real-time adjustment of limb movements and foot placements. These advanced locomotor requirements remain underexplored in the Yucatan Mini Pig (YMP), an emerging large-animal model for spinal cord injury (SCI). Thus, our laboratory is developing and using a series of adaptive gait tasks to evaluate motor performance before and after SCI. These tasks are designed to test key features of gait during different task challenges. These include modulation of hindlimb (HL) trajectories, hoof placement precision, weight support capacity, posture, and balance control. Standard spatiotemporal features of each task also are determined along with scoring on the Porcine Thoracic Injury Behavioral Scale (PTIBS). In the current study, eight neurologically intact female YMPs were conditioned to perform a range of locomotor tasks, including level surface (basic) overground walking, traversing a horizontal agility ladder, a step-up/step-down maneuver, and obstacle negotiation. HL gait data were captured using a marker-based 3D motion capture system in the neurologically intact YMPs and in a subset of three animals that received low thoracic lateral hemisections (Hx) across an 8-week recovery period. Hx injuries are used as they permit assessment of skilled forms of gait and enhance interpretation of circuitry contributions. Our findings indicate that the neurologically intact YMP smoothly modulates its HL trajectory according to the task height, and significant changes in efficiency are seen with increasing height challenges. Following Hx, the ability to negotiate height challenges is disrupted and weight acceptance occurs on a significantly more flexed limb. Neither fully recover. Compared to basic walking, navigating the agility ladder shows significant reductions in self-selected walking speed along with adjustment of stride length to meet rung spacing normally and after Hx. Similarly, hoof placement accuracy and consistency decrease. In contrast to no HL preference in the intact YMP, the ipsilateral HL becomes the preferred leading limb during obstacle negotiation and step-up/step-down tasks post-Hx. These findings demonstrate that supraspinal circuitry influences HL gait functions in the YMP and that these contributions along with sub-lesional circuitry likely reorganize to support partial recovery. Anatomical tract tracing studies of the descending pathways are ongoing along with continued gait data assessments.

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Poster

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Topic: F.06. Posture and Gait

Support: NIH/NIA 1R01AG07553201A1
DoD OPORP OP180014

Title: Postural modeling toward understanding imbalance in aging

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Abstract: Understanding sensorimotor control of posture is critical for addressing balance issues and fall risks in older adults, and postural modeling offers a valuable tool to investigate underlying sensorimotor control mechanisms. Postural models often approximate the human body as a single-link inverted pendulum, because these models are sufficiently simple to study sensory function and neuromuscular control. Some studies have secured participants to a backboard during data collection to enforce the single-link assumption; however, similar results are achieved with and without a backboard in young participants [1]. To the authors' knowledge, this has not been validated in older adults, which is relevant because prior literature [2] shows that older adults have greater differences between lower and upper body sway than younger adults. This study investigates the effects of multi-segment body motion on the ability to fit single-link models to experimental body sway in older adults. We hypothesize that postural control in older adults can be described using a single-link model, which would be supported if 1) measures of model fit in older adults are similar to those of younger adults and 2) there is not a strong correlation between measures of multi-segment body sway and model fit. Our work leverages data collected from 6 older adults (63-72, 67.3±3.1 yrs) who provided informed consent and completed several variations of a perturbed posture task. Upper and lower body sway were measured, and center of mass (CoM) sway was estimated from body segments. To characterize multi-segment body motion, we use the mean and standard deviation of the difference between lower and upper body position. A single-link postural model (input: perturbation stimulus, output: body sway) was optimized to reduce the error between experimental and predicted CoM sway, using 7 free parameters. Mean squared error was computed as a measure of model fit. Preliminary results show that single-link models are well-fitted to experimental data in the older adults. Correlations between measures of model fit and multi-segment body motion were not significant in most conditions. These preliminary results suggest that single-link models can be used to describe posture in older adults and are minimally affected by multi-segment body sway. Overall, these findings will support upcoming work investigating the contributors to imbalance and fall risk in older adults and contribute to the continued development of improved postural models. [1] Peterka. J Neurophysiol, 2002. 88(3):1097-1118. [2] Wiesmeier, et al. Front Aging Neurosci, 2015. 7:97.

Disclosures: R. Bellisle: None. A. Goodworth: None. K. Love: None. F. Karmali: None.

Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR285.16/V8

Topic: F.06. Posture and Gait

Title: Development and Decline of Dynamic Postural Control: Age, Sex, and Anthropometric Influences on Limits of Stability

Authors: *N. M. A. ALSALMIE¹, V. ROBLES-CERDAS¹, M. MEYER VEGA¹, D. J. GOBLE², N. SHAH¹, H. S. BAWEJA¹;

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Abstract: Limits of Stability (LOS) are a fundamental measure of dynamic postural control during standing, reflecting the maximum distance an individual can intentionally lean in any direction without losing balance or stepping. Reduced LOS is a known risk factor for falls and is commonly used in clinical and research settings to assess balance ability. While it is reasonable to assume that anthropometric factors such as height, weight, and age influence LOS, their specific impact remains unclear. This study aims to quantify how these anthropometric characteristics affect LOS and to examine how LOS changes across the lifespan. A total of 1559 healthy participants (881 female and 678 male) volunteered to participate in the study. Participants were grouped into eight age-groups - 5-12, 13-17, 18-25, 26-35, 36-44, 45-55, 56-65, and 66+ years. LOS was tested while standing on a force plate. Participants moved around as far as possible in all directions without lifting their feet for 60 seconds. LOS develops and changes across the lifespan ($F=56.5$, $p<.001$) and is significantly different between males and females ($F=142.23$, $p<.001$). We found that LOS areas increase from ages 5-12 (355 ± 102 cm²), stabilize from 13-44 years (459 ± 101 cm²), and then decline starting at the age of 45 (412 ± 110 cm²) and continue to decline beyond 60 years of age. Shoe sizes were weak predictors of LOS in males ($F=92.17$, $p<.001$, $R^2=.135$), while height a weak predictor of LOS in females ($F=32.11$, $p<.001$, $R^2=.038$). These findings indicate a weak sex-specific contribution of anthropometrics to LOS across the lifespan with no consistent overall determinant. LOS increases through childhood and adolescence, peaks in early adulthood, and begins to decline around age 45 - with further decline observed beyond age 60. Our findings demonstrate that Limits of Stability vary significantly across the lifespan and are partially influenced by sex-specific anthropometric factors. These results offer important normative reference values and emphasize the need to consider age and sex when assessing dynamic postural control in clinical and research settings.

Disclosures: N.M.A. Alsalmie: None. V. Robles-Cerdas: None. M. Meyer Vega: None. D.J. Goble: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Eligible for royalties from a pending patent (OMB 0651-0032) related to the technology used in this study. In addition, he has an equity stake (stock options) in Balance Tracking Systems. N. Shah: None. H.S. Baweja: None.

Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Program #/Poster #: PSTR285.17/V9

Topic: F.06. Posture and Gait

Support: NIH Grant R01HD100416
NSF Grant 1937968

Title: Heteronymous modulation of reflex excitability may not contribute to post-stroke stiff-knee gait

Authors: *J. S. CORREA^{1,2}, R. SIU¹, D. LORENZ², D. A. CUNNINGHAM^{2,1}, J. S. SULZER^{2,1};

¹Biomed. Engin., Case Western Reserve Univ., Cleveland, OH; ²Physical Med. and Rehabil., Case Western Reserve Univ. and The MetroHealth Med. Syst., Cleveland, OH

Abstract: Stiff-Knee gait (SKG) is a post-stroke impairment defined by reduced knee flexion linked to excessive quadriceps activity during pre-swing. Clinical treatments for SKG have shown variable efficacy, highlighting the need for understanding the underlying neural mechanisms. Prior studies have shown heteronymous reflexes produce increased facilitation after stroke in static conditions. It remains unclear whether such pathways contribute to excessive muscle activity in dynamic settings. This study aimed to investigate the role of heteronymous reflexes during gait in people with post-stroke SKG. We hypothesized individuals with SKG would exhibit increased facilitation during pre-swing. Eighteen individuals with chronic post-stroke SKG and 10 neurologically intact (NI) individuals participated in this study. The soleus H-reflex served as a model to measure the modulation of reflex excitability by two distinct heteronymous pathways: the vastus lateralis (VL) and tibialis anterior (TA). Peripheral nerve stimulation parameters used to innervate VL and TA afferents were optimized in the seated position. Participants then walked on a treadmill at self-selected speeds while reflex modulation was measured during pre-swing and loading phases. Modulation was expressed as the percent change of the soleus H-reflex amplitude of conditioned trials to unconditioned control trials. In a separate session, rectus femoris (RF) EMG and peak knee flexion were recorded during treadmill walking. Pre-swing RF activity was quantified by normalizing integrated EMG from pre-swing to loading. In individuals with post-stroke SKG, soleus reflex excitability was facilitated by VL input when seated ($H=144 \pm 63\%$) but showed no modulation at pre-swing ($H=96 \pm 12\%$) or loading ($H=100 \pm 10\%$). TA input inhibited reflexes when seated ($H=68 \pm 22\%$) and similarly showed no modulation at pre-swing ($H=84 \pm 38\%$) or loading ($H=88 \pm 22\%$). In NI participants, TA modulation did not yield any change during loading ($H=110 \pm 50\%$), contrasting the inhibition observed when seated ($H=62 \pm 25\%$). In individuals with post-stroke SKG, there were no correlations between reflex modulation and RF activity or knee flexion. This study aimed to understand the role of heteronymous reflexes in post-stroke SKG. Results showed VL and TA modulation of soleus reflex excitability was task dependent but not related to quadriceps muscle activity or knee flexion during gait. These findings suggest that while heteronymous reflexes

may play a role in post-stroke SKG, it is limited. Future work should examine if the role of reflex modulation varies across subgroups of SKG defined by impairment severity.

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Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

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Title: Foot-ground force quantifies impaired balance control mechanisms post-stroke

Authors: *K. SHIOZAWA¹, R. SUGIMOTO DIMITROVA², K. G. GRUBEN⁴, N. HOGAN³;
¹Mechanical Engin., ³Mechanical Engin. & Brain and Cognitive Sci., ²MIT, Cambridge, MA;
⁴Kinesiology & Biomed. Engin., Univ. Wisconsin, Madison, WI

Abstract: Stroke can have detrimental effects on the ability to maintain postural balance, which can impede mobility and negatively impact overall quality of life. Although there has been considerable effort to study post-stroke balance, the specific changes of the underlying control mechanisms following stroke remain unclear. This work sought to use foot-ground force data to quantify the apparent quiet-standing balance controller of stroke survivors compared to that of similarly-aged unimpaired participants. A previous study showed that the intersection-point-height measure, which captures frequency-domain foot-ground force behavior, was able to differentiate quiet-standing data from paretic and non-paretic limbs of 12 post-stroke participants and those of 22 similarly-aged unimpaired participants. In this study, the same data set was used to identify a descriptive model, in the form of a double-inverted pendulum controlled via full-state feedback, that reproduced the intersection-point-height data for each participant group. The best-fit models revealed a distinct joint-torque-coordination pattern in the paretic limb of post-stroke participants, which differed substantially from that of the similarly-aged participants. The post-stroke participants also showed greater reliance on neural control. Identifying joint coordination patterns associated with impairment, quantifying their severity, and distinguishing them from compensatory strategies in this way can offer valuable guidance for designing targeted rehabilitation interventions and monitoring balance recovery.

Disclosures: **K. Shiozawa:** None. **R. Sugimoto Dimitrova:** None. **K.G. Gruben:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent holder. **N. Hogan:** None.

Poster

PSTR285: Posture and Gait: Influence of Aging, Injury, and Disease

Location: SDCC Halls B-H

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Program #/Poster #: PSTR285.19/V11

Topic: F.06. Posture and Gait

Support: Major Basic Research Fund of Jiangsu Province Hospital (TS202402)

Title: Activation of hypothalamic-pontine-spinal pathway promotes locomotor initiation and functional recovery after spinal cord injury in mice

Authors: *C. Ji¹, Y. Li²;

¹Chinese Acad. of Sci. Ctr. for Excellence in Brain Sci. and Intelligence Technol., Nanjing, China; ²Neurol., Boston Children's Hosp., Boston, MA

Abstract: The hypothalamus is critical for regulating behaviors essential for survival and locomotion, but how it integrates internal needs and transmits locomotion commands to the spinal cord (SC) remains unclear. We found that glutamatergic neurons in lateral hypothalamic area (LHA) are essential for regulating motivated locomotor activity. Using single-neuron projectome analysis, trans-synaptic tracing, and optogenetic manipulation, we showed that LHA facilitates motivated locomotion during food seeking via pontine reticular nucleus, oral part (PnO) projection neurons, rather than direct SC projections or indirect stress signaling via medial septum and diagonal band. Activating PnO-SC projection neurons also initiated locomotion. Importantly, LHA-PnO projection neurons were crucial for regulating locomotor recovery following mouse spinal cord injury (SCI). Motor cortex signals gated LHA deep brain stimulation treatment markedly promoted long-term restoration of hindlimb motor functions after severe SCI. Thus, we have identified a hypothalamic-pontine-spinal pathway and the stimulation paradigm for potential therapeutic intervention after SCI.

Disclosures: **C. Ji:** None. **Y. Li:** None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.01/W1

Topic: G.01. Neuroethology

Title: Decoding handwriting trajectories from electrocorticogram (ECoG)

Authors: *X. WANG^{1,3}, W. YANG¹, C. ZHENG¹, H. CUI²;

²Lab. of Neural Mechanism of Motor Control, ¹Chinese Inst. for Brain Research, Beijing(CIBR), Beijing, China; ³Chinese Acad. of Med. Sci. & Peking Union Med. Col., Beijing, China

Abstract: Handwriting, a complex sequence of highly dexterous movements, is ubiquitous in human life. However, the neural basis of fine motor control involved in handwriting remains largely unexplored. In the present study, we intraoperatively recorded electrocorticogram (ECoG) signals from eight participants using 20 to 32 channels temporarily placed on the frontoparietal cortex for tumor resection. Additionally, we recorded ECoG signals from three participants with 36 to 64 electrodes implanted in the temporal lobe and prefrontal cortex for epilepsy monitoring over three days. During the ECoG recording, participants were instructed to write the digits from 0 to 9 using their index finger while a GoPro recorded their hand movements. We employed multi-taper and wavelet transforms for feature extraction, support vector machines (SVM) and decision trees for classification, and a single-layer recurrent neural network (RNN) to reconstruct finger trajectories. We observed event-related desynchronization and synchronization (ERD/ERS) rhythms in all participants during digit writing. The decoding algorithms were able to predict the digits of the hand movement significantly above the chance level. We found that electrodes in the sensorimotor area (primary motor cortex [M1] and dorsal premotor cortex [PMd]) contributed most significantly to decoding, with the strongest signals primarily from the high gamma (80-160 Hz) and beta (13-30 Hz) frequency bands. Participants demonstrated heterogeneous spatiotemporal dynamics during digit writing. The decoding algorithms could predict the type of movement for four participants even at the preparation phase before movement onset. The decoding results varied depending on the temporal structure of the participants' movements while writing digits. We reconstructed finger trajectories for digits 0 to 9 and calculated Pearson correlation coefficients between the actual and reconstructed trajectories. In one awake intraoperative case, we achieved Pearson correlation coefficients of 0.39 ± 0.14 for the X-axis and 0.42 ± 0.12 for the Y-axis. In one participant with implanted electrodes, the correlation coefficients reached 0.46 ± 0.06 for the X-axis and 0.48 ± 0.08 for the Y-axis. Above findings provide new insights into the neural control mechanisms underlying fine motor behavior in humans.

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Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.02/W2

Topic: G.01. Neuroethology

Support: The J. Scott McFadyen Fund for Excellence in Parkinson's Disease Research

Title: Assessing rare-earth materials' magnetic resonance response for use as fiducials in rat brain MRI

Authors: *Y. DONG¹, T. CHALLENGER², O. V. FAVOROV¹, R. MURROW²;

¹Biomed. Engin., Univ. of North Carolina Chapel Hill, Chapel Hill, NC; ²Neurol., Univ. of North Carolina, Chapel Hill, NC

Abstract: Technological advances in recording electrophysiological activity from large numbers of neurons at single-cell resolution have opened new opportunities to investigate the brain mechanisms underlying purposeful animal behaviors. To gather such data, multichannel recording probes must be chronically implanted in specific divisions of the forebrain, which in rats requires submillimeter precision. Stereotaxic methods, which rely on coordinates relative to the bregma, are commonly used to place electrode probes but are limited in precision due to individual variations in forebrain anatomy. In principle, high-resolution MRI scans can localize an individual's forebrain structures and use skull-embedded fiducial markers to guide recording probe insertion. However, commonly used fiducial markers, such as fiducial screws filled with 2% copper-sulfate or Gd-DTPA solution, have a precision of 1-2mm. In rodents, forebrain targets are small, and higher precision is necessary. Combining stereotaxy with MRI guidance requires high-precision fiducials in order to register where electrodes should be implanted. As rare-earth elements are weakly paramagnetic, REE-containing crystals might be highly visible on MR images and suitable as fiducials. We explored several such crystals, including Gadolinium Aluminum Gallium Garnet (Ce:GAGG), Europium Fluorite (Eu:CaF₂), Terbium Gallium Garnet (TGG), Erbium-doped lithium niobate (Er:LN), and opal impregnated with gadolinium contrast agents. These were selected for fluorescence as well as rare-earth content in order to increase visibility of small markers. We also explored paramagnetic silver and tungsten wires. All these materials proved visible in the MRI. However, under both T1 and T2, they produced black distortions without any bright spots, making them highly visible in fatty tissues but virtually invisible in the skull itself. The Gd-DTPA opals were an exception; however, the contrast agent escaped the opal matrix quickly, and sealing the surface proved nonviable. While labeling the skull with these markers directly did not prove workable, tungsten wires anchored to the skull could be detected if they were extended into the overlaying skin.

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Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.03/W3

Topic: G.01. Neuroethology

Title: Topological architecture of the brain-spinal cord communication

Authors: *L. CHEN^{1,2}, Q. DENG^{1,3}, X. DU^{1,3}, J. DU^{1,2,3};

¹Inst. of Neuroscience, Chinese Acad. of Sci., Shanghai, China; ²ShanghaiTech Univ., Shanghai, China; ³Univ. of Chinese Acad. of Sci., Beijing, China

Abstract: The communication between the brain and spinal cord (SC) is crucial for integrating external signals and generating behavior. However, the brain and SC differ significantly in shape and neuronal organization principles, making it critical to unravel the wiring logic underlying brain-SC communication. Here, we systematically investigate the topographic organization of brain-SC projections using single-neuron reconstructions in zebrafish larvae. Brain-to-SC projections follow a horizontally rotated and axially distributed pattern, while SC-to-brain projections exhibit a horizontally conserved and axially condensed principle, both involving excitatory and inhibitory neurons associated with axial elongation. Together, they constitute a topological architecture of brain-SC communication, with parallel sensory and motor pathways, corresponding to their information flow. This architecture is further complemented by widespread neuromodulatory projections from the brain to the SC, and integrate diverse sensory signals in a modality-specific manner. Together, our findings reveal a comprehensive framework for understanding of brain-SC communication, highlighting differential information transmission, and offering new insights into its functional organization and inter-regional interactions.

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Poster

PSTR286: Sensory Motor Systems

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Program #/Poster #: PSTR286.04/W4

Topic: G.01. Neuroethology

Support: Alfred P Sloan Foundation
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Title: Recent breed formation has altered white matter organization in domestic dogs

Authors: *S. BARTON;
Harvard Univ., Cambridge, MA

Abstract: Contrary to popular belief, most Western dog breeds have recent origins. These modern breeds have rapidly evolved neural, cognitive, and behavioral traits that distinguish them from premodern dogs, which include ancient breeds, free-roaming village dogs, and New Guinea singing dogs. Notably, modern breed dogs show widespread expansion of gray matter volume in the cerebral cortex. By comparison, premodern dogs have larger subcortical structures. However, it remains unknown as to whether there are also significant differences in white matter

organization between modern breed and premodern dogs. Using tract-based spatial statistics and a whole brain probabilistic tractography analysis, we compared fractional anisotropy and connectivity networks between 60 modern breed and 24 premodern dogs. We report differences in patterns of white matter organization in these two canine lineages, extending previous gray matter morphometry results. Our findings highlight the remarkable extent to which recent and intensive artificial selection has remodeled the canine brain. We discuss the relevance of these results to our understanding of neural changes during domestication, as well as to the more specific, open question of human self-domestication.

Disclosures: S. Barton: None.

Poster

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Program #/Poster #: PSTR286.05/W5

Topic: G.01. Neuroethology

Title: Eliciting a sequence of orienting behaviors by optogenetic manipulation in an echolocating Bat

Authors: *G. SOMASEKHAR¹, E. MARSHALL¹, S. CADIGAN¹, W. NGUYEN², K. KASPER¹, V. NARAYANAN¹, P. GUTRUF³, M. J. WOHLGEMUTH²;

²Neurosci., ¹Univ. of Arizona, Tucson, AZ; ³The Univ. of Arizona, Tucson, AZ

Abstract: Spatial localization recruits a sequence of motor behaviors to orient the animal towards a target of interest. Humans use vision for active sensing, and we frequently articulate our eyes, head, and body when orienting towards an object of interest. To study how the brain sequences behaviors for spatial orientation, we conducted studies on the echolocating bat. The bat produces sonar calls and moves its head and ears when orienting towards an object of interest. We focused on the mammalian midbrain superior colliculus (SC) for our work because the SC has been shown to play a role in species-specific spatial orientation, including visuomotor behaviors in humans, and sonar orienting in bats. We developed a wireless optogenetic manipulation technique to initiate a sequence of motor behaviors to orient an unrestrained bat towards egocentric coordinates. We used a light-activated virus to stimulate the excitatory neurons in the SC, followed by the implantation of a custom, wireless optogenetic device. The microLED on the wireless optogenetic device was experimenter-controlled, allowing modulation of stimulation parameters such as duration, duty cycle and frequency. Our results show a difference in the latency of response to spatial orientation with head movement beginning almost instantaneously post stimulation, followed by ear movements and then vocalizations. This methodology establishes the groundwork for studying orienting behavior in free-moving, unrestrained animals. Future work will couple circuit-level optogenetic perturbations with multichannel electrophysiology to examine how sensory information influences motor planning in a behaving animal.

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Poster

PSTR286: Sensory Motor Systems

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Topic: G.01. Neuroethology

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NSF EFRI-BRAID 22223822
NIH R01DC018446
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Title: Species Differences in Real-Time Sensorimotor Error Processing During Vocalization

Authors: *L. L. STANWICKS¹, L. M. OSTROWSKI², M. TODESCO³, A. SIDDIQUI³, T. MCPHERSON², J. C. GORMAN⁴, M. MCPHERSON³, E. M. ARNEODO⁵, T. GENTNER⁶; ¹UCSD NGP, SAN DIEGO, CA; ²Dept. of Neurosciences, UCSD, La Jolla, CA; ³UCSD, San Diego, CA; ⁴Neurosci., UC San Diego, La Jolla, CA; ⁵Psychology, ⁶Psychology, Univ. Of California San Diego, La Jolla, CA

Abstract: Control theoretic models of motor control posit that behavioral goals generate internal expectations (forward models) of the sensory feedback associated with kinematic actions, and use deviations between expected and actual sensory input to adjust motor actions. For learned, sequentially complex vocal communication signals that unfold across time in flexible ways, such as speech and some bird songs, this integration process must be nearly continuous. How the brain integrates continuous sensory feedback to drive adaptive changes in vocal motor output remains unknown. We are investigating sensorimotor feedback in two songbird species with differing vocal flexibility: zebra finches (*Taeniopygia guttata*), which produce highly stereotyped song, and European starlings (*Sturnus vulgaris*), which produce variable, dynamic vocal sequences modified throughout their lives. We hypothesize that differential sensorimotor feedback during singing underlies these species differences in vocal flexibility. Thus, modulating auditory feedback of a bird's own-song should alter vocalizations differentially for each species in real-time. Specifically, starlings' songs should be more disrupted than zebra finches', reflecting greater reliance on real-time vocal error correction. To test this, we recorded songs in ambient air and heliox (80% helium, 20% oxygen), which shifts vocal tract resonance without altering motor commands. Song recordings were collected from 3 zebra finches (482 regular air, 260 heliox) and 6 starlings (1892 regular air, 806 heliox). We analyzed the vocalizations using unsupervised embedding and assessed whether heliox altered song structure at three levels: acoustic (physical properties, e.g. fundamental frequency), phonetic (production-related features, e.g. duration), and syntactic (sequence structure e.g. syllable order). As expected, heliox altered acoustic properties in both species. In starlings, however, acoustic shifts were accompanied by

both phonetic and syntactic changes, while zebra finches showed minimal phonetic and/or syntactic changes to their songs in helioX. These results support functionally distinct and species-specific roles for sensorimotor integration, consistent with our hypothesis that sensory feedback-driven error processing functions in vocal-motor flexibility.

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Poster

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Topic: G.01. Neuroethology

Support: MEXT/JSPS KAKENHI Grant Numbers JP23H05428, JP24H00912, JP24KJ0886

Title: Development of auditory-vocal mirror neurons in the basal ganglia (Area X) of Bengalese finches

Authors: *Y. SUZUKI^{1,2}, H. KODA¹, K. OKANOYA^{2,1}, S. YANAGIHARA²;

¹Grad. Sch. of Arts and Sci., The Univ. of Tokyo, Tokyo, Japan; ²Advanced Comprehensive Res. Organization, Teikyo Univ., Tokyo, Japan

Abstract: Imitation learning requires associating sensory inputs with motor commands necessary to reproduce observed behaviors. Mirror neurons, which fire both when an action is produced and when the same action is perceived, have been proposed to support imitation learning. To test the hypothesis, it is necessary to record neural activity throughout the entire course of imitation learning and identify how mirror neuron properties change as learning progresses. However, such developmental investigations have been technically challenging in mammals. In contrast, songbirds learn songs by imitating their tutors as they develop, which enables us to examine how mirror neuron activity changes across development at the single-neuron level. Within the cortico-basal ganglia circuitry of songbirds, which is crucial for song learning, the premotor cortical nucleus HVC contains neurons that fire during both song playback and singing. These “auditory-vocal mirror neurons” project to the basal ganglia nucleus Area X and may contribute to vocal imitation learning in songbirds. However, whether similar auditory-vocal mirror neurons exist in Area X and how their properties change during development remain unclear. To address these questions, we first conducted extracellular single-unit recording in Area X of adult Bengalese finches during both singing and passive listening to songs. We found neurons that fired at specific syllable timing when the birds were singing and when the birds were listening to their own songs, confirming the presence of auditory-vocal mirror neurons in Area X. If these neurons are involved in vocal imitation, their neural properties

should change as imitation learning progresses in juveniles. To examine how these mirror neurons develop, we next recorded the neural activities of juvenile Bengalese finches across the sensorimotor learning period. We found auditory-vocal mirror neurons in Area X of juveniles, but the auditory response properties changed across development. In the early stage of song learning, many of the neurons responded exclusively to tutors' songs without responding to birds' own songs. As learning progressed, more neurons responded to the birds' own songs, and some neurons no longer responded to their tutors' songs. These findings suggest that auditory-vocal mirror neurons initially encode the auditory information of the tutors' songs and gradually shift to represent learned vocal output. We propose that auditory-vocal mirror neurons may contribute to vocal imitation by flexibly linking sensory input with vocal output across developmental stages.

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Poster

PSTR286: Sensory Motor Systems

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Title: Social familiarity strengthens neural and vocal responses to conspecific calls in zebra finches

Authors: *C. GOMEZ-GUZMAN¹, J. BENICHOV², D. VALLENTIN³;

¹Vallentin, Max Planck Inst. for Biol. Intelligence, Starnberg, Germany; ²Max Planck Inst. Seewiesen, Seewiesen, Germany; ³MPI for Biol. Intelligence, Seewiesen, Germany

Abstract: Across animals, dyadic vocal interactions often occur within complex acoustic environments containing numerous signalers. The influence of socially relevant acoustic signals on the neural circuits controlling interactive vocal behavior remains poorly understood. We address this question in zebra finches, highly social songbirds that maintain nearly continuous vocal contact through the exchange of short innate calls. We developed a behavioral paradigm that elicits differential responses to familiar and unfamiliar vocal partners, enabling the prediction of social context based on individual birds' response patterns. We then used high-density Neuropixels probes to record neural activity within a vocal premotor nucleus in the songbird forebrain, while birds listened to familiar and unfamiliar contact calls. We found that the activity of putative projection neurons and interneurons in this vocal premotor nucleus was modulated by the familiarity of heard calls, with interneurons exhibiting stronger responses to

familiar calls. Furthermore, we found that measures of vocal responsiveness correlated with neural response parameters during listening. Specifically, we observed that increased vocal response rates, rapidity, and temporal consistency for familiar call playbacks were correlated with elevated mean and peak firing rates, as well as prolonged activity, in HVC interneurons. These results demonstrate how socially salient auditory information can affect a forebrain premotor circuit to maintain the specificity of vocal interactions within complex and dynamic social environments.

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Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.09/W9

Topic: G.01. Neuroethology

Support: JSPS KAKENHI Grant 24KJ1956

Title: Pigeons use both frontal and lateral vision for targets at different locations during sequential pecking

Authors: *K. WADA, H. KAWABATA;
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Abstract: Foraging behaviour consists of the reach-to-grasp toward feeding targets successfully in many species with different body anatomies. Unlike primates or mantises, whose eyes and arms are separated, birds experience unstable visual inputs during the reach because their eyes and bills share the same head. Previous observations pointed out that fixation in birds, or a brief stop phase before starting their reach, stabilized the retinal image to make it easier to plan their pecking. Many birds have two high-acuity retinal regions: the fovea centralis, which receives input from the farther lateral field, and the area dorsalis from the nearer frontal field. Frontal vision has been implicated in single-target pecks, yet ground-feeding often involves sequential pecking at targets scattered on their lateral sides. This study with five pigeons was conducted to investigate their use of each vision during sequential pecking. Based on the hypothesis that pigeons relied on their frontal vision even in sequential pecking, we predicted that pigeons would primarily peck the target in front of them during the sequence. Pigeons pecked 54 pieces of beans arrayed on the tray with 1.75 cm spacing as a daily session. Each session was video-recorded for 3D reconstruction of the geometric relation between their eyes and targets. In each peck toward the initial or following target during the sequence, fixating duration and visual distance to the target were extracted. Their transition to the next target during each sequence was logged. We compared the extracted data with our previous observation of single pecking. Pigeons fixated 30 % longer on the initial target in sequential pecking (around 150 ms) than on the single target (around 120 ms). In addition, pigeons fixated on the following targets 25 %

closer (around 75 mm) than on the initial or single target (around 100 mm). When many beans remained, pigeons fixated on nearer targets with their frontal vision. As the tray emptied, they fixated on farther targets with their lateral vision. These results suggested they already planned their pecks toward following pecks at the beginning of the sequence, and they used their frontal and lateral vision more flexibly depending on the density of feeding targets.

Disclosures: K. Wada: None. H. Kawabata: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.10/W10

Topic: G.01. Neuroethology

Support: Field-Spicer Fellowship in Research

Title: Environmentally relevant exposure to bisphenol-S in the perinatal and juvenile period impacts morphology and reproductive behavior of rats

Authors: A. ALMOND¹, A. GANDHI¹, X. GUIMBA¹, N. JAMES², S. MOBERLY¹, K. MULLIN¹, A. TAYLOR¹, *G. M. LANGE¹;

¹Saginaw Valley State Univ., University Center, MI; ²Saginaw Valley State Univ., Saginaw, MI

Abstract: In classic work by William C. Young's research laboratory, it was established that organization of mammalian brain morphology is guided by gonadally expressed hormones both in-utero and continuing into the perinatal environment. This suggests, while the genetic sex of an organism may drive development of sexual morphology and the brain, the environment can and does also shape phenotypic expression. Bisphenol-S is a chemical agent found in many plastics used in everyday life. Use of bisphenol-S has increased in recent years in comparison to the primary bisphenol used in industry, bisphenol-A, due to concerns about known environmental and physiological effects of bisphenol-A exposure. Bisphenol-S can and does leach from plastics and is inadvertently ingested by organisms. Bisphenol-S has been identified as an endocrine disruptor and has the potential to alter the neuroendocrine system of developing organisms. We theorize that perinatal and juvenile exposure to bisphenol-S may influence phenotype by reshaping organization of the mammalian central nervous system during perinatal and juvenile development altering expression of morphology and reproductive behavior. In this research, exposure to bisphenol-S occurred at environmentally relevant levels during post parturition days 1-21. At maturity, neuromuscular development was assessed and neurobehavioral responses related to anxiety were examined. Additionally, the presence and timings of reproductive behaviors were measured. We have identified ways in which bisphenol-S alters development in ways that are different from that of bisphenol-A exposure.

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Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.11/W11

Topic: G.01. Neuroethology

Title: Rats learn and use intuitive physics knowledge to solve fetch tasks

Authors: *J. YAN, R. GATTONI, A. KEMTUR, J. TALBOT, R. OTHAYOTH, M. LINDSAY, S. SAWTELLE, S. C. TURAGA, R. JOHNSON;
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Abstract: Humans have a rich capacity to understand and predict how objects in 3D scenes are likely to move and interact over time. This cognitive ability, called intuitive physics, allows humans to mentally simulate how their local environment is likely to change over time and to plan interactions with the world around them. Whether and how other animal species, e.g. rodents, learn and use intuitive physics knowledge is poorly understood. To explore this knowledge gap, we train rats to play fetch in large reconfigurable environments and challenge them to predict interception routes to earn rewards. Fetch is easy to learn, but has open-ended complexity, allowing us to systematically probe rodent understanding of intuitive physics concepts like momentum, occlusions, collisions, containment, and stability in a freely-moving cognitive task. To maximize task engagement and experiment control, we use collaborative robotic arms (cobots) to procedurally generate fetch trials across months of gameplay. To measure fine postural dynamics and animal-object interactions across learning, we use high-speed cameras and detailed 3D pose estimation to reconstruct all observed behavioral data in a digital twin of our experimental environment. Finally, we use imitation and reinforcement learning to train a virtual rodent to move and solve problems in the physics simulator MuJoCo like real rats do in our experiments, allowing us to model rodent decision making and locomotion using embodied agents with increasingly realistic biomechanical constraints. Our data reveals that in simple scenarios (e.g. a rolling ball with no obstacles), rats gradually improve task efficiency and develop a consistent interception strategy. In more complex variations (e.g. dropping a ball into a container), rats exhibit few-shot learning of an intuitive physics concept that generalizes to new scenarios. Overall, this project lays early groundwork to measure and model how rodents learn and use intuitive physical reasoning to solve problems in dynamic scenes.

Disclosures: J. Yan: None. R. Gattoni: None. A. Kemtur: None. J. Talbot: None. R. Othayoth: None. M. Lindsay: None. S. Sawtelle: None. S.C. Turaga: None. R. Johnson: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.12/W12

Topic: G.01. Neuroethology

Title: Influence of wheel running on home cage activity levels

Authors: ***R. P. WATERS**¹, I. VANDERHART², I. BURCH², S. BRONSKI²;
²Biol., ¹Univ. of Mary Washington, Fredericksburg, VA

Abstract: Exercise has potent beneficial effects on physical and mental well-being. As such, exercise serves as a popular model to explore multiple scientific fields, including physiology, neurobiology, and psychology. In the laboratory, voluntary wheel running is a primary exercise behavior in rodents and is used to approximate overall levels of physical activity. We housed outbred CD-1 female mice in social cages (4 per cage) with zero, one, or two running wheels (MedAssociates, Vermont) to determine voluntary utilization of the running wheel. We measured total home cage movement of mice using an RFID tracking system (Phenosys, GmbH, Berlin) to determine if wheel running is performed at the expense of home cage activity. We measured behavioral responses to the elevated plus maze and Lashley maze following 42 days in these environments, as exercise influences behavior in these paradigms. Mice housed with two wheels ran nearly twice the distance of mice with one wheel, suggesting that wheel running is limited when 4 mice are housed with a single wheel. We also observed increased memory performance and decreased anxiotypic behavior in mice housed with two wheel versus one wheel. We are currently analyzing RFID estimated home cage activity to determine if home cage activity is diminished by wheel running.

Disclosures: **R.P. Waters:** None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.13/W13

Topic: G.01. Neuroethology

Support: R01MH119250
C.M. Robertson Foundation
The Feil Foundation

Title: Dopamine dynamics in the nucleus accumbens during maternal behavior in postpartum and virgin CBA/CAJ mice

Authors: *K. R. DAY¹, S. D. SHEA²;

²Neurosci., ¹Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: Attentive maternal care is critical for the survival of altricial offspring and is regulated by a broad network of brain areas, including pathways that detect conventional rewards such as palatable food or drugs of abuse. In mice, a complex suite of caregiving behaviors is performed by both dams and virgins, including pup retrieval. In this study, we observed the dynamics of dopamine (DA) release during maternal behavior in nucleus accumbens (NAc) with fiber photometry and a fluorescent dopamine sensor (dLight1). We compared DA release and how it fluctuates over time and experience between postpartum CBA/CaJ dams, and age-matched virgin females (with no prior pup experience) with a focus on DA transients during pup retrieval. Mice underwent repeated multi-pup retrieval trials with multiple trials per day for 5 days. Latency to retrieve pups, as well as success rate (ability to place pups to the nest) were also quantified. We observe that both dams and virgin female CBA mice have a transient increase in NAc DA to pup retrieval that begins when the mouse contacts the pup with her mouth and slowly decays to baseline as the pup is transported to the nest. This DA response is consistent across multiple trials in a session as well as across multiple days. Interestingly, there is no such contact response to pup grooming, or mouthing the pup when it does not include a retrieval or transportation of the pup to another location. Notably, the CBA virgin female mice perform spontaneous pup retrieval without any prior personal maternal experience or tutoring from a dam, achieving both successful pup retrievals to the nest and latencies that improve across sessions/days. Considered together with our previous work (Xie et al., *Neuron* 111(4): 557), these findings suggest that NAc DA release acts as a positive reinforcer that motivates future performance of pup retrieval behavior by similar mechanisms in both dams and virgin females. Understanding the biological processes that initiate the onset and maintenance of caregiving actions like pup retrieval are important to advance our understanding of maternal motivation and maternal mental health.

Disclosures: K.R. Day: None. S.D. Shea: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

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Program #/Poster #: PSTR286.14/W14

Topic: G.01. Neuroethology

Support: CONAHCyT CF-2023-G-518

Title: Sex-specific effects of appetite suppressants on stereotypy in rats

Authors: *A. LOPEZ¹, R. GUTIERREZ²;

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Abstract: This study investigated the sex-specific effects of commonly prescribed appetite suppressants on body weight and the manifestation of motor side effects, specifically stereotypy.

Employing video recordings and DeepLabCut (DLC) for precise behavioral quantification, we analyzed stereotypy, defined as purposeless, repetitive motor behaviors, in male and female rats. Under control (saline) conditions, male rats exhibited a greater propensity for weight gain compared to females. However, in contrast, female rats demonstrated greater and more homogenous weight loss than males following the administration of diethylpropion and tesofensine. Phentermine and mazindol induced comparable weight loss in both sexes, whereas cathine elicited weight reduction exclusively in males. 5-HTP and d-amphetamine administration only prevented weight gain relative to controls. Analysis of motor side effects revealed that drugs primarily targeting dopamine pathways - specifically, phentermine, mazindol, diethylpropion, cathine, and d-amphetamine - induced pronounced stereotypies, particularly head-weaving, in both sexes. Interestingly, tesofensine elicited head-weaving behavior exclusively in female subjects, albeit to a lesser extent than that observed with other dopaminergic agents; conversely, tesofensine was most frequently associated with orolingual dyskinesia. Moreover, one of the most potent forms of stereotypy—backward locomotion, here referred to as "moonwalking"—was sporadically observed only following the administration of phentermine, diethylpropion, cathine, and mazindol, with diethylpropion inducing it most frequently. Male subjects treated with these same drugs exhibited an unexpected effect: spontaneous ejaculations, potentially attributable to the combined effects on dopamine and serotonin signaling in brain regions regulating sexual function. Network analysis and Markov transition matrices revealed distinct behavioral profiles associated with head-weaving, which emerged as the dominant attractor state, suggesting potential mechanistic differences among these drugs. Collectively, this study provides a valuable database characterizing the behavioral side effects of appetite suppressants.

Disclosures: A. Lopez: None. R. Gutierrez: None.

Poster

PSTR286: Sensory Motor Systems

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Topic: G.01. Neuroethology

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Title: Early song learning experiences regulate developmental dynamics of the auditory to motor circuit in zebra finches

Authors: *J. A. KOMOROWSKA-MULLER¹, S. NOMURA², Y. MOROHASHI¹, B. KUHN², Y. YAZAKI-SUGIYAMA¹;

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Abstract: Like human infants learn to speak, juvenile songbirds learn to sing by memorizing and then matching their vocalizations to their tutor's song (TS) during the developmental song learning critical period. In zebra finches, a commonly used songbird model, only males learn to sing an individually unique song and keep it for their entire lives. Recently, we found transient sensorimotor projections into the song premotor area, HVC, from the neurons activated by hearing TS in zebra finch higher auditory area, caudomedial nidopallium (NCM). In contrast to the dense NCM axonal projections observed in HVC in the juveniles in sensorimotor song learning period, significantly lower amount of NCM-HVC projections were found in the older juveniles at the end of song learning period. This suggests that NCM-HVC projections disappear during song crystallization (Louder et al., 2024). Here, we further examined the dynamics of NCM projections within HVC using longitudinal structural two-photon in vivo imaging and confirmed the disappearance of NCM-HVC projections at the end of song learning period. We then investigated whether song experiences and their timing affect the time course of NCM-HVC projection dynamics and song learning. We raised the juveniles with two tutors sequentially and thus made juveniles learn to sing from the first tutor (T1), and then from the second tutor (T2) after an isolation period. We found that the sequentially tutored juveniles learned songs from T2 beyond the normal learning period. Also, their songs were less stable than songs of normally reared birds at the end of normal sensorimotor learning period. We further visualized the NCM neurons responsive to each tutor's song in sequentially tutored birds by using a combination of viruses which express fluorescent proteins in an activity-dependent manner (AAV9-cFos-TetON-EGFP and AAV9-cFos-TetOFF-mRFP). The projections from NCM neurons responding to T2 song were found in HVC within 12 hrs after being exposed to the T2 in sequentially tutored juveniles. Notably, after sequential learning from two tutors, projections remained in HVC from both NCM neurons activated by hearing T1 or T2 songs in adulthood when NCM-HVC projections were rarely found in normally raised birds. Taken together, our results suggest that early song learning experiences and their timing shape auditory-motor neuronal circuits and learning time course.

Disclosures: J.A. Komorowska-Muller: None. S. Nomura: None. Y. Morohashi: None. B. Kuhn: None. Y. Yazaki-Sugiyama: None.

Poster

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Topic: G.01. Neuroethology

Support: NIH Grant 5R01DC014989

Title: Evidence of sensory-motor integration at the neuronal level across a network of auditory-motor areas in non-human primates

Authors: *S. BAUMANN¹, M. KIM¹, S. OGUNTAYO¹, P. A. WIKMAN², R. SAUNDERS³, J. P. RAUSCHECKER¹;

¹Dept. of Neurosci., Georgetown Univ., Washington, DC; ²Dept. of Psychology and Logopedic, Univ. of Helsinki, Helsinki, Finland; ³NIMH, McHenry, MD

Abstract: Auditory-motor integration is crucial for the development and maintenance of speech and music performance in humans but also for vocalizations and auditory-guided actions in other animals. In humans, defects in the auditory-motor system have been implicated in speech deficits such as various aphasia (e.g. after stroke) or stuttering. Evidence from neuroimaging in humans and non-human primates suggests auditory and motor information interacts in a network of auditory, motor, parietal and subcortical regions during auditory-motor activities (Baumann et al., 2007, Archakov et al., 2020). We developed a model system based on an auditory-motor task performed by rhesus macaques where we can study sensory-motor interactions in said network at a neuronal level. We recorded single units as well as local field potentials from four candidate areas of the non-human primate auditory-motor network: posterior auditory cortex, dorsal premotor cortex, posterior parietal cortex and the putamen in the basal ganglia using. From each of these areas, we recorded single-cell action potentials (AP) and local field potential (LFP) data using linear Neuropixels electrophysiology probes, while animals replayed presented tone sequences (piano task, Archakov et al., 2020). The high density of recording sites of these probes (4416 sites selectable from 384 channels) typically yields solid data from 50-100 uniquely identifiable neurons per recording session. We find frequent neurons responding to the piano sounds, piano key actions and juice rewards in all four recorded brain regions, both in separate neurons as well as in neurons that respond concurrently to two or all three main events of the auditory-motor task. These different responses tend to cluster at different depths in the cortical layer organization. The processing of sound and key actions in auditory-motor candidate areas provides evidence of information sharing across a sensory-motor network with auditory-motor neurons providing correlates for integration. The specific arrangement across cortical layers provides potential clues about and input processes.

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Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.17/Web Only

Topic: G.01. Neuroethology

Title: The effects of temperature and TRPA1 overexpression in *Drosophila melanogaster* behavior and physiology

Authors: *J. KIM¹, Y. KIM¹, J. NADOLSKI², R. L. COOPER³;

¹The Gatton Acad. of Mathematics & Sci., Richmond, KY; ²Benedictine Univ., Lisle, IL; ³Dept. of Biol., Univ. of Kentucky, Lexington, KY

Abstract: All cells are made of proteins, but abnormal expressions of proteins can lead to cellular malfunction, thus harming the organism. One type of protein, transient receptor potential ankyrin 1 (TRPA1), is a Ca²⁺-permeable ion channel involved in physiological roles of temperature sensing and mechanosensory transduction. TRPA1 is overexpressed in most cancerous cells, but the physiological mechanisms and pathological processes are not yet fully understood. Thus, this study investigated the effects of TRPA1 overexpression in various sensory neurons and muscle tissues on behavioral and physiological responses, specifically (1) mechanotransduction, (2) locomotion, (3) thermotaxis, and (4) membrane potential of *Drosophila melanogaster* larvae. First, for mechanotransduction and locomotion, head-abdomen-tail, body wall, and mouth hook assays were performed in 20°C and 33°C. For thermotaxis, larval crawling behavior was recorded on Petri dishes with IR imaging. Finally, resting membrane potential was recorded in muscles without and with TRPA1 overexpression. The hypothesis tested in this study was that TRPA1 overexpression will alter behavioral and physiological responses by heightening sensory responsiveness when thermally activated. The results indicated that the hypothesis was supported when TRPA1 was overexpressed in certain tissues. When overexpressed in muscles, TRPA1 hindered locomotion and altered mechanosensory responses to localized stimuli. When overexpressed in sensory neurons, TRPA1 apparently caused larvae to be more responsive and to prefer warmer temperatures. TRPA1 also caused significant but transient depolarization in resting membrane potential. These results highlight the role of TRPA1 in behavior and physiology, with potential implications for cancer-related mechanisms.

Disclosures: J. Kim: None. Y. Kim: None. J. Nadolski: None. R.L. Cooper: None.

Poster

PSTR286: Sensory Motor Systems

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Program #/Poster #: PSTR286.18/X2

Topic: G.01. Neuroethology

Support: GR533359

Title: Sex differences in context-dependent behavioral flexibility in *C. elegans*

Authors: *C. BAINBRIDGE¹, D. S. PORTMAN²;

²Dept. of Biomed. Genet., ¹Univ. of Rochester, Rochester, NY

Abstract: Animals must constantly integrate external sensory cues with internal-state information to optimally select adaptive behavioral strategies. Because nutritional requirements and reproductive strategies often differ by sex, flexible deployment of behavioral strategies are

often sexually dimorphic. Previous work from our lab and others indicates that *C. elegans* exhibits sexually dimorphic behavioral and neuronal responses to some nutrient-derived cues. However, the mechanisms by which biological sex impinges on neural circuits to orchestrate appropriate nutrient-dependent behavioral strategies remains poorly understood.

Here, we investigate sex differences in behavioral responses to nutrient stress by examining changes in locomotion during food encounter for three nutritional conditions (fed, 30min, and 3hr fasted). In hermaphrodites, food encounter causes a stereotyped reduction in speed (“basal” slowing) which is exaggerated upon nutrient stress (“enhanced” slowing). Whether males exhibit a similar integration of external and internal cues is unknown. To investigate this, we recorded both sexes for each nutritional condition as they navigated from a foodless environment onto food to capture both pre- and post-food encounter behavior. Interestingly, males only showed exaggerated slowing after food encounter under prolonged (3hrs) of nutrient stress. This was unlike hermaphrodites, which showed increased slowing during both pre- and post-food encounter with any nutrient stress. This suggests sex-specific tuning allowing males to decouple slowing behavior under different sensory contexts. Mutant analysis and optogenetic stimulation of the serotonergic food-sensing NSM neurons suggest that sex differences in serotonergic signaling may generate nutrient-dependent sex-specific differences in slowing behavior. Our approach provides a unique opportunity to explore mechanisms by which sex-specific modulation of circuit logic can bring about flexible and adaptive behavioral strategies.

Disclosures: C. Bainbridge: None. D.S. Portman: None.

Poster

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Program #/Poster #: PSTR286.19/X3

Topic: G.01. Neuroethology

Support: NIH Grant 5T32MH067564-22
NIH Grant 5R00NS119749-04

Title: Modeling decentralized control of swimming in the jellyfish *Clytia hemisphaerica*

Authors: *R. LU¹, K. CUNNINGHAM³, J. E. FITZGERALD², B. WEISSBOURD⁴, A. KENNEDY⁵;

¹Northwestern Univ., Chicago, IL; ²Neurobio., Northwestern Univ., Evanston, IL; ⁴Biol., ³MIT, Cambridge, MA; ⁵The Scripps Res. Inst., La Jolla, CA

Abstract: Generating and modulating behavior is an important function of nervous systems. To understand how early nervous systems achieved flexible control of behavior, we study the jellyfish *Clytia hemisphaerica*, a recently developed model organism for neuroscience. *Clytia* has around 10,000 neurons but lacks a brain; instead, it has a diffuse nerve net and two dense nerve rings. Still, *Clytia* can produce a range of internal-state-modulated behaviors. Historical

electrophysiology results suggest that a distributed network of coupled neurons with intrinsically generated subthreshold oscillations drives rhythmic swim pulses. Here, we use these results and new behavioral data to study how *Clytia*'s distributed pacemakers coordinate swimming and create different types of pulses.

To characterize swimming behavior, we recorded videos of semi-restrained, swimming jellyfish. We used pose estimation software (SLEAP) to track the tentacle bulbs lining the jellyfish's outer margin. We found that swim pulses varied in their degree of synchrony—the difference in contraction timing of different tentacle bulbs within a single pulse—and asymmetry—the difference in contraction depth of bulbs within a single pulse. Using a state-space reconstruction technique, we furthermore found six behavioral modes that capture the jellyfish's postural dynamics.

To study how *Clytia*'s nervous system modulates synchrony and symmetry, we next modeled the activity of neural pacemakers that generate swim pulses. We adapted a previously published Hodgkin-Huxley-like model of the *Aplysia* R15 neuron to replicate findings demonstrating subthreshold oscillations that can persist in the absence of spiking and synaptic inputs. We further propose a phenomenological model that qualitatively replicates the biophysical model by modeling a slow oscillatory variable interacting with fast spiking variables. To study network properties, we coupled our models into rings. We found that randomly initialized ring neurons will synchronize across a range of coupling strengths, and that transient asynchrony can be induced in a subset of neurons by applying external current. Notably, the network's ability to synchronize or desynchronize is critically dependent on the pacemakers' phase response curve, suggesting that single-cell electrophysiology can narrow down the necessary network control mechanisms. Taken together, our analyses suggest that our simple network can qualitatively replicate components of the behavioral data; however, further analysis and modeling are necessary to fully elucidate the jellyfish's behavioral dynamics and to explore how a neural system can generate those dynamics.

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Poster

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Program #/Poster #: PSTR286.20/X4

Topic: G.01. Neuroethology

Title: Neuronal dynamics underlying behavioral response variability in sensorimotor transformation

Authors: *J. MADHUSUDHANAN^{1,2}, C. FIESELER^{1,2}, A. PARINOV^{1,2}, M. ZIMMER^{1,2};
¹Neurosciences and Developmental biology, Univ. of Vienna, Vienna, Austria; ²Vienna Bio Ctr., Vienna, Austria

Abstract: Variability is a fundamental feature of many animal behaviors, including those in response to a sensory stimulus. Studies across different model organisms, including our previous work in *C. elegans*, show that the intrinsic brain-wide neuronal dynamics encode motor actions. However, it is unclear how such large-scale intrinsic neuronal dynamics process external sensory information and how they contribute to behavioral variability. One way to approach this question would be to look at the whole-brain neuronal dynamics at single-cell resolution and investigate where behavioral response decisions originate and how they are executed. Here, we leverage the tractability of the *C. elegans* nervous system to address these questions using oxygen chemosensory responses as a paradigm. When challenged with aversive oxygen stimuli (21% O₂), worms reliably reduce crawling speed, followed by a backward-crawling (reversal) escape response in approximately 60% of the trials. First, we tested whether the behavioral state of the animals prior to stimulus onset could predict the response outcome, but found that only locomotion speed revealed relatively weak predictive power. This finding led us to hypothesize that an ongoing neuronal state affects this response decision. We could reproduce a similar variable response to oxygen stimuli under whole-brain imaging conditions, where immobilized animals trigger fictive behavioral commands, which can be decoded from the activities of motor-related neurons. This provided a unique opportunity to record the sensorimotor transformation process at a whole-brain scale with single-cell resolution. Interestingly, we found that the response variability is not observed at the primary sensory neuron layer but only at the downstream inter- and motor neuronal layers of the worm brain. Moreover, multiple interneurons in the pre-stimulus brain state partially predict the future response to oxygen. Using machine learning approaches, we show that a distributed initial neuronal state, which evolves prior to stimulus onsets, best predicts the response decision. Finally, we confirm the contribution of some of the neurons participating in the pre-stimulus brain state to cause the decision using targeted neuronal manipulations via chemogenetics and optogenetics. Our findings show that sensory systems operate reliably to report information from the environment, while seemingly random variability in subsequent behavioral responses arises from non-random mechanisms like distinct initial conditions reflected in distributed neuronal population activity across the brain.

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Poster

PSTR286: Sensory Motor Systems

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Program #/Poster #: PSTR286.21/X5

Topic: G.01. Neuroethology

Support: NIH 2R15GM125073-03
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Title: Mommy Knows Best: Genetic and Circuit basis of female *Drosophila* decision-making

Authors: *D. SITARAMAN¹, Y. LEE², N. DHUNGANA³;

¹California State Univ. East Bay, Hayward, CA; ²California State University, East Bay, Hayward, CA; ³California State University- East Bay, San Mateo, CA

Abstract: Decision making is a fundamental process that influences all living organisms for their behavior and survival. In oviparous animals, the choice of where to lay eggs is especially important for the survival of their offspring. Here, we propose to use the fruit fly *Drosophila melanogaster*, as a system to understand the neural circuit mechanisms underlying decision making by studying the effects of neuronal and genetic manipulation on egg laying site selection or oviposition. By

systematically modifying the genetic tractability and relatively simple nervous system of *Drosophila*, we showed the sensory pathways, neural processing pathways, and motor outputs which are involved in oviposition site selection for the organism. We utilized the UAS-GAL4 system to manipulate gene expression in specific neurons and coupled with behavioral assays to test the egg-laying preferences under different substrate conditions. As the first level of brain processing, the sensory receptor mutants from the gustatory (Gr) family were screened to identify key pathways involved in substrate tasting choices. To investigate the circuit basis of decision making we systematically inhibited input, outputs and core processing regions of the mushroom body, a structure implicated in associative learning and other decision making tasks and assayed oviposition preferences. Silencing of these mushroom body and mushroom body output neurons (MBONs) help support the data of understanding a higher-order processing of sensory information. With the support of the corresponding inputs of MB such as octopaminergic neurons (OAs), dopaminergic neurons (DANs), anterior paired lateral (APL) and dorsal paired medial (DPM), we were able to show a full picture of decision-making neural process that during egg-laying behavior in *Drosophila*. In addition to the oviposition assay, we further perform feeding assay and positional tracking to identify the exploratory motivation of the fly during this decision-making process. With our result, it suggests a complex interplay of sensory inputs and internal processing. Through a combination of behavioral assays, imaging, and connectomics, our research builds a detailed neural map of the oviposition decision-making circuit in *Drosophila* which provides insights into the fundamental principles of neural computation and behavior.

Disclosures: D. Sitaraman: None. Y. Lee: None. N. Dhungana: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.22/X6

Topic: G.01. Neuroethology

Title: Beyond male enhancements: a synaptic mechanism for the evolution of sexually dimorphic singing

Authors: *J. V. GOGOLA^{1,2}, D. MARGOLIASH³, N. B. KASTHURI⁴;
²Med., ³Organismal Biol. and Anat., ⁴Neurobio., ¹Univ. of Chicago, Chicago, IL

Abstract: Understanding the neural basis of sex differences in behavior is crucial for deciphering brain function. Yet studies of sexual dimorphism often prioritize male specializations, implicitly treating females as the "default" state. In zebra finches, only males sing complex songs, traditionally attributed to females lacking male neural adaptations. Here, we reveal that female zebra finches possess a distinct synaptic organization in the syrinx (avian voice box) that actively suppresses singing ability. In adult females (n = 3), over half of syringeal synapses are weak *en grappe* (48.7%, $348.64 \mu\text{m}^2 \pm 23.47 \text{ SE}$), dispersed (Spatial Distribution Index: $G = 0.94$), and concentrated on small muscle fibers ($X^2 = 16.016$, $p < 1\text{E-}4$). These features contrast sharply with the strong *en plaque* (98.4%, $850.0 \mu\text{m}^2 \pm 62.91$, $p < 1\text{E-}26$), centrally organized ($G = 0.04$, $p < 1\text{E-}5$) synapses in males (n = 3). This female pattern differs from synaptic organization in other muscles we examined in both sexes (tongue: Male = $1046.8 \mu\text{m}^2 \pm 72.14$, Female = $1051.9 \mu\text{m}^2 \pm 61.10$, $p = 0.92$; lat: Male = $648.0 \mu\text{m}^2 \pm 24.89$, Female = $657.11 \mu\text{m}^2 \pm 45.32$, $p = 0.88$), and from typical vertebrate muscle innervation. Developmentally, both sexes initially possess similar proportions (Male = 48.2%, Female = 50.6%, $p = 0.77$; n = 3 each sex) and spatial organization of strong and weak synapses (Male $G = 0.85$, Female $G = 0.89$, $p = 0.83$), but males selectively prune weak synapses while females retain them into young adulthood (P70: Male = 17.6%, Female = 52.2%, $p < 1\text{E-}6$). Hormonal manipulations early in development altered synaptic morphology and vocal ability in both sexes, with synapse type and endplate band organization strongly predicting vocal competency (Masculinized Female = 22.6%, $p < 0.001$, $G = 0.84$; Feminized Male = 37.4%, $p < 0.001$, $G = 0.78$; n = 3 each treatment). Thus, sexual dimorphism in the syrinx emerges principally in the regulation of weak synapses in females, and is correlated with vocal behavior. Our comparative analysis with female chickens (47.0%, $p = 0.76$; $181.68 \mu\text{m}^2 \pm 36.33$; $G = 0.75$), combined with existing literature, suggests an evolutionary trajectory wherein endplate band formation first evolved in both sexes of songbirds, likely driven by hormone sensitivity of synapse type, but was subsequently suppressed in females. This suggests a new mechanism for the widespread evolutionary loss of female song in temperate zone songbirds compared to basal tropical species where both sexes sing. These findings demonstrate that female song absence reflects an active suppression mechanism established during development rather than a passive lack of specialization, challenging prevailing views of sexual dimorphism.

Disclosures: J.V. Gogola: None. D. Margoliash: None. N.B. Kasthuri: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.23/X7

Topic: G.01. Neuroethology

Support: NIMH DA031389

Title: Neural control of wing and leg movements in a terrestrial social display

Authors: *S. CAO¹, G. M. RUBIN², D. ANDERSON¹;

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Abstract: Terrestrial threat displays by flying insects communicate aggressive intent towards conspecifics using both the wings and legs, appendages whose primary functions are to control aerial vs. terrestrial locomotion, respectively. How such appendicular coordination is achieved is not well understood. Previously we identified a small (6-8 cell) cluster of AIP neurons in *Drosophila* that causally controls threat displays, raising the question of how this circuit node flexibly coordinates wing and leg movements. Here we have used connectomics, cell type-specific functional perturbations and calcium imaging to elucidate the circuit logic underlying male threat displays. We identified two distinct appendage-specific combinatorial modules that mediate threat actions. The wing module consists of two descending neurons (DNs), CL341 and DNp60, which act synergistically to control the wing-based actions: wing elevation and/or pumps. The leg module comprises two interneuron (IN) types, AVL491 and 10588. AVL491 activation promotes extended locomotion, while its co-activation with 10588 (an inhibitory IN) truncates this behavior into brief, discrete bouts of turning and charging - the leg-based threat actions. The common control of these two modules by AIP neurons serves to coordinate these appendages and to activate them in the context of agonistic interactions. These findings reveal a hierarchical and combinatorial neural circuit logic that coordinates independent appendicular control systems using distinct computations, to generate a flexible social display.

Disclosures: S. Cao: None. G.M. Rubin: None. D. Anderson: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.24/X8

Topic: G.01. Neuroethology

Support: NIH Grant NS105595
NIH Grant NS111477
Simons Foundation Grant 543015SPI

Title: Towards guidance laws that govern visual pursuit in mice

Authors: *D. J. POLLAK¹, M. MEISTER²;

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Abstract: Mice coordinate head and body movements during pursuit in a sequence of dynamic orienting behaviors. These behaviors take place on the order of tens of milliseconds and are subject to modification from unexpected changes in the environment. To elicit reliable and prolonged hunting bouts, we developed a motorized gantry to maneuver prey items through an

open field via a magnet beneath the floor. Mice pursued targets under two hunting paradigms: open- and closed-loop pursuit. In open-loop, the target moves along a pre-determined trajectory. In closed-loop, the target actively avoids the mouse. Food-deprived mice learned to pursue target objects, usually a chocolate chip, within minutes of being introduced to the setup. We compared various guidance law models to predict angular velocity of the head, body, and trajectory from directional variables using angular target position (known as proportional pursuit) and/or line of sight velocity (known as parallel navigation). Head-turning was best modeled by parallel navigation, while body movements were well approximated by multiple guidance laws. However, the head and the body do not respond instantaneously to visual input: head movements were delayed by 60-90 ms, trajectory changes were delayed by ~120 ms, and body movements were delayed by nearly 200 ms. This approach to visual orienting during natural behavior provides a promising avenue for understanding the neural circuits that produce it by condensing a complex behavior into a few informative variables that may govern vigorous pursuit behavior across species.

Disclosures: D.J. Pollak: None. M. Meister: None.

Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR286.25/X9

Topic: G.01. Neuroethology

Support: 1ZIAMH002950-09

Title: State-dependent gating of visual threat processing in area prostriata

Authors: *R. LINGG¹, C. ANCATEN², G. NNAMDI¹, K. YU³, H. WANG⁵, J. O'MALLEY⁴, Y. LENG⁶, H. A. TEJEDA⁷, M. A. PENZO⁴;

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Abstract: Internal hunger states dynamically modulate the behavior of foraging animals in complex environments with uncertain resource, or predator, densities. When starved, neuronal circuits indicating basic survival requirements bias attentional systems from vigilance (i.e. active monitoring of animal's surroundings) and towards actions promoting survival through resource acquisition. Reduced vigilance when foraging leads to a greater chance that a threat is not detected, or that detection is delayed sufficiently to challenge the likelihood of survival when a predator is encountered. In ecological situations, threatening stimuli often arise from the visual periphery unexpectedly, which creates a unique problem for a foraging animal when attentional

resources have been directed toward appetitive stimuli that tend to be located toward the ground in front of the organism. Here, we characterize a limbic-cortical visual processing area known as area prostriata (APr) involved in the detection of a rapidly advancing threat occurring in an organism's peripheral visual field that is uniquely sensitive to metabolic state. Using the looming visual threat task in combination with single-cell calcium imaging we show APr activity encodes the detection of threat, as well as the initiation and termination of escape from overhead looming threat; an effect associated with neurons that express the genetic marker for pituitary adenylate-cyclase-activating polypeptide (PACAP). Using head-fixed imaging of single-cell APr activity we observed excitatory APr responses to looming stimuli were greatest for rapid (300 °/s) stimuli occurring near the far periphery (i.e. 120° along the azimuthal plane), relative to stimuli presented toward center field. Excitatory APr responses to peripheral visual stimuli were abolished when mice were food deprived for 24hrs. Either 24hr food deprivation, or injection of the hunger-related hormone ghrelin also dramatically suppressed escape behavior in both male and female mice; a decrement that could be prevented by chemogenetic excitation of APr^{PACAP}. Follow-up *In vitro* recording of APr neurons that express PACAP indicated food deprivation reduced the intrinsic excitability of these neurons. We further characterize a novel midline thalamic projection stemming from the paraventricular thalamus (PVT) to APr that exhibits monosynaptic connectivity with APr^{PACAP} neurons and drives escape vigor. Further research is aimed at the broader circuit architecture whereby APr drives visuomotor responses, and how this is influenced by internal physiological conditions.

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Poster

PSTR286: Sensory Motor Systems

Location: SDCC Halls B-H

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Topic: G.01. Neuroethology

Support: Searle Scholars Award (M.L.-B.)
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Sloan Research Fellowship (M.L.-B.)
NIH New Innovator Award (M.L.-B., DP2EY036251).M

Title: Social action detection in the midbrain enables collective escape

Authors: *J. YU, J. MILAN, G. MEYERHOF, J. L. NAPOLI, M. LOVETT-BARRON;
UC San Diego, La Jolla, CA

Abstract: Social animals can transmit information about danger to improve survival for group members. Collective information transmission has been observed in fish schools, bird flocks, and

human groups, but the neural mechanisms enabling the detection of socially transmitted danger is poorly understood. Using the schooling glassfish *Danionella cerebrum*, here we find that escape from mechanosensory or visual threats is enhanced in groups, and mediated by visual access to escaping conspecifics. By performing two-photon calcium imaging in adult *Danionella*, we found neurons in the midbrain optic tectum responsive to visual stimuli mimicking freely swimming fish. Among this population, we found subsets of neurons responding to the rapid escape or sudden disappearance of social stimuli, but not disappearance of non-social stimuli. Accordingly, individual *Danionella* engaged with virtual conspecifics executed rapid avoidance behaviors when these virtual social partners escaped or disappeared. These results suggest that midbrain populations sensitive to conspecific disappearance engage escape behaviors in schooling fish, enabling the social transmission of threat information in animal collectives.

Disclosures: J. Yu: None. J. Milan: None. G. Meyerhof: None. J.L. Napoli: None. M. Lovett-Barron: None.

Poster

PSTR286: Sensory Motor Systems

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Topic: G.01. Neuroethology

Support: Helen Hay Whitney Foundation
Searle Scholars Award
Klingenstein-Simons Fellowship in Neuroscience
Kavli Institute for Brain and Mind
NIH R00MH112840

Title: Hunger promotes food seeking by modulating color vision

Authors: *K. A. MARTIN¹, E. R. DEAN³, M. LOVETT-BARRON²;
²Biol. Sciences, Neurobio., ¹Univ. of California San Diego, La Jolla, CA; ³Biol. Sciences, Neurobio., UCSD, San Diego, CA

Abstract: To enable adaptive behavior, animals must flexibly respond to the sensory input relevant to their physiological needs, such as the representation of food when hungry. Such adaptations can occur from changes to neural processes regulating sensory perception and motor action, and are subject to neuromodulation. However, little is known about how the nervous system selects subsets of sensory features in natural environments to enable need-motivated behaviors. Here, we study visually-driven movements in larval zebrafish, to investigate how hunger alters the sensory-motor circuits underlying prey capture. While the sensory-motor circuits driving prey pursuit and other visually-guided behaviors have been primarily studied in wavelengths visible to humans, zebrafish perceive prey in ultraviolet (UV) in natural sun-lit aquatic environments. Thus, we wanted to explore how hunger modulates behavior and neural

representation of prey in spectrally-realistic environments - specifically environments enriched for ultraviolet and red wavelengths.

We found hunger increases food-seeking behavior in both freely-moving and head-tethered fish. To better understand how hunger is mediating this increase, we first measured whether fish preferred red or UV light. Both hungry and sated fish displayed a strong preference towards red over UV light, but hungry fish comparatively greater engagement with UV light. We next explored how sensory responses changed across hunger state by recording from two primary visual areas involved in hunting using two-photon imaging during behavior. We presented hungry or sated zebrafish with either red or UV prey-like stimuli while systematically altering movement pattern and elevation of the stimulus to mimic different types of prey. Hungry fish showed less selective behavioral and neural responses to a diverse array of prey-like stimuli, compared with sated fish; both hungry and sated fish hunt UV prey-like stimuli, but hungry fish also hunt red stimuli. Taken together, this indicates hunger may expand the range of sensory stimuli fish will pursue, potentially facilitating riskier behavior to reduce physiological need. Overall, this work allows us to explore the interplay between physiological needs, state-dependent modulation, and sensory representation in ethologically-relevant behavior.

Disclosures: **K.A. Martin:** None. **E.R. Dean:** None. **M. Lovett-Barron:** None.

Poster

PSTR286: Sensory Motor Systems

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Topic: G.01. Neuroethology

Support: Helen Hay Whitney Foundation
The Searle Scholars Award
The Klingenstein-Simons Fellowship in Neuroscience
The Kavli Institute for Brain and Mind
NIH R00MH112840

Title: Larval zebrafish movement is modulated by visual wavelength and internal state

Authors: ***E. R. DEAN**¹, K. A. MARTIN², M. LOVETT-BARRON¹;
¹Biol. Sciences, Neurobio., UCSD, La Jolla, CA; ²Dept. of Biol. Sci., Univ. of California San Diego, La Jolla, CA

Abstract: Perception of external stimuli and animal's own internal state are both important in shaping that animal's behavioral repertoire. The presence of light can trigger different sets of behaviors in animals, from avoidance to food consumption, in a state-dependent manner. Aquatic environments have non-uniform spectral content due to diffraction; therefore, different wavelengths of light could provide distinct information about the environment, including the potential location of food. We are using larval zebrafish to explore how hunger influences

behavioral repertoires across ethologically-relevant wavelengths of light. We developed an open field arena for freely-swimming larval zebrafish that automatically switches between illumination with long and short wavelengths. Using SLEAP to automatically extract kinematic information about eye and tail movements, we found that hungry fish moved faster than sated fish, with greater speeds in red light versus ultraviolet (UV) light. This indicates hungry fish may use red light more than UV light for routine swimming, such as obtaining sensory feedback from self-generated movements. To perturb sensory input in these fish, we applied conditional cell ablation to red- or UV- cones to abolish specific channels of color vision. In hungry fish, red cone ablation significantly reduced the overall speed of movements and decreased the number of tail movements used in behavioral routines similar to prey-hunting. In contrast, UV-cone ablation showed a specific decrease in the number of tail movements used during hunting-like routines, but not other movement types. Therefore, internal state and the spectral features of environments interact to guide movement decisions. These experiments allow us to explore the impact of environmental statistics on state-dependent behavior. We are also exploring the neural underpinnings of hunger modulation of behavior across different wavelengths of light (see poster by KA Martin).

Disclosures: E.R. Dean: None. K.A. Martin: None. M. Lovett-Barron: None.

Poster

PSTR286: Sensory Motor Systems

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Klingenstein-Simons Fellowship in Neuroscience (ML-B)
Kavli Institute for Brain and Mind (ML-B)
NIH R00MH112840 (ML-B)

Title: Persistent brainstem inhibitory dynamics maintain a long-lasting internal state

Authors: *G. PADILLA¹, M. NGAN², J. JEFFERSON², J. SWINDLEHURST CHAN², K. MARTIN², M. LOVETT-BARRON²;

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Abstract: The ability to identify and respond to danger in one's environment is necessary for animal survival. For instance, brief exposure to a threatening sensory stimulus can result in sustained alterations to physiology, behavior, and neural activity that facilitate defensive behaviors. While the neural mechanisms of rapid threat-evoked behaviors are well studied, the mechanisms that mediate sustained changes in neural activity and behavior are poorly

understood. Here we address this question by studying an odor-driven persistent state in larval zebrafish. We found that a one-minute exposure to the death-associated odor cadaverine induces an elevated heart rate and locomotor suppression over 10-15 minutes in head-tethered fish. These effects are concentration-dependent, and specific to death/putrefaction-associated odors, but not other chemosensory threats (ie: NaCl). Using brain-wide cellular-level calcium imaging, we found that brief cadaverine exposure induces long-lasting activity in the lateral hindbrain - specifically GABAergic neurons. These persistently-active cells are thus poised to inhibit movement after threat detection. Accordingly, we found that brief cadaverine exposure suppressed visually-driven swimming at the level of the motor system; visually-responsive neurons were not modulated by cadaverine. Furthermore, optogenetic activation of local GABAergic neurons was sufficient to suppress spontaneous and visually-evoked movement. Together, this work identifies a brain-spanning network that transform brief salient events of ecologically-relevant threats into long-lasting changes in behavioral and neural dynamics.

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Poster

PSTR286: Sensory Motor Systems

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Program #/Poster #: PSTR286.30/X14

Topic: G.01. Neuroethology

Support: HFSP LT0002/2022L
Zuckerman STEM Program

Title: Transcriptomic comparison in schooling-impaired fish reveals convergent genetic pathways for social behavior

Authors: *D. ZADA;
Bar Ilan Univ., Ramat Gan, Israel

Abstract: Collective behaviors, such as fish schooling or bird flocking, arise from coordinated social interactions, yet the developmental and genetic foundations of these interactions remain unclear. Using the transparent teleost *Danionella cerebrum*, we investigated how social behavior develops and what genetic factors underlie its impairment. We first characterized the normal emergence of visually guided schooling, finding that fish initially exhibit aggregation behavior, followed by postural alignment with conspecifics. This behavioral sequence was accompanied by development of midbrain neuronal populations responsive to biological motion and shape of schooling fish. Disruption of social experience using developmental isolation impaired both the schooling behavior and the selectivity of neural responses to social stimuli. To identify the genetic basis of these impairments, we performed brain transcriptomic analysis in three conditions: juvenile fish, socially isolated adults, and crispants of autism-associated genes.

Despite distinct causes, these models of impaired schooling behavior shared a set of differentially expressed genes, suggesting a common molecular signature. Notably, we identified changes in genes related to thyroid hormone signaling, circadian rhythm regulation, and synaptic function. Using targeted genetic and pharmacological interventions, we validated the functional roles of several candidate genes in the regulation of schooling behavior. Our findings suggest that social and developmental disruptions converge on shared genetic pathways, providing insight into the biological foundations of collective behavior and present a new framework for understanding social deficits in neurodevelopmental disorders.

Disclosures: D. Zada: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.01/X15

Topic: G.03. Stress and the Brain

Support: Doctorado Nacional 21221349 (ANID) Scholarship
Puente UC

Title: Dopamine D2 receptor and LSD1: an epigenetic pathway to compulsivity

Authors: ***B. I. RIVERA**^{1,2}, P. AGUILERA MATURANA², B. ALMONACID², M. SCHENKE², F. GONZÁLEZ-REINOSO², M. JORQUERA², R. I. GATICA², M. E. ANDRES²; ²Fac. of Biol. Sci., ¹Pontificia Univ. Católica de Chile, Santiago, Chile

Abstract: Compulsivity is the inability to halt an ongoing action driven by an anxious state. It is a common dimension in various psychiatric disorders, including drug addiction and obsessive-compulsive disorder. The perpetuation of behaviours has been linked to epigenetic changes that modulate the strength of the involved synapses. Lysine-Specific Demethylase 1 (LSD1) is an epigenetic enzyme that plays a pivotal role in neuroplastic adaptation, modulating the expression of genes in response to stimuli. We propose that LSD1 plays a critical role in the development of compulsivity through the establishment of an epigenetic program that promotes the type of inflexible neuronal response observed in such behaviors. To test this idea, we employed a gradually developed compulsive model induced in rats by repeated administration of Quinpirole (QNP), a dopamine D2 receptor (D2R) agonist. Our findings reveal a reduction in D2R in the nucleus accumbens (NAc) and dorsal striatum (DS) of rats acutely treated with QNP. These changes dissipate following QNP repeated treatment. Additionally, an increase in the abundance of LSD1 substrates H3K4me2 and H3K9me2 was observed in the NAc of compulsive rats, in contrast to a downregulation in midbrain dopaminergic nuclei. Furthermore, changes in the expression of LSD1 splicing variants were detected pre- and post-compulsivity. These findings highlight significant epigenetic changes mediated by D2R in the development of compulsivity and suggest a regulation of LSD1 splicing by the D2R signalling.

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Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.02/X16

Topic: G.03. Stress and the Brain

Support: CZI grant

Title: Is neuronal DNA repair a cellular function of sleep

Authors: *Y. ZHAO¹, W.-J. BIAN², O. C. GONZALEZ², L. DE LECEA³;

¹Stanford Univ., Palo Alto, CA; ²Dept. of Psychiatry and Behavioral Sci., Stanford Univ., Stanford, CA; ³Psychiatry, Stanford Univ., Palo Alto, CA

Abstract: Neuronal activity triggers the formation of DNA double strand breaks (DSBs). Interestingly, activity-induced DSBs facilitate the expression of immediate early genes, which is crucial for experience driven changes to synapses, learning, and memory. Brain activity associated with wakefulness requires high metabolic rates/energy consumption to support various neuronal functions like action potential propagation and synaptic transmission to perform learning and memory. DSB repair pathways, non-homologous end joining (NHEJ) and homologous recombination (HR), both consume significant cellular energy to eliminate DSBs accumulated. How does the brain coordinate these two competing processes that both demand high energy consumption? Here we tested whether DNA damage accumulates in neurons *in vivo*. We first compared immunostaining of γ H2AX, an established marker for DNA damage across light dark cycle in mice. An increase in γ H2AX immunoreactive cortical neurons during the dark cycle was observed, with the mean puncta number per neuron increased by 475%. We then combined two-photon live imaging of 53BP1-eGFP fusion protein with EEG recording in natural sleep mice and those with sleep deprivation (SD). 53BP1 is an important regulator of Non-homologous End Joining (NHEJ), the main repair pathway in post-mitotic neurons. We monitored fluorescent 53BP1 puncta, which other studies have shown to coincide with immunostaining for γ H2AX. We visualized DNA repair by virus injection (AAV9-hSyn-GFP-53BP1) in excitatory Calcium/calmodulin-stimulated protein kinase II (CaMKII-positive) or inhibitory somatostatin-expressing (SST+) neurons. Our data show that, among supragranular neurons, sleep deprivation led to a decrease in 53BP1 puncta in 58% of CaMKII+ neurons compared to baseline. Notably, there is a negative correlation between delta power and the rate of puncta change in this subset of CaMKII+ neurons, suggesting that higher delta power is associated with a greater reduction in 53BP1 puncta and, consequently, increased DNA repair. We then induced slow wave by stimulating Chr2-transduced cortical SST+ neurons with blue light (1Hz). Slow wave induction reduced γ H2AX, with the mean puncta number per neuron

reduced by 30%. These results strongly suggest that DNA damage accumulates during wakefulness, while increased delta power during sleep facilitates DNA repair. Our findings indicate that DNA repair may represent a primary cell-autonomous function of sleep.

Disclosures: Y. Zhao: None. W. Bian: None. O.C. Gonzalez: None. L. de Lecea: None.

Poster

PSTR287: Cellular Actions of Stress

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Topic: G.03. Stress and the Brain

Support: NIH R01 Grant MH130399
NIH T32 Grant NS007453-22

Title: Stress-induced formation of DNA double strand breaks in neurons lead to molecular adaptations in the prefrontal cortex and hippocampus

Authors: *K. BOSIS¹, L. HEADY³, L. L. VOLLMER², J. L. BOLLINGER², R. MADABHUSHI³, E. S. WOHLER²;

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Abstract: Chronic stress exposure causes molecular and cellular adaptations in the prefrontal cortex (PFC) and hippocampus (HPC) which contribute to behavioral and cognitive maladaptation. Several molecular pathways have been identified as mediators of these neurobiological effects, however the transcriptional catalysts initiating these pathways are largely unknown. Recent work indicates that spatial constraints on chromatin organization can dictate gene expression through restriction of interactions between genes and their regulatory regions. Interestingly, other studies suggest that formation of neuronal DNA double strand breaks (DSBs) result from increased cell activity and can influence chromatin topology. This is intriguing as stress exposure is known to increase excitatory neurotransmission in corticolimbic regions as a result of increased glucocorticoid signaling. As such, we aimed to test the hypothesis that chronic stress induces neuronal DNA DSBs, and that this establishes the transcriptional patterns underlying resulting neurobiological and behavioral changes. In initial exploratory studies, four groups of male C57BL/6J mice (n=6/group) were exposed to 0, 1, or 14 days of chronic unpredictable stress (CUS), the fourth being left undisturbed for 14 days after CUS. Immunohistology in the PFC and HPC revealed that CUS exposure increased the DSB marker, γ H2A.X+, in neurons co-labeled with FosB. To follow-up, covalent DNA complexes were isolated from male PFC and HPC (n=6/group) to assess activity of topoisomerase-2 β , the enzyme controlling DSB formation. We observed that Top2 β covalent complexes were increased following 14 day CUS, with the HPC exhibiting higher levels than the PFC. These results

indicate that stress exposure causes formation of neuronal DNA DSBs in the PFC and HPC. Ongoing studies are using γ H2A.X ChIP-seq and RNA-seq analyses on sorted male and female PFC and HPC neurons in CUS-exposed mice to identify genes containing DSBs and assess their transcript levels. These studies may reveal a genomic mechanism orchestrating some of the molecular and cellular adaptations underlying the neurobiology of stress.

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Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.04/Y2

Topic: G.03. Stress and the Brain

Support: Funded by Unione Europea NextGeneration EU (Missione 4 Componente 2 Investimento 1.1 Avviso MUR PRIN 2022 PNRR D.D. n. 1409 del 14 09 2022 Prot. P20222WFZ3 - CUP: J53D23016000001

Title: Chronic mild stress impairs HDAC4 signaling in the nucleus accumbens: implications for glutamatergic synaptic transmission

Authors: C. MARCHETTI¹, G. CASTELLITTO², A. BERTOZZI¹, G. ACETO¹, C. GRASSI¹, C. COLUSSI², *M. D'ASCENZO¹;

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Abstract: Histone deacetylase 4 (HDAC4), which can target both histone and non-histone proteins, has gained substantial interest in translational neuroscience, as its expression and activity have been implicated in several neuropsychiatric disorders. However, the molecular and cellular mechanisms by which HDAC4 contributes to neuronal adaptations underlying depressive-like behaviors remain poorly understood. Here, we aimed to address this gap by investigating whether, and through which mechanism, HDAC4 contributes to neuronal adaptations in the nucleus accumbens (NAc) in the chronic unpredictable mild stress (CUMS) mouse model of depression. We found decreased levels of HDAC4 in CUMS mice in both total lysates and synaptosomes. This reduction did not depend on HDAC4 delocalization to the nucleus but was induced by GSK3 β -dependent phosphorylation of HDAC4 and its targeting to degradation. Indeed, GSK3 β inhibition (CT-99021) or treatment with the proteasome inhibitor MG132 reversed HDAC4 degradation. Decreased HDAC4 in NAc of CUMS mice was paralleled by reduced levels of synapsin I at the presynaptic compartment and by altered AMPA expression at the postsynaptic density regions. To link these findings to HDAC4-dependent synaptic regulation, we inhibited HDAC4 activity in brain slices using the selective inhibitor LMK235 and assessed glutamatergic synaptic transmission in medium spiny neurons (MSNs) of the NAc through whole-cell patch-clamp recordings. HDAC4 inhibition led to a significant

reduction in the frequency of miniature excitatory postsynaptic currents (mEPSCs) compared to control slices. In LMK235-treated slices, we also observed increased NMDA receptor-mediated currents, a decreased AMPA/NMDA ratio, and an upward shift in the input-output curve, indicating enhanced synaptic responsiveness and strength. Taken together, these results suggest that both pharmacological and stress-related downregulation of HDAC4 impairs glutamatergic synaptic transmission in the NAc.

Disclosures: C. Marchetti: None. G. Castellitto: None. A. Bertozzi: None. G. Aceto: None. C. Grassi: None. C. Colussi: None. M. D'Ascenzo: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.05/Y3

Topic: G.03. Stress and the Brain

Support: NCN OPUS 2021/41/B/NZ3/04099
Horizon Europe SAME-NeuroID: 101079181

Title: Dissecting the role of astrocyte-specific synaptic proteins in glucocorticoid receptor - dependent plasticity and its behavioral manifestations.

Authors: *A. M. LECH, Senior¹, P. ZIUZIA^{1,2}, B. ZGLINICKI¹, M. SKULA^{1,3}, M. SLEZAK¹; ¹Life Sci. & Biotech. Ctr., Lukasiewicz Res. Network – PORT Polish Ctr. for Technol. Develop., Wroclaw, Poland; ²Wroclaw Univ. of Envrn. and Life Sci., Wroclaw, Poland; ³Wroclaw Univ. of Sci. and Technol., Wroclaw, Poland

Abstract: Depression is one of the most prevalent mental health conditions worldwide, characterized by a diverse range of clinical symptoms, including impaired emotional regulation, social interactions and decision-making. While extensive evidence implicates disturbed synaptic homeostasis in the pathophysiology of depression, the role of astroglia in regulating synaptic activity and shaping neural connectivity remains underexplored. In previous studies, we found profound changes in transcriptomic profile of prefrontal cortex (PFC) astrocytes in a mouse model of chronic stress, which were mediated by glucocorticoid receptor (GR). One group of genes regulated by stress included astrocyte-specific factors which mediate synapse formation and elimination. Here, we investigated how manipulation of genes controlling synapse number influences behavioral effects of chronic stress and how these genes contribute to the action of antidepressant. To achieve astrocyte-specific protein silencing in the mPFC, we bilaterally injected mice with vectors carrying shRNA against each of target genes and a GFP reporter. Behavioral phenotyping was conducted using an automated system that enables high-throughput tracking of individual and group behaviors in freely moving mice. Chronic corticosterone (CORT, 21 days in the drinking water) paradigm was used to induce depressive-like behavior. We assessed individual behaviors (e.g., locomotion, and speed) as well as social interactions

(e.g., nose-to-nose and nose-to-tail contacts, and following behavior) in both sexes during three sessions: 3 weeks after vector administration (baseline), after completion of stress paradigm (CORT), and 24h after injection of a single subanesthetic dose of ketamine. Immunostaining against GFP and astrocyte-specific markers was used to validate the distribution and efficacy of the gene silencing. Principal Component Analysis (PCA) of behavioral traits revealed that silencing of specific astrocytic proteins distinctly altered the behavior of animals following chronic CORT and ketamine exposure. We further identified the behavioral variables contributing most to the observed variance in PCA. These findings underscore the pivotal role of astrocyte-derived synaptic proteins in GR-dependent stress mechanisms, advancing our understanding of how astroglia modulate neural circuits in depression.

Disclosures: A.M. Lech: None. P. Ziuzia: None. B. Zglinicki: None. M. Skula: None. M. Slezak: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.06/Y4

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant R01MH109382

Title: Genetic variants of EIF2AK3 in astrocytes is associated with altered stress tolerance in neuron and astrocyte coculture

Authors: *J. K. ARYA¹, S. GHURA¹, S. B. NEWTON⁴, E. ALVAREZ PERIEL², K. L. JORDAN-SCIUTTO³, C. AKAY-ESPINOZA²;

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Abstract: PERK, an endoplasmic reticulum stress response kinase, is tightly regulated at multiple levels, and subtle variations in its activity and signaling can shift stress response outcomes. PERK has three commonly occurring exonic single nucleotide variants frequently inherited together as PERK haplotype B, which occurs in 28% of the global population and is associated with several chronic neurodegenerative diseases, although its impact on PERK activity is contradictory. We hypothesized that PERK-B was associated with increased kinase activity, altering stress response and stress tolerance and that small but consistently elevated PERK activity, in combination with additional environmental or genetic risk factors, would lead to adverse outcomes. Comparison of stress response between PERK-A and PERK-B neuroglial cultures from the novel knock-in PERK-B and wild-type PERK-A mice demonstrate differential and transient changes. Following ER stress, neuroglial, but not enriched neuronal, cultures expressing PERK-B exhibited higher eIF2 α phosphorylation, the best-known PERK substrate, at 4-6 h and activation of PERK signaling, specifically the proapoptotic CHOP, at 12 h. However,

ER stress-mediated death at 24 h was comparable between the PERK-A and PERK-B neuroglial cultures. PERK-B neuroglial cultures pretreated with a low-dose PERK-specific activator exhibited increased vulnerability to N-methyl-D-aspartate-induced death compared to PERK-A cultures; the observed difference in stress tolerance was driven by astrocytes. The association of PERK-B with transient changes in stress response and decreased stress tolerance with higher vulnerability to cellular stress, albeit transient and small/moderate sized, has clinical relevance as they might contribute to the overall genetic vulnerability in chronic diseases.

Disclosures: J.K. Arya: None. S. Ghura: None. S.B. Newton: None. E. Alvarez Perial: None. K.L. Jordan-Sciutto: None. C. Akay-Espinoza: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.07/Y5

Topic: G.03. Stress and the Brain

Support: NIH grant MH119283
The Catherine and Hunter Pierson Chair in Neuroscience
Carol Lavin Bernick Faculty Grant from Tulane University

Title: The membrane glucocorticoid receptor is associated with the primary cilium

Authors: *L. HARRISON¹, E. BURKE², J. G. TASKER^{1,3};
¹Dept. of Cell and Mol. Biol., ²Dept. of Biol. Chem., ³Tulane Brain Inst., Tulane Univ., New Orleans, LA

Abstract: Glucocorticoids (GCs) exert rapid effects via a membrane-localized receptor, but the nature of this receptor remains unknown. Cell culture studies and mouse models indicate an interaction between GCs and the Hedgehog (Hh) system, a signaling system critical for neural development that requires the primary cilium for proper function. Specifically, a potential locus of interaction between GCs and Hh is the GPCR-like protein Smoothed of the Hh pathway. Thus, we hypothesized that rapid GC effects are mediated via a membrane receptor that utilizes the primary cilium for signaling. Most cells contain a single primary cilium, an immotile projection from the cell body that forms a signaling hub critical for such functions as neurodevelopment and metabolic homeostasis. We detected glucocorticoid receptor (GR) immunoreactivity in primary cilia of the 3T3 mouse embryonic fibroblast cell line, a model for primary cilia. Interestingly, we also consistently detect GR immunoreactivity in an area at the base of the primary cilium, co-localizing with a marker for the Golgi apparatus. Like 3T3 cells, neuronal cells of the N42 hypothalamic cell line also show a distribution of GR immunoreactivity in and near the base of the cilium. The GR immunoreactivity can also be detected in non-permeabilized cells, indicating that the antibody can access the GR on the surface of the cell. A similar pattern of surface localization of GR can also be observed in live

cells incubated with the GR antibody prior to fixation. In contrast, another cilia- and periciliary-localized protein, Dexras1, which is constrained to the inside of the cell, is not labeled in non-permeabilized cells, suggesting that the antibody does not penetrate the membrane. The distribution of GR immunoreactivity in the periciliary region is regulated by agonist. Thus, co-incubation with GCs and an agonist of the Smoothened receptor results in a striking redistribution of GR signal to puncta in the cytosol. Collectively, these results suggest that a membrane GR is associated with the primary cilium and interacts with the Hh pathway.

Disclosures: L. Harrison: None. E. Burke: None. J.G. Tasker: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.08/Web Only

Topic: G.03. Stress and the Brain

Title: Stress-induced histone post-translational modifications in peripheral immune cells

Authors: *A. GRUSCA;

Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY

Abstract: Psychosocial stress can induce persistent immune alterations that impact psychiatric vulnerability, yet the epigenetic mechanisms driving these changes remain poorly defined. Peripheral immune cells rapidly respond to environmental stimuli, and recent evidence suggests that stress may specifically alter chromatin landscapes via histone post-translational modifications (PTMs). Here, we investigate stress-responsive histone PTMs—e.g., H3 lysine 56 acetylation (H3K56ac) and H3 lysine 27 trimethyl serine 28 phosphorylation (H3K27me3S28ph)—within mouse peripheral blood mononuclear cells (PBMCs) following chronic social defeat stress. Unbiased histone mass spectrometry identified distinct PTM profiles in stress-exposed animals that differed by behavioral phenotype. Flow cytometry studies aim to determine whether stress-induced PTM changes are primarily enriched in monocytes, a stress-reactive PBMC subset implicated in inflammatory signaling and CNS trafficking. To assess downstream impact, we are integrating CUT&RUN, RNA-seq, and cytokine profiling in sorted monocytes to evaluate relationships between chromatin state, transcriptional programs, and systemic inflammatory profiles, including key cytokines such as IL-6, TNF- α , and IL-10. Parallel analyses are underway on brain-infiltrating monocytes to assess whether stress-induced transcriptional signatures are preserved across compartments. Together, this work seeks to identify epigenetic mechanisms by which stress reprograms peripheral immune cells and shapes immune phenotypes linked to resilience or vulnerability. These insights may ultimately inform new targets for the treatment of inflammation-associated psychiatric disorders.

Disclosures: A. Grusca: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.10/Y6

Topic: G.03. Stress and the Brain

Title: Stress and noradrenaline trigger distinct response profiles in hippocampal neurons and astrocytes

Authors: *S. DUSS¹, M. WILHELM¹, F. HELMCHEN², P. RUPPRECHT², J. BOHACEK¹;
¹ETH Zurich, Zuerich, Switzerland; ²Brain Res. Inst., Univ. of Zurich, Zuerich, Switzerland

Abstract: The acute stress response is comprised of complex biological processes that help organisms cope with challenging situations. A crucial component of the acute stress response in the brain is the release of noradrenaline (NA) from the locus coeruleus (LC). While most research has focused on neurons, recent studies have pointed to a pivotal role of astrocytes in the stress response [1]. Astrocytes express all adrenergic receptors, they are highly sensitive to noradrenaline [2], and it has even been proposed, that astrocytes can mediate some of the effects observed in neurons [3][4]. Here, we set out to systematically investigate how the LC-NA system regulates the activity of astrocytes, excitatory and inhibitory neurons in the mouse hippocampus, and how this compares to the impact of stress on these cell types. To this end, we titrated optogenetically controlled release of NA by the LC to match NA-levels triggered by natural stressors. We then used fiber photometry recordings and two-photon imaging in awake mice to compare the effects of stress to isolated LC-NA release. We found distinct response profiles across different stressors and LC stimulation paradigms. While astrocytic activation was highly similar between LC stimulation and stress, neuronal calcium imaging revealed dissimilar effects of stress or isolated LC-NA activation. Additionally, we tracked the same cells across multiple imaging days and uncovered heterogeneous but predominantly stable response profiles within the different cell-types. Finally, interfering with astrocytic calcium levels supported the notion of a key role of astrocytes in the noradrenergic signaling cascade. Together, our results foster the understanding of the cellular mechanisms underlying the stress response in the brain. Such a paradigm shift away from a neuro-centric approach may be crucial for developing targeted interventions for stress-related disorders.

References

1. Murphy-Royal, C., Gordon, G. R. & Bains, J. S. Stress-induced structural and functional modifications of astrocytes—Further implicating glia in the central response to stress. *Glia* **67**, 1806-1820 (2019).
2. Hertz, L., Lovatt, D., Goldman, S. A. & Nedergaard, M. Adrenoceptors in brain: Cellular gene expression and effects on astrocytic metabolism and $[Ca^{2+}]_i$. *Neurochem. Int.* **57**, 411-420 (2010).
3. Chen, A. B. *et al.* Norepinephrine changes behavioral state through astroglial purinergic signaling. *Science* **388**, 769-775 (2025).
4. Lefton, K. B. *et al.* Norepinephrine signals through astrocytes to modulate synapses. *Science* **388**, 776-783 (2025).

Disclosures: S. Duss: None. M. Wilhelm: None. F. Helmchen: None. P. Rupprecht: None. J. Bohacek: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.11/Y7

Topic: G.03. Stress and the Brain

Support: 1RF1AG082388-01

Title: Effects of chronic restraint stress on de novo protein synthesis in the hippocampus

Authors: *G. HOWARD¹, N. I. NICOL³, H. M. JESTER⁴, T. MA²;

¹Wake Forest Sch. of Med., WINSTON SALEM, NC; ²Intrnl. Medicine-Geriatrics, Wake Forest Sch. of Med., Winston Salem, NC; ³Wake Forest Univ., Winston-Salem, NC; ⁴Geriatrics and Gerontology, Wake Forest Univ. Sch. of Med., Germanton, NC

Abstract: Prolonged or chronic stress can be detrimental for both physical and mental health, leading to various diseases. Investigation of the molecular/cellular mechanisms underlying chronic stress can provide insights into novel therapeutic strategies. Chronic restraint stress (CRS) has been shown to induce a depressive-like phenotype in rodents, yet the biochemical component of CRS remains elusive. Substantial evidence demonstrates that integral de novo protein synthesis (mRNA translation) is essential for synaptic and cognitive function. This study aims to explore the relationship between chronic restraint stress, behavioral changes, and de novo protein synthesis. Wildtype male C57B6L mice, aged 3-5 months, were randomly separated into four experimental groups: group housed, singly housed (SH), food and water deprived for six hours a day for seven days, and CRS subjected to six hours of restraint stress for seven days. Two-three mice per group were sacrificed immediately following their stress condition to determine immediate effects of stress, while the remaining mice completed nine, single-day cycles of sucrose preference testing (SPT), which is a widely used behavioral test to assess the anxiety-like phenotype associated with restraint stress. Four days following the stress conditions, a trending decrease ($n = 11/12$, $p = 0.066$) in the CRS group's sucrose preference was seen, indicating the paradigm causes an anxiety-like phenotype. Hippocampal slices were harvested for measurement of de novo protein synthesis using the Surface Sensing of Translation (SUnSET) assay. We found a trending decrease in de novo protein synthesis in hippocampus immediately following the stress condition ($n = 5$, $p = 0.0528$), which recovered to the level of SH one-month after stress ($n = 6$, $p = 0.9942$) using a one-way ANOVA. In summary, our study demonstrates an association between chronic restraint stress and impairment of de novo protein, which could lead to cognitive deficits and synaptic failure.

Disclosures: G. Howard: None. N.I. Nicol: None. H.M. Jester: None. T. Ma: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.12/Y8

Topic: G.03. Stress and the Brain

Support: 1RF1AG082388-01

Title: Effects of acute restraint stress on translational control in mouse hippocampus

Authors: *Z. WAN¹, G. HOWARD², H. M. JESTER⁴, N. I. NICOL⁵, T. MA³;

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SALEM, NC; ³Intrnl. Medicine-Geriatrics, Wake Forest Sch. of Med., Winston Salem, NC;

⁴Geriatrics and Gerontology, Wake Forest Univ. Sch. of Med., Germanton, NC; ⁵Wake Forest Univ., Winston-Salem, NC

Abstract: Stress can be detrimental for physical and mental health, leading to various disease state. Elucidation of the molecular mechanisms underlying stress could provide insights into novel therapeutic strategies for stress-related diseases. In this study, we aim to establish a model for acute restraint stress (ARS) in mice, which involves a single 6-hour restraint session in 1 day, and examine its effects on cognitive function and protein synthesis (mRNA translation), which plays a critical role in long-lasting forms of memory and synaptic plasticity. We designed a restraint tube just big enough to accommodate one mouse and has 16 holes punctured for adequate breathing. Male C57B6L mice were separated into four different groups: group housed (GH, n = 4), single housed (SH, n = 7), single housed with food and water deprivation (FWD, n = 7), and single housed with food and water deprivation plus acute restraint stressed (ARS, n = 7). All four groups were habituated with sugar preference test (SPT) three days prior to ARS exposure to establish baseline measure for later behavioral evaluation. Two mice from each group were sacrificed on the day of stress exposure, while the remaining underwent SPT the same day and every 4 days for another 34 days. Surface Sensing of Translation (SUnSET) was conducted on acute hippocampal slices to evaluate the overall de novo protein synthesis, and the result didn't reveal any significant changes between groups after the ARS exposure. We further investigated multiple signaling pathways involved in protein synthesis (translational control). Western blot analysis on hippocampus revealed that levels of eukaryotic elongation factor 2 (eEF2), a key translational factor controlling elongation phase of the protein synthesis, are significantly decreased in the ARS and FWD groups compared with the SH group when using unpaired T-test, with $p = 0.002$ and $p = 0.001$, respectively. Interestingly, total level of postsynaptic density protein 95 (PSD-95) also shown significant reduction in the FWD group comparing with the SH group using unpaired T-test, $p = 0.03$. In summary, our study suggest that acute restraint stress can result in dysregulation of mRNA translational control, which may contribute to impaired cognition and synaptic function associated with stress condition.

Disclosures: Z. Wan: None. G. Howard: None. H.M. Jester: None. N.I. Nicol: None. T. Ma: None.

Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

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Program #/Poster #: PSTR287.13/Y9

Topic: G.03. Stress and the Brain

Title: Spatial transcriptomics reveals stress hormone-dependent modulation of gene expression in hippocampal area CA2

Authors: *B. KANDEMIR, C. GU, G. M. ALEXANDER, S. M. DUDEK;
Neurobio. Lab., Natl. Inst. of Environ. Hlth. Sci., Research Triangle Park, NC

Abstract: The stress hormone corticosterone, produced by the hypothalamic-pituitary-adrenal (HPA) axis, modulates stress responses via glucocorticoid receptor (GR) and mineralocorticoid receptor (MR) signaling in the brain. Although GRs are broadly expressed in the hypothalamic paraventricular nucleus (PVN), hippocampus, and anterior pituitary, MRs are specifically enriched in the hippocampus, particularly within area CA2. Mouse conditional knockout studies have shown that MRs regulate baseline stress responses, learning, memory, and social behaviors, as well as influencing expression of CA2-specific marker genes (e.g. *Amigo2* and *Fkbp5*) and synaptic plasticity in the hippocampus. Therefore, we asked whether MR-related gene expression in CA2 can also be modulated by changes in corticosterone levels. To do this, we surgically removed the adrenal glands (adrenalectomy; ADX), which nearly eliminates endogenous corticosterone. We found that ADX increased MR protein expression, consistent with previous work, and changed MR cellular localization (increased cytosolic/nuclear ratio) compared to sham-operated control mice (sham), suggestive of MR inactivation in ADX animals. Furthermore, sham hippocampus had higher nuclear MR levels than in non-surgical control (NSC) mice, indicating a stress (or history of stress) condition for sham animals. Using Visium HD spatial transcriptomics, we examined CA2 gene expression profiles under these conditions. Our findings demonstrate that *Nr3c2* (encoding MR protein) expression is significantly increased in ADX mice relative to sham (approximately 2-fold increase) and slightly increased for sham mice comparing to NSC groups consistent with MR protein expression in these conditions. Notably, the expression of the CA2 marker gene *Amigo2* decreased more than 4-fold in ADX animals but increased by approximately 3-fold in sham mice. Additionally, the expression of the stress-responsive gene *Fkbp5* was reduced by ~3-fold in ADX mice compared to sham and NSC groups. Overall, ADX conditions, associated with decreased MR activity, increased the number of downregulated genes, whereas sham animals, with enhanced MR activity, showed a higher number of upregulated genes in CA2. These differential gene expression patterns were further validated using MERSCOPE spatial transcriptomics. Our results highlight the sensitivity of MR-mediated CA2 gene expression to corticosterone fluctuations, suggesting dynamic modulation of CA2 genes in response to adrenal hormone levels and stress-related conditions. Future work will attempt to disentangle MR-specific gene changes from those regulated by GRs.

Disclosures: B. Kandemir: None. C. Gu: None. G.M. Alexander: None. S.M. Dudek: None.

Poster

PSTR287: Cellular Actions of Stress

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Program #/Poster #: PSTR287.14/Y10

Topic: G.03. Stress and the Brain

Support: CU Anschutz Endowed Chair seed funds

Title: Extracellular vesicle stress dynamics in a healthy adult cohort: building a normative framework

Authors: ***R. ORSO**, S. NICKLES, P. MOON, S. BERKOWITZ, T. L. BALE;
Dept. of Psychiatry, Univ. of Colorado, Aurora, CO

Abstract: Extracellular vesicles (EVs) are cell-derived nanoparticles that play a crucial role in cell-to-cell communication to maintain homeostasis and have emerged as promising biomarkers for various health and disease states. Acute stress can alter EV composition and secretion, but little is known regarding EV proteomic changes across biological fluids. Our current study aimed to assess whether salivary EV responses to acute stress could serve as a non-invasive proxy for plasma EVs, and to evaluate their association with physiological stress markers in healthy adults. Twenty-seven healthy participants (14 males, 13 females; aged 21-35 years) completed a single visit session consisting of three 20-minute experimental phases: baseline (relaxation video), stress exposure (stress-inducing video), and recovery (quiet reading). Heart rate and skin conductance were continuously monitored, while blood and saliva samples, along with subjective ratings of perceived stress, were collected after each timepoint. Cortisol concentrations in plasma and saliva were quantified using enzyme-linked immunosorbent assay. EVs were isolated from both plasma and saliva samples using Izon size exclusion chromatography columns, and EV concentration and size were analyzed via nanoparticle tracking analysis using the ZetaView platform. Male and female participants reported increased perceived stress and skin conductance in response to the stress exposure, with both measures returning to baseline after recovery. Cortisol level changes over the course of the study were sex-specific, with males showing differences in both plasma and salivary cortisol levels following stress exposure, while females showed little change. Plasma EV concentration was significantly increased in males following stress exposure, but no significant changes were observed in females. In saliva, only females exhibited a significant increase in average EV particle size after stress exposure. In conclusion, this study demonstrates that acute stress exposure elicits changes in stress-related markers and EV characteristics, with distinct sex-dependent outcomes. Although changes in salivary EV characteristics did not fully mimic those of blood plasma EV responses, their sensitivity to stress suggests potential use as a noninvasive biomarker. Current studies are examining the unbiased EV proteomics changes in these samples over time and between sexes. These findings support the relevance of EVs in capturing stress-related alterations and highlight sex- and fluid-specific EV dynamics.

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Poster

PSTR287: Cellular Actions of Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR287.15/Y11

Topic: G.03. Stress and the Brain

Title: Oxidative stress and APE-1 suppression in the chronically stressed brain

Authors: *Y. REYES;
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Abstract: Neuronal activity constantly generates reactive oxygen species (ROS), which, in excess, lead to oxidative stress and damage to proteins, lipids, and DNA. To counteract this, neurons rely on defense mechanisms, including APE-1 (apurinic/apyrimidinic endonuclease 1), an enzyme involved in DNA repair and antioxidant regulation. APE-1 detects and corrects oxidative lesions, preserving neuronal function. Chronic stress disrupts homeostasis, triggering physiological and behavioral changes. In animal models, unpredictable chronic stress (UCS) mimics human conditions like depression and anxiety. Although chronic stress affects neurotransmission and neurogenesis, its impact on DNA repair—particularly APE-1—remains understudied. This project examines whether UCS alters APE-1 expression in the prefrontal cortex, a region involved in emotional and executive functions. We exposed CD1 mice to six weeks of UCS and assessed both behavior and APE-1 expression. Stressed animals exhibited decreased nest-building quality and reduced grooming latency—behaviors associated with depressive-like states. Molecular analysis revealed a significant reduction in APE-1 levels in the prefrontal cortex of stressed mice compared to controls. These results suggest that chronic stress impairs the brain's ability to activate DNA repair mechanisms, increasing vulnerability to oxidative damage and emotional dysregulation. Reduced APE-1 expression may be a key link between sustained stress and neurobehavioral susceptibility. Our findings provide new insights into stress-related molecular changes and highlight APE-1 as a potential therapeutic target. Future studies could explore interventions aimed at restoring APE-1 function to enhance brain resilience under chronic stress.

Disclosures: Y. Reyes: None.

Poster

PSTR288: Adolescent Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR288.01/Y12

Topic: G.03. Stress and the Brain

Support: Huck Institutes of the Life Sciences

Title: Does sleep in a mouse model of developmental allergic asthma explain adult behavioral outcomes?

Authors: *V. E. CONRAD¹, R. N. HALLAHAN², W. J. HORTON³, S. A. CAVIGELLI⁴;
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Abstract: Asthma is a global health concern that affects over 300 million people of all ages, resulting in substantial socioeconomic losses (Giacco et al., 2016). With prevalence rates of approximately 9.5%, it remains the leading chronic illness of children and adolescents. Asthma is further related to other adverse outcomes, including an increase in learning difficulties, behavioral problems, and social problems (Dudeney et al., 2017). Notably, psychological disorders—particularly internalizing disorders like anxiety—are higher in asthmatic youth than non-asthmatic youth, with 22.7% and 6.9% prevalence rates, respectively (Dudeney et al., 2017; Lawrence et al., 2015). We hypothesized that sleep could be an exacerbating factor that leads to the threefold increase of anxiety observed in individuals with asthma. To investigate this, we utilized a mouse model of asthma induced by intranasal administration of house dust mite (HDM) extract. Pups were bred in-house and, beginning in week two, we exposed pups to HDM by administering 60 µg intranasally three times a week until adulthood. During HDM exposure, we monitored home cage activity for 5 consecutive days during weeks 3, 5, and 7. Sleep parameters, including sleep timing and circadian alignment, were derived from these activity data. In early adulthood, baseline, reactivity, and recovery of circulating corticosterone concentrations after restraint stress were measured to assess physiological stress regulation. In week 9, a final round of sleep monitoring was conducted during which behavioral tests were performed to measure anxiety-related behavior. Behavioral tests included the elevated plus maze, novel social partner, sucrose preference test, and light-dark box. Results compared the effect of induced asthma on anxiety phenotypes and the interaction between sleep and these two variables. This work may have implications for the future treatment of asthma comorbidities.

Disclosures: V.E. Conrad: None. R.N. Hallahan: None. W.J. Horton: None. S.A. Cavigelli: None.

Poster

PSTR288: Adolescent Stress

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR288.02/Y13

Topic: G.03. Stress and the Brain

Support: Huck Institutes of the Life Sciences

Title: Brain gene expression as a possible mechanism underlying the relationship between allergic asthma and behavioral and biological outcomes in a mouse model

Authors: *S. VELU¹, H. HUSAK², W. J. HORTON³, H. M. KAMENS⁴, S. A. CAVIGELLI⁵;
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Abstract: Children and adolescents with asthma often suffer from different psychological, neurological, and physiological co-morbidities such as anxiety, depression, social/cognitive difficulties, and growth impairment. A causal relationship between asthma and these outcomes has been investigated and established using rodent models in recent years; however, the factor(s) mediating these changes is less clearly understood. We suspect that modifications in brain gene expression may be a critical component in this pathway. In the present study, to further understand the effects of allergic asthma on brain gene expression, we repeatedly exposed male and female Balb/cJ mice to intranasal control saline or 60 ug house dust mite (HDM) extract in saline throughout development (3x/week for 7 weeks). In adulthood, behavioral tests were conducted to measure anxiety-like (elevated plus maze, light dark test), depression-like (sucrose preference test), social (novel social partner test), and cognitive-related (object location memory) behaviors. Body weight was monitored throughout development, and blood and tissue samples were collected from adults to quantify asthma-like symptoms (e.g. airway inflammation) and glucocorticoid secretion (which is often altered in asthma patients). The expression levels of various genes suspected to contribute to these outcomes were measured using RNA extraction, cDNA synthesis, and QT PCR. Mice exposed to intranasal allergen throughout development gained less weight, had increased inflammation and expression of allergic asthma-related cytokine gene expression (IL-4,-5,-13), increased anxiety-like behavior, decreased social behavior, and decreased spatial memory. Some of these outcomes were associated with altered expression of HPA-regulation genes while others were associated with altered expression of inflammation-related genes in emotion regulation areas of the brain. The results suggest multiple changes in the brain each of which may account for different aspects of allergic asthma comorbidities. This contributes to our growing understanding of possible mechanisms by which allergic asthma affects development.

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Poster

PSTR288: Adolescent Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR288.03/Y14

Topic: G.03. Stress and the Brain

Support: Fairfield University Science Institute Research Grant
Fairfield University Kathleen B. Trainor Student Research Funds

Title: Evaluating the impact of adolescent social isolation on depressive-like behavior and alcohol preference in male and female Long-Evans rats

Authors: *S. M. HARDING, M. KENNY, C. SURPRENANT;
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Abstract: Previously, our lab has shown that social isolation beginning in adolescence increases anxiety-like behaviors and impairs spatial memory in male and female Long-Evans rats. In the present study, we extended this work to examine the effects of prolonged social isolation on depressive-like behavior and alcohol preference. Male and female Long-Evans rats were assigned to one of four groups on postnatal day (P) 28: socially isolated males (SI-M) and females (SI-F); and group-housed males (GH-M) and females (GH-F) (n = 9 per group). SI rats were singly housed in standard cages while GH rats were housed in larger, open-top cages in groups of three and received environmental enrichment. Weekly behavioral tests were conducted in adulthood beginning on P64. Anxiety-like behaviors were assessed using the open field test (OFT) and elevated plus maze (EPM); depressive-like behavior was measured with the novelty-suppressed feeding test (NSF); and alcohol preference was evaluated in a subset of rats using conditioned place preference (CPP). SI rats of both sexes displayed increased anxiety-like behaviors in the OFT and EPM compared to GH controls. Additionally, females traveled more distance and spent more time in the center of the OFT than males, demonstrating sex-specific patterns of anxiety-like behavior. In the NSF test, SI rats showed an increased latency to consume food pellets and reduced food consumption compared to GH rats suggesting enhanced depressive-like behaviors, and males ate more overall indicating sex differences. In the CPP task, SI rats showed a trend toward spending more time in a chamber previously paired with alcohol compared to GH rats ($p = 0.05$). These findings suggest that prolonged social isolation beginning in adolescence enhances anxiety- and depressive-like behaviors, and may increase vulnerability to alcohol consumption in both sexes. Furthermore, the Long-Evans strain provides a useful model for investigating the neurobehavioral effects of adolescent isolation. These findings have important implications given the large number of adolescents that experienced social isolation during the COVID-19 pandemic.

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Poster

PSTR288: Adolescent Stress

Location: SDCC Halls B-H

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Program #/Poster #: PSTR288.04/Y15

Topic: G.03. Stress and the Brain

Title: Juvenile social isolation is associated with alterations in neurovascular markers in the hippocampus

Authors: *W. J. GORDIÁN-VÉLEZ, S. ORBETA, F. RANDI, Y. BAI, S. D. CROLL, G. BATISTA;
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Abstract: Social isolation and loneliness are public health problems affecting around 1 in 3 adults and 5-15% of adolescents. This type of chronic stress, especially during periods of heightened brain plasticity (i.e., early life, adolescence), increases the risk for physical and mental disorders. The mechanisms of how social isolation leads to changes in brain function and behavior are not fully understood. To address this knowledge gap, we used a model of post-weaning social isolation, in which C57BL/6J male mice are single-housed at P28 through adulthood (>P60). These mice exhibit anxiety-like behaviors and impaired sociability, as well as blunted serum corticosterone levels as adults. We have investigated the transcriptomic, molecular, and cellular mechanisms that potentially underlie the effects of this stress. Bulk RNA sequencing was used to profile the hippocampus, a region involved in cognition, learning, memory, social behavior, and with known sensitivity to stress. We identified differentially expressed genes (DEGs) associated with the neurovasculature and the blood-brain barrier. A top hit among these DEGs was intercellular adhesion molecule-1 (*Icam1*), which is expressed in endothelial cells and has a critical role regulating vascular permeability and immune cell trafficking. ICAM1 alterations in the hippocampus were validated in an independent mouse cohort, with significant reductions of ~37% and ~27% in mRNA (qPCR) and protein (Western blot), respectively. We also found ~25% less expression of the adhesion molecule CD31 and the tight junction protein claudin-5, suggesting broader neurovascular alterations. To better understand the mechanisms leading to these alterations, we investigated NF-κB signaling as a known regulator of neurovascular gene expression. We found a potential dysregulation of NF-κB in the hippocampus, characterized by significant reductions in its p65 and p50 protein subunits. Future studies will test our working mechanistic model by investigating whether modulating pathways associated with neurovascular changes can either reproduce stress-induced behavioral deficits in group-housed mice or rescue these deficits in isolated mice. Ultimately, this work could significantly expand our understanding of the neurobiology of stress, particularly the mechanisms involved in stress-induced neurovascular dysregulation.

Disclosures: **W.J. Gordián-Vélez:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals, Inc. **S. Orbeta:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals, Inc. **F. Randi:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals, Inc. **Y. Bai:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals, Inc. **S.D. Croll:** A. Employment/Salary (full or part-time): Regeneron Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals, Inc. **G. Batista:** A. Employment/Salary (full or part-time):

Regeneron Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Regeneron Pharmaceuticals, Inc..

Poster

PSTR288: Adolescent Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: G.03. Stress and the Brain

Support: NARSAD Young Investigator Grant from the Brain & Behavior Research Foundation awarded to C.A.V.
Schimmel Research Grant from Ohio Wesleyan University awarded to C.E.H.

Title: Determining the Effects of Early Adolescent Stress on Psychiatric-Related Behaviors and GABA_A Receptor Subunit Expression in C57BL/6J Adult Mice

Authors: *C. E. HAMMOND¹, K. R. SAIKALY¹, A. R. BIDDLE¹, J. KUBACEK¹, C. A. VADNIE²;

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Abstract: Anxiety and mood disorders are highly prevalent and often diagnosed in late adolescence or early adulthood, with stress being a major risk factor. However, limited studies focus on the long-term neurobiological and behavioral effects of adolescent stress in mouse models. Past work indicates that adolescent stress may increase anxiety- and depressive-like behavior and alter the expression of GABA_A receptor subunits in specific brain regions. In this study, we sought to determine if we could replicate work on the effects of adolescent stress on behavior and GABA receptor gene expression in adult mice. Male and female C57BL/6J mice were separated into stressed and unstressed groups (n = 7-8 of each sex). Stressed mice experienced 3 days of stressors (forced swim, restraint, and elevated platform stress) from postnatal day (PND) 25 to PND 27 during the light phase. Control mice remained undisturbed. We hypothesized that early adolescent stress would increase anxiety- and depressive-like behaviors in adult mice and alter GABA_A receptor subunit expression in brain regions implicated in regulating psychiatric-related behaviors. Behavioral testing started during early adulthood at PND 60. We used the open field (OF) and elevated plus maze (EPM) as measures of anxiety-like behavior, along with assays for depressive-like behavior. Brain tissue was collected at PND 69. Adolescent stress increased anxiety-like behavior in adult mice in the OF as measured by decreased center entries and center time (*p<0.05). However, stress had no impact on EPM behaviors or measures of depressive-like behavior. Thus, stress during early adolescence increases some anxiety-like behaviors in adult C57BL/6J mice. Results so far show that early adolescent stress decreased GABA_A α3 receptor subunit expression in the medial prefrontal cortex (mPFC) (p<0.001). These findings, similar to previous work, suggest that GABAergic

signaling in the mPFC plays a significant role in increased anxiety-like behavior after early adolescent stress. To further explore the effect of a longer period of adolescent stress, mice were exposed to 6 days of adolescent stress (forced swim, restraint, elevated platform stress, restraint, forced swim, and elevated platform) from PND 25 to PND 30. The mice were then examined for changes to a battery of psychiatric-related behaviors in adulthood starting at PND 60 using the OF, EPM, light-dark box, Y-maze, novel object test, and sucrose preference test. The results of this six day paradigm will allow us to examine the impact of the length of stress experienced in early adolescence and better understand its impact on psychiatric-related behaviors.

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Poster

PSTR288: Adolescent Stress

Location: SDCC Halls B-H

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Program #/Poster #: PSTR288.06/Z1

Topic: G.03. Stress and the Brain

Title: Genetic Background Modulates Anxiety-like Behaviors and Nicotine Sensitivity Following Adolescent Social Stress in BXD Mice

Authors: *C. NOVOA¹, T. J. GOULD²;

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Abstract: Adolescence is a period of increased sensitivity to environmental influences, which is marked by significant physiological changes and shifts in typical behaviors, such as increased social behavior, risk-taking, and the initiation of drug use. This stage is also associated with the onset of common psychiatric disorders, including anxiety, mood, and substance use disorders. Adolescents exhibit an amplified response to stress, and combined with the ongoing reorganization of neural circuits, this creates a window of biological plasticity where significant exposures can shape long-term health trajectories. Social stressors play a key role in contributing to negative psychiatric outcomes both during adolescence and later in life. Here we investigated the effects of adolescent social stress on anxiety-related behaviors and sensitivity to acute nicotine exposure in late adolescence. Using male and female mice from the recombinant inbred family BXD, we explored how sex and genetic background influence the interactions between juvenile stress and drug exposure. We observed substantial strain differences in spontaneous exploration in the elevated plus maze (EPM) and locomotor activity in the open field following acute nicotine exposure, where mice exhibited a biphasic hypolocomotor response. Additionally, exposure to social instability stress altered activity in the EPM and altered nicotine-induced locomotor responses in a strain- and phase-dependent manner. Furthermore, we observed that initial responses to nicotine were modulated by sex in addition to genetic background. These findings underscore the role of genetic factors in shaping mental health-related traits and drug

susceptibility during adolescence. Current bioinformatics analyses will leverage publicly available BXD mouse family data to further investigate the biological mechanisms underlying these results.

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Poster

PSTR288: Adolescent Stress

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Program #/Poster #: PSTR288.07/Z2

Topic: G.03. Stress and the Brain

Support: JSPS25K03022
JSPS 22K11528

Title: Development of a corporal punishment exercise model for investigating the negative impact of exercise under social stress on brain function

Authors: M. SHIMODA, R. TSUCHIDA, H. ONISHI, *T. NISHIJIMA;
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Abstract: [Background] Exercise is widely recognized for its positive effects on both physical and mental health, with notable benefits in the hippocampus. These effects are evident in children as well; for example, aerobic exercise interventions have been shown to improve hippocampus-dependent memory and enhance academic performance in adolescents. However, it is important to note that, depending on the conditions, exercise can also act as a stressor that impairs hippocampal function. Previous studies have demonstrated that intense exercise above the lactate threshold triggers stress responses, which in turn inhibit the positive effects of exercise on the hippocampus in rodents. It is conceivable that other factors, beyond exercise intensity, could also serve as potential stressors. Verbal and physical abuse during sports remain prevalent, and some victims of such abuse develop post-traumatic stress disorder. We therefore hypothesized that exercise combined with social stress would inhibit the beneficial effects of exercise on the hippocampus. However, no experimental animal model has yet been developed to investigate this hypothesis. [Purpose] The aim of this study was to develop an animal model of exercise under social stress that closely resembles corporal punishment in mice. [Methods] Male C57BL/6J mice were divided into four groups based on the presence or absence of exercise and social stress. The treadmill running intensity was set at a mild (10 m/min) to ensure that exercise alone did not induce a stress response. We employed a social defeat stress model, which is a well-validated approach designed to simulate the stress resulting from violent interpersonal experiences. The social stress group was subjected to social defeat stress with a retired ICR mouse the day before the acute exercise test, and exercised in the presence of the ICR mouse placed at the rear end of the treadmill. To assess whether exercise under these conditions induced stress responses, we conducted immunohistochemical analysis of neural activity in the

paraventricular nucleus (PVN) of the hypothalamus. [Results] Treadmill exercise under social stress significantly increased the density of c-Fos-positive cells in the PVN, despite the mild intensity of exercise. In contrast, exercise without social stress did not induce a similar response. [Conclusion] These results demonstrate that exercise combined with social stress, even at mild intensity, induces stress responses in mice. The experimental model established in this study will be valuable for future research investigating the effects of exercise under conditions of corporal punishment on hippocampal function.

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Poster

PSTR288: Adolescent Stress

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Program #/Poster #: PSTR288.08/Z3

Topic: G.03. Stress and the Brain

Support: Emmanuel College Research Funding

Title: Two-hit developmental stress decreases glucocorticoid receptor levels in mouse hippocampus when compared to maternal separation or adolescent social isolation alone.

Authors: *E. DEL MORO¹, Z. PALERMO¹, A. GEMOS¹, R. SULLIVAN¹, R. JUSTINIANO¹, M. P. LEUSSIS^{2,1};

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Abstract: Early adversity increases the likelihood of developing a psychiatric or mood-related disorder later in life and can alter the stress regulation pathway, but there is limited research examining the possible cumulative impact of multiple early adverse events during development. This study examined the long-term effects of a two-hit model composed of stressors during both childhood and adolescent development and evaluated the impact of these different stressors on mood and anxiety-related behaviors and glucocorticoid receptor (GR) levels in the hippocampus. Maternal separation was used to model early adversity and stressful events in childhood, while social isolation was used as a stressor in adolescence. Male and female CD-1 mice were exposed to either maternal separation (MS; PND 2-12), adolescent social isolation (ASI; PND 35-56), a combination of both (MS-ASI), or were controls (CON) that experienced no stress. Analyses of GR protein levels in the hippocampus following the developmental stress treatments showed that those mice experiencing two developmental stress hits, MS-ASI, had substantially lower GR levels in the hippocampus than mice in the other three groups (CON, MS or ASI) ($p < 0.001$). There were no sex differences ($p > 0.1$) and no stress condition x sex interaction ($p > 0.1$) for GR level in the hippocampus. GR levels in the cortex are in the process of being assessed. These findings point to a possible increase in stress system dysregulation, as higher GR levels in the hippocampus are known to help positively modulate hypothalamic pituitary adrenal (HPA) axis

activity. Impairments in the ability to regulate the HPA axis can in turn impact the likelihood of developing a mood or anxiety-related disorder. In fact, mice exposed to two-hits (MS-ASI) exhibited increased anxiety in an elevated plus maze test compared to control mice (CON) for both males and females ($p < 0.05$), suggesting the decrease in GR could be at least partially mediating this effect. Exposure to two-hits (MS-ASI) did not appear to impact measures of depression in either the forced swim test or sucrose preference test. Further, there was no overall impact of stress treatment on locomotor behavior in an open field. Future research on two-hit models of stress exposure should examine other psychiatric related behaviors and consider whether stress hormone levels are directly affected.

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Poster

PSTR288: Adolescent Stress

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Program #/Poster #: PSTR288.09/Z4

Topic: G.03. Stress and the Brain

Support: RSCA Grant (CSU, Chico)

Title: From Sugar to Substance: Adolescent Binge Eating Heightens Adult Morphine Reward and Withdrawal

Authors: *A. BARKLEY¹, P. REZAIE BOROON², S. BATES³;

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³Psychology, CSU Chico, Chico, CA

Abstract: Binge eating disorder (BED) is characterized by chronic overeating, as well as a feeling of loss of control during binge episodes. Substance Use Disorder (SUD) is characterized by chronic, relapsing drug use and users experience a negative emotional state when access to their drug of choice is denied. BED is similar to SUD as both disorders are characterized by recurrent binge episodes and involve similar neuronal pathways. Adolescence is an important developmental period consisting of extreme physical, psychological, and neurological changes. Many mental health disorders start to manifest during this period, including BED. Additionally, 17.9% of adolescents misuse opioids. However, despite the prevalence of BED and substance use in adolescents, there is a lack of research in this area. Our laboratory has sought out to explore the relationship that BED has on SUD because of the similarities. Previous findings in our laboratory suggest that binge eating in adolescents potentiate cocaine reward and impulsivity in adult mice. To further explore the relationship between SUD and BED, this study examines the effects of binge eating on response to morphine. To test the level of morphine reward, a Conditioned Place Preference (CPP) paradigm was implemented. Following CPP, an elevated plus maze test and a naloxone challenge were used to assess level of withdrawal. In a separate

group of animals, immunohistochemistry (IHC) was conducted, in which, brain slices from animals that were exposed to the binge-eating protocol are stained with a fluorescent dye and examined under a confocal microscope. Results from CPP show increased morphine reward in the female binge eating condition. This suggests that female adolescents with BED may be more vulnerable to the rewarding effects of morphine. Additionally, the binge eating condition showed greater withdrawal-induced behaviors, regardless of sex. This finding suggests that adolescents with BED may be at a greater risk for experiencing greater withdrawal following morphine use. Additionally, results from the IHC demonstrates an increase in activity in the hypothalamus in binge eating mice. This suggests that binge eating leads to hyperactivity in the hypothalamus. In humans, overactivation of the hypothalamus can increase the chances of developing a mood disorder, sleep disorder, or obesity. Our results suggest that adolescent BED potentiates morphine reward and withdrawal, which suggests an increased vulnerability to an opioid addiction in individuals with BED. Understanding the risks associated with adolescent binge eating will allow for future studies on treatment and intervention strategies for BED.

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Poster

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Topic: G.03. Stress and the Brain

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Title: Sex-specific neural adaptations to adolescent trauma-induced binge eating in a rat model

Authors: *J. SIERRA¹, T. SIMON², P. ONTIVEROS-ANGEL², V. WILLIAMS², A. M. WILLIAMS², B. NOARBE³, A. OBENAU⁴, J. D. FIGUEROA²;

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Abstract: Binge eating is the most common eating disorder symptom, impacting more than 40 million individuals globally. Recent studies suggest that disruptions in brain circuits responsible for food reward processing may contribute to the development of binge eating. These reward-regulating regions undergo protracted maturation throughout adolescence, a period marked by heightened vulnerability to environmental insults. Among these, exposure to traumatic stressors during adolescence has been associated with reduced volume and altered function in key reward-related brain regions. Despite these observations, the precise neural mechanisms connecting adolescent trauma to dysregulated eating remain unclear. In this study, we investigated how adolescent trauma influences the developmental trajectories of reward circuits implicated in binge eating.

To test our hypothesis, we developed a Lewis rat model of adolescent trauma-induced binge eating. At postnatal day 21 (PND 21), male and female rats were assessed for baseline acoustic startle response and body weight, then stratified into trauma-exposed and control groups to ensure balanced allocation across conditions. Trauma-exposed rats endured a 31-day protocol (PND30-60) consisting of two predator exposures and daily social instability. To evaluate binge eating behavior, adult rats were provided a Western-like high-saturated fat diet (WD) for 24 hours, followed by 6-day access to a low fat control diet (CD) for three weeks (cycle 1-3). WD consumption was measured at 2.5 and 24 hours after WD (re)introduction. At the end of the study, brains were prepared for ultrahigh resolution diffusion-MRI (dMRI, 17.6T) and immunofluorescence.

Our results demonstrate that trauma-exposed female rats displayed elevated food intake compared to trauma-exposed males (cycle 1) and unexposed females (cycle 3). Furthermore, female subjects exhibited a blunted acoustic startle response following trauma exposure compared to controls. We calculated region-specific z-scores for fractional anisotropy (FA), a dMRI metric that reflects white matter microstructural organization, influenced by factors such as axonal density, fiber coherence, and myelination. FA analyses demonstrated selective trauma-induced structural differences in hypothalamic and reward regions (increase in males, decrease in females) compared to controls. Our findings demonstrate the impact of early trauma on novel neurocircuits implicated in binge eating. Current work is focused on evaluating neuronal activity and neuroendocrine markers that may offer avenues to identify neuropathological targets of susceptibility for binge eating.

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Poster

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Title: Adaptative and dysfunctional behaviors following traffic noise exposure in adolescent rats: effects of sex and enriched housing - preliminary results

Authors: *G. CORSI¹, L. D'ALESSIO^{2,3}, L. R. GUELMAN^{1,3}, S. J. MOLINA¹;

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Abstract: Central nervous system (CNS) development is a complex process that can be affected by aversive environmental stimuli. In particular, white noise exposure can affect the hippocampus (HC), leading to cognitive deficits and changes in HC-related behaviors in experimental animals. However, little is known about the effects of traffic noise on the HC, a stimulus to which children and adolescents living in large cities are frequently exposed. Moreover, environmental enrichment (EE), a non-pharmacological neuroprotective strategy, has shown efficacy in preventing different CNS injuries. Thus, the aim of this work was to assess the effects of traffic noise on HC-related behaviors in adolescent rats and to determine whether these alterations can be prevented by EE housing. Adolescent Wistar rats of both sexes were exposed to traffic noise (2h/day, for 5 days). A subgroup of rats was housed in an EE after weaning, while others remained in standard cages. Finally, behavior was assessed using open field, inhibitory avoidance and elevated plus maze tasks devices. Results showed that traffic noise improved habituation memory in males, whereas it impaired performance in females, changes that were prevented by EE housing. However, associative memory remained unaffected. Male and female rats housed in EE, exposed or not to noise, exhibited increased exploratory activity and decreased anxiety-related behaviors. Finally, EE housing alone reduced risk-assessment behaviors in females, whereas an increase was found when combined with noise exposure. In contrast, the combination of EE housing and noise exposure led to a reduction in risk-assessment behaviors in males. These findings suggest that traffic noise exposure and EE housing might induce different HC-related behavioral changes in a sex-specific manner. In males, noise may promote faster habituation to novel environments, indicating an adaptative response. In contrast, females seem more vulnerable to its negative effects, showing deficits in habituation memory. Moreover, EE housing could promote beneficial effects by preventing noise-induced memory deficits, and reducing anxiety-like behaviors, thereby increasing exploration. However, EE housing might also reduce defensive behaviors that potentially might increase their exposure to threats. Interestingly, concurrent noise exposure and EE housing may enhance caution in females, suggesting a compensatory response to stressful situations. In summary, animal responses to aversive stimuli like traffic noise could be influenced by prior experience and context interactions, showing sex-specific effects that may be either adaptive or detrimental.

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Poster

PSTR288: Adolescent Stress

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Program #/Poster #: PSTR288.12/Z6

Topic: G.03. Stress and the Brain

Support: NIH Grant HD091376

Title: Impact of pubertal stress on behavioral and physiological responses to later stressors and the hypothalamic transcriptome

Authors: *M. M. NICODEMUS¹, L. A. M. LUTHER¹, M. A. FOUAL¹, I. M. LINTON¹, K. E. MORRISON²;

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Abstract: Women are at increased risk of being diagnosed with an anxiety or mood disorder, although this is not uniform across the lifespan. Women are at a particular risk during the time before and shortly after birth, known as the peripartum period. One risk factor for such disorders is exposure to stress. In humans, both preconception and gestational stressors have been associated with an increased risk of peripartum mental health issues. We have previously established a translationally relevant mouse model, where pubertal stress led to a disrupted glucocorticoid response to stress during the peripartum window and lasting alterations in the transcriptional and epigenetic landscape within the paraventricular nucleus of the hypothalamus (PVN). However, whether the molecular and physiological results are related to altered behavior and the involvement of brain regions beyond the PVN have yet to be determined. Here, we address these gaps by examining how stress during puberty and later stress during pregnancy might interact to shape such outcomes. We utilized differing levels of stress during pregnancy to examine how pubertal stress alters the behavioral, hormonal, and neural responses to a “second hit” of stress during pregnancy. Female mice were exposed to chronic variable stress (CVS) during puberty (postnatal days 21-34) and were bred to become pregnant in adulthood. During pregnancy, factors that are known stressors were systematically varied: housing (pair vs single housing), repeated testing (1 vs 3), and resources available (standard vs limited bedding and nesting). Mice were tested for maternal behavior (pup retrieval task), depressive- and anxiety-like behavior (sucrose preference test, splash test, open field test, social interaction test), and corticosterone response to pup retrieval in late pregnancy. Initial results show that control and pubertally stressed pregnant mice respond differently to singly housing and repeated testing. CVS females exposed to increased stress during pregnancy show a longer latency to engage with pups, but faster retrieval once maternal behavior sequence begins. The corticosterone response was also influenced by both pubertal stress and degree of stress during pregnancy. Ongoing work is examining depressive- and anxiety-like behavior. We are also examining the transcriptome of two key hypothalamic regions, the medial preoptic area (mPOA), which is involved in the expression of maternal behavior, and the PVN, which regulates the hormonal stress response. This work will add to our understanding of the molecular underpinnings and the complex risk factors for mood and anxiety disorders in the lifespan of women.

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Poster

PSTR288: Adolescent Stress

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR288.13/Z7

Topic: G.03. Stress and the Brain

Support: NIH Grant HD091376

Title: Effects of preconception stress on maternal behavior and offspring behavior and brain development

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Abstract: Women who experience early life adversity have an increased risk for mood and anxiety disorders in adulthood. This risk is further increased if they become pregnant. Perinatal mood and anxiety disorders are associated with disrupted maternal behavior, which may have lasting effects on offspring. The lasting consequences of preconception early life stress on maternal care and offspring behavior and brain are not well understood. We hypothesized that maternal preconception stress would alter offspring behavior in an open field test, which probes risk-taking behavior, and social preference, which examines preference for social interaction and social novelty. We exposed female mice to two weeks of chronic variable stress (CVS) from postnatal days (PN) 21-34 or to no stress (Control). They were undisturbed until 10 weeks old, when they were all bred to become pregnant. One male and one female offspring from each dam underwent open field and social preference tests at PN34 (adolescence) and 10 weeks (adulthood). In adolescence, mice of CVS dams spent more time in the center of the open field test, though no differences were observed in adulthood. To address whether this transmission of preconception maternal stress to offspring is via prenatal versus postnatal mechanisms, we generated another cohort that underwent cross fostering. Ongoing work is characterizing the behavior and brains of the offspring. On the day of birth of their litter (PN1), pups will be counted, sexed, and weighed and swapped between CVS and Control dams. In adolescence (PN34) or in adulthood, one male and one female from each litter will undergo open field and social interaction tests. After behavior, brains will be collected from the offspring that underwent behavior and a matched naive animal. Regions including the prefrontal cortex, amygdala, and hypothalamus will be examined for relevant gene expression. The results will further our understanding of how stress can be passed down through generations and the mechanisms by which maternal preconception stress may alter offspring development.

Disclosures: I.M. Linton: None. M.M. Nicodemus: None. K.E. Morrison: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.01/Z8

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R01AG077731

Title: Multi-modal analysis of microvascular diameter across cortical layers and corpus callosum

Authors: *S. NAIR¹, S. STAMENKOVIC², G. GÜRLER², A. Y. SHIH^{2,3,4};

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Abstract: Control over the size of blood vessels is a key factor in the proper regulation of blood flow to the brain. Capillaries confer the highest resistance to blood flow because of their small diameter. In prior studies, we used deep in vivo two-photon imaging in mice to show that capillaries of the deeper tissues (cortical layer 6 and corpus callosum) were on average 11.4% larger in diameter and supported higher rates of flow than upper cortical layers ($3.5 \pm 0.6 \mu\text{m}$ in mid-cortex vs. $3.9 \pm 0.6 \mu\text{m}$ in deep layers; $p < 0.0001$). This was presumably necessary to ensure sufficient blood flow to more vulnerable deep tissues distant from the perfusion source from the pial surface. However, in vivo imaging at greater depths is affected by optical aberrations that can make vessels appear larger than in reality. We hypothesized that capillaries are indeed physically larger in deep tissues. To address this hypothesis we examined capillary diameters in volume electron microscopy data and histological preparations from post-mortem mouse brain tissues.

We first used the publicly available Machine Intelligence from Cortical Networks (MICrONS) dataset, which is a cubic millimeter volume electron microscopy reconstruction of a mouse visual cortex. We assessed several capillary features, such as number of endothelial tight junction number, capillary diameter, and capillary lumen area between mid-cortical regions (including layer 2/3 and 4) and compared these to capillaries in deeper layers (including layer 6 and superficial corpus callosum). Capillary diameter and area was, on average, 7.3% larger in deep tissues compared to superficial cortex ($4.0 \pm 1.1 \mu\text{m}$ in mid-cortex vs. $4.3 \pm 0.9 \mu\text{m}$ in deep layers; $p = 0.04$). Capillaries in deep tissues tended to have more tight junctional strands, suggesting that they may be composed of more endothelial cells. As expected, deep capillaries also had greater cross-sectional areas consistent with larger diameters.

We then used a separate histological dataset to examine the same question. Mouse brain tissues were sectioned and stained with fluorescent lectin to label the vascular wall. Mid-cortical layers and deeper layers in the sensory cortex were then imaged by two-photon microscopy, and capillary diameters in these images were measured using Vasometrics. This data also revealed that capillary diameters in deep layers were, on average, 15.5% larger than mid-cortex ($3.8 \pm 0.8 \mu\text{m}$ in mid-cortex vs. $4.5 \pm 0.9 \mu\text{m}$ in deep layers; $p = 0.0001$). Overall, independent analyses of capillary diameters from datasets insensitive to depth related imaging artifacts were consistent with results from in vivo imaging. They confirm capillary size differences across layers.

Disclosures: S. Nair: None. S. Stamenkovic: None. G. Gürlér: None. A.Y. Shih: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.02/Z9

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R01NS078168
NIH Grant U19NS128613
NIH Grant T32NS115667
NIH Grant T32GM108563
American Heart Association Grant 24PRE1201066
American Heart Association Grant 25PRE1371919

Title: Roles of Norepinephrine and Pyramidal Neuron Activity in Controlling Cerebral Hemodynamics Across Arousal States

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Abstract: Cortical arteries exhibit larger dilations and constrictions during sleep than wakefulness. These arterial dynamics are thought to drive periarterial pumping, displacement of cerebrospinal fluid (CSF) that helps clear waste from the brain. While norepinephrine-mediated vascular dynamics during sleep have been linked to CSF clearance, the combined roles of neuromodulators and local neural activity remain unexplored. In this study, we explored how norepinephrine (NE), acetylcholine (ACh), and cortical pyramidal neurons jointly regulate vascular dynamics in a state-dependent manner. Using fiber photometry, we measured cerebral blood volume (CBV) via liver-secreted fluorescent protein mAlb-mScarlet, neuromodulator levels via GRAB-NE and GRAB-ACh biosensors, and pyramidal calcium activity (CaMKIIa-GCaMP7s) in the barrel cortex of head-fixed mice during wakefulness, NREM, and REM sleep. Sensory stimulation of the whiskers caused vasodilation and an increase in NE, ACh, and CaMKIIa-GCaMP. There was an increased blood volume during the awake-to-NREM sleep and NREM-to-REM sleep transitions. Cortical CaMKIIa-GCaMP activity was strongly correlated with the CBV. These CBV increases were more prominent than sensory stimulus-induced changes. ACh levels decreased during awake-to-NREM sleep transition but increased during the NREM-to-REM sleep transition. NE levels decreased during awake-to-NREM sleep and NREM-to-REM sleep transition. Awakening from either sleep stage was associated with large increases in NE and ACh levels, tied to sharp decreases in CBV and pyramidal calcium activity. Optogenetic stimulation of locus coeruleus (LC)-NE neurons drives pupil dilation and increases norepinephrine release in the cortex while causing vasoconstriction. The positively correlated pyramidal neuron activity and blood volume became anticorrelated during optogenetic activation of the LC, showing vasoconstriction while the pyramidal calcium activity was high. This shows that NE activity can override local cortical activity and drive vasomotion, opposing local cortical activity. Our findings suggest that local pyramidal neuron activity may have a key role in maintaining vasomotion, but is not always required, and NE modulation can drive arterial pulsation, overriding local cortical activity.

Disclosures: M. Hossain: None. M.R. Wenke: None. J. Flores: None. D.I. Greenawalt: None. K.L. Turner: None. P.J. Drew: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.03/Z10

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R01NS078168
NIH Grant U19NS128613
NIH Grant T32NS115667
AHA Grant 24PRE1201066

Title: Two-photon imaging of norepinephrine and arteriole dynamics in sleep-deprived mice

Authors: *M. WENKE^{1,2}, M. HOSSAIN¹, P. J. DREW¹;
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Abstract: One important feature of sleep is global vasodilation of cerebral arteries with periodic pulsations that drive cerebrospinal fluid flow and promote waste clearance from the brain. Norepinephrine (NE) release from cortical axonal projections originating from the locus coeruleus (LC) precedes vasoconstriction and arousal from sleep, as well as the pulsations throughout sleep. LC dysfunction is a hallmark of many neurologic conditions, including chronic sleep insufficiency. Sleep deprivation increases sleep drive, but it also increases the magnitude and frequency of NE release from the LC, correlating with sleep fragmentation. However, the effect of sleep deprivation-induced LC hyperexcitability on arterial dynamics throughout sleep is unknown. This study used two-photon imaging to observe cortical NE release alongside arterial diameter in the somatosensory cortex of head-fixed C57BL/6J mice as they sleep. We visualized local NE tone around single penetrating arterioles through a thinned-skull window over the somatosensory cortex by expressing a G protein-coupled Receptor-Associated Biosensor (GRAB_{NE-2m}). We monitored non-rapid eye movement (NREM) and rapid eye movement (REM) sleep states using electrocorticography, electromyography, pupillometry, and motion sensors. Preliminary results show that NE around penetrating arterioles was highest during wakefulness, decreased during NREM accompanied with vasodilation, and was lowest during REM sleep. Immediately prior to arousal from sleep, local NE levels rapidly increased back to waking baseline levels, followed by vasoconstriction. Sleep deprivation increased NE release and altered the frequency of arteriole pulsations throughout NREM sleep. Together these experiments suggest that LC hyperexcitability after sleep deprivation changes arteriolar dynamics throughout NREM sleep and at arousal events.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.04/Z11

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH NS121543

Title: Modulation of Neurovascular Coupling Through 5-HT_{2A} Agonism

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Abstract: The brain relies on a tightly regulated and continuous blood supply to meet its moment-to-moment metabolic demands, ensuring that regions with heightened neuronal activity receive an adequate influx of oxygen and nutrients. Central to this process is neurovascular coupling (NVC), in which cerebral blood flow increases in response to increased local synaptic activity. While neurons are considered the essential drivers of this process, astrocytes—glial cells that form intricate networks with both neurons and blood vessels—also play a critical role in coordinating these vascular responses. Although several neurotransmitters have been implicated in NVC, an important but often-overlooked aspect is the role of neuromodulators. Serotonin (5-HT), a key neuromodulator, is a robust vasoconstrictor in peripheral systems, but its effects on cerebral vascular tone and NVC are less understood. Serotonergic neurons project to various brain regions, and serotonin receptors located on astrocytes, neurons, vascular smooth muscle cells, and endothelial cells further complicate this signaling pathway. The interaction between serotonin and components of the neurovascular unit may contribute to the spatial-temporal coordination of blood flow across different brain regions. Activation of serotonin 2A receptors (5-HT_{2A}s) has shown therapeutic promise for mental illnesses such as depression, with psychedelics—5-HT_{2A} agonists—demonstrating the ability to promote cortical structural and functional neuroplasticity. However, their potent vasoactive effects have largely been overlooked. We aim to investigate the contributions of psychedelic 5-HT_{2A} receptor agonist, 2,5-Dimethoxy-4-iodoamphetamine (DOI), to neurovascular coupling in the cerebral cortex of awake mice. We assessed the effects of DOI on NVC using two-photon imaging in awake mice under resting-state and stimulus-evoked conditions. A craniotomy was performed over the barrel cortex. Mice were trained on a treadmill and imaged after two training sessions. DOI was administered either via intraperitoneal injection or topical perfusion. Astrocyte Ca²⁺ dynamics and vascular responses were measured. We found that DOI differentially modulated astrocyte Ca²⁺ responses and vascular activity, during resting-state and stimulus-evoked conditions. DOI influence the spatial and temporal dynamics of neurovascular coupling. Our findings suggest that DOI modulates neurovascular coupling by activating 5-HT_{2A} receptors expressed in various cells of the neurovascular unit, highlighting the complex role of serotonin in regulating brain blood flow.

Disclosures: M. Kim: None. C.T. Tran: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.05/Z12

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: American Physiology Society Summer Undergraduate Research Fellowship

Title: Adenosinergic modulation of brain vasculature in awake behaving mice

Authors: *J. D. CLAYPOOL¹, O. MORRIS¹, Q. ZHANG²;

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Abstract: Various pathologies, such as infections, autoimmune disorders, and neurodegenerative diseases, are linked to neuroinflammation, which can lead to cognitive impairments and even neuronal cell death. These effects are often the result of ischemia of the brain tissue. The neurotransmitter adenosine has long been recognized of playing both pro- and anti-inflammatory roles in the brain as well as influencing the cerebral vasculature. The relationship between inflammation and adenosine is important when evaluating how pathologies can impact brain hemodynamics and the blood-brain barrier. While there are theories behind these interactions, the dynamics under normal physiological conditions have not been fully examined. Here, we investigated the effects of acute caffeine injection, a nonselective adenosine receptor antagonist, on neurovascular coupling by measuring neuronal and hemodynamic responses to voluntary locomotion and whisker stimulation in awake, head-fixed mice. Widefield fluorescence imaging was used to measure cerebral blood volume (CBV), oxygenation and metabolism activity through a polished, reinforced thin-skull window, while neural activity (electrocorticogram) was measured in the contralateral cortex concurrently. Both neural activity and hemodynamic signals were collected before and after systemic administration of caffeine and vehicle controls. Compared to vehicle controls, both resting and locomotion/whisker stimulation-evoked neuronal response were not changed. There was a clear vasoconstriction at resting state, and CBV responses to locomotion/whisker stimulation were speed up by caffeine injection. These results support a role of adenosine in the regulation of hemodynamics. Future studies will be conducted to evaluate extracellular adenosine levels using G-protein coupled receptor activation-based adenosine (GRAB_{Ado}) sensors and evaluate brain hemodynamic responses during induced systemic inflammation. These experiments will provide us with a better understanding of how inflammation impacts cerebrovascular hemodynamics, and to reveal more about the role adenosine may play in these responses.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.06/AA1

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Brain Initiative funding RF1NS124778
NIH Brain Initiative funding R01NS122904
NIH Brain Initiative funding R01NS120594
NSF grant 2123971
S10 instrument grant S10 MH124733-01

Title: Detecting Vaso-Dynamic Signal Propagation through the Cortex of Awake Mice Using 14T CBV fMRI

Authors: X. LIU¹, D. HIKE¹, X. A. ZHOU¹, Y. JIANG¹, J. DUCKWORTH², D. KLEINFELD^{2,3}, *X. YU¹;

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Abstract: Vasomotion, the spontaneous rhythmic oscillation of arteriole diameter, is well characterized in rodents via two-photon microscopy in upper cortical layers^{1,2}. Studying vasomotion was extended across the full cortical depth with high-resolution single-vessel CBV fMRI³. We applied awake mice whole-brain high-resolution (100×100×200 μm; TE=6.2 ms; TR=1 s; 5 mice 46 trials) and cortex-specific fast (200×200×200 μm; TE=4 ms; TR=0.125 s; 19 mice 111 trials) CBV fMRI to characterize global propagation of specific oscillation bandwidths based on coherence phase gradients derived from singular value decomposition (SVD). We scanned 24 C57BL/6J mice at 14T by multi-slice 2D EPI following MION injection (25 mg/kg) (Fig 1A). Data was processed with AFNI^{4,5} and segmented by the Allen Brain Atlas. Space-frequency SVD^{6,7} shows dominant spatial components as magnitude and phase maps for vaso-dynamic analysis (Fig 1C D). Cortical phase gradients were linearly fitted in 3 mm windows in three directions (AP, LR, IS). Propagation speed was calculated as $c = 2\pi f / k$; f = center frequency, k = phase gradient. High-resolution CBV fMRI with ~0.1 Hz space-frequency coherence peaks showed complex vasomotion patterns over hours in 31 of 46 trials, maybe reflecting brain state-dependent neuromodulation (Fig 1B-D). 8 Hz CBV fMRI estimated coherence phase gradients over three axes (tangential: AP, LR; radial: IS) (Fig 1E). PCA was applied to group-level speed maps to see dominant phase patterns across center frequencies. Three spatial patterns showed primary vasomotion dynamics, explaining ~60% variability (Fig 1F). A one-sample t-test of spherical-coordinate speed vectors showed faster propagation near large sinuses and opposite directions in frontal cortex (Fig 1G). The same analysis was applied to visual/whisker-stimulated BOLD fMRI⁸ for validation, confirming propagation from activated visual/barrel cortices to other areas (Fig 1H I). Our work demonstrates that high-resolution and fast CBV fMRI captures vaso-dynamic patterns, highlighting cortical neurovascular dynamics in awake mice.

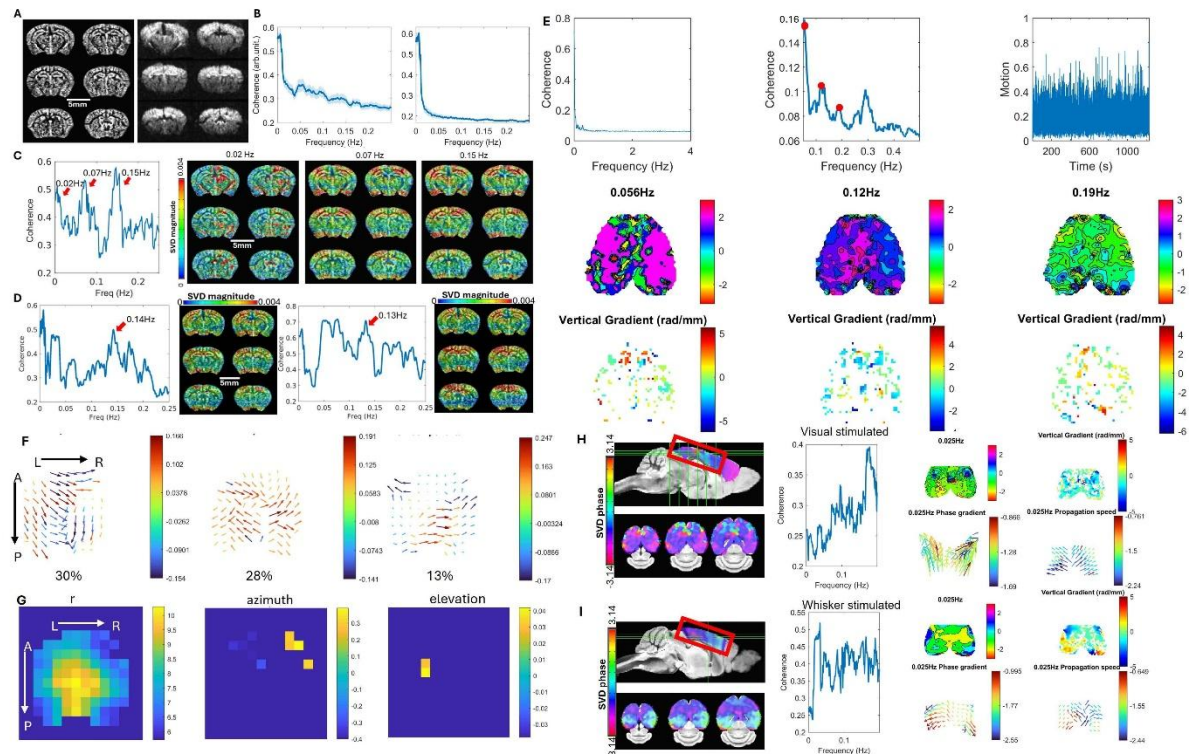


Fig 1 A) Anatomical (left) and CBV EPI (right) images. B) Averaged spatial coherence with peaks at 0.05–0.2 Hz (left) and no features (right). C) Spatial coherence with peaks at 0.02, 0.07, and 0.15 Hz and corresponding magnitude. D) Coherence and magnitude maps from the same animal at different times. E) Coherence, motion trace, tangential phase, and radial phase gradient maps at 0.056, 0.12, and 0.19 Hz. F) Top 3 PCA components (30%, 28%, 13%). Radial direction encoded by sign: positive from inferior to superior, negative in the opposite direction. G) Mean amplitude, azimuth, elevation ($p < 0.001, 0.05, 0.05$). H&I) Phase map, coherence, tangential phase, radial phase gradient, phase gradient and propagation speed patterns at 0.025 Hz (visual(H)/whisker(I) stimulation frequency).

Disclosures: X. Liu: None. D. Hike: None. X.A. Zhou: None. Y. Jiang: None. J. Duckworth: None. D. Kleinfeld: None. X. Yu: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.07/AA2

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Title: Endothelial calcium signalling regulates capillary blood flow dynamics and vessel tone in the cerebral microvasculature

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¹Div. of Med. Sci., ²Island Med. Program, Univ. of Victoria, Victoria, BC, Canada

Abstract: Endothelial calcium (Ca^{2+}) signalling is a key regulator of vascular tone, yet its *in vivo* role in controlling cerebral microvascular function remains incompletely understood. Using *in vivo* two-photon microscopy in transgenic mice expressing GCaMP8m selectively in cerebral endothelial cells, we assessed the effects of acutely elevating Ca^{2+} through DREADD-mediated

activation. Activation of hM3Dq receptors with deschloroclozapine (DCZ) resulted in a robust and sustained elevation in endothelial Ca^{2+} levels compared to saline, as quantified by the peak amplitude, half-width, and area under the curve (AUC) of $\Delta F_0/F_0$ signals. This Ca^{2+} elevation was observed across multiple capillary branch orders, demonstrating widespread activation of Ca^{2+} signalling in the capillary network by DCZ. Functionally, DCZ injection induced a rapid and significant increase in capillary red blood cell (RBC) flux, measured within 2 minutes post-injection, indicating enhanced blood flow. This effect was validated by laser Doppler flowmetry, which demonstrated a significant increase in cortical perfusion following DCZ, as indicated by changes in peak amplitude, half-width, and area under the curve (AUC) of the Doppler signal compared to saline controls. Concomitant with increased Ca^{2+} and flow, we observed dilation in capillaries, penetrating arterioles, and ascending venules. Vessel diameter changes were quantified across vascular segments and were significantly greater after DCZ injection compared to saline. Importantly, a positive correlation was found between the magnitude of Ca^{2+} transients and vessel dilation during the first 200 seconds post-injection, suggesting that Ca^{2+} elevations directly contribute to the dynamic regulation of vessel tone. Additionally, we observed that spontaneous increases in endothelial Ca^{2+} coincide with recanalization of stalled capillaries. Ongoing DCZ experiments aim to determine whether temporally elevating Ca^{2+} is sufficient to clear existing stalls or minimize their occurrence altogether. These findings highlight a key role for endothelial Ca^{2+} signalling in the regulation of cerebral blood flow and microvascular dynamics *in vivo*. Ongoing work is examining the spatial organization and timing of these Ca^{2+} responses across vascular compartments. Together, these results establish endothelial Ca^{2+} as a rapid and powerful modulator of cerebrovascular function.

Disclosures: D.A. Hancock: None. C.E. Brown: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.08/AA3

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH U19 NS123717
NIH R35 NS097265
AHA 906077

Title: The microvascular architecture in the arteriole-to-capillary transition zone enables local control of cortical perfusion

Authors: *X. JI^{1,3}, L. BAI⁴, Y. ZHAO⁴, K. WANG⁴, D. KLEINFELD^{1,2};

¹Dept. of Physics, ²Dept. of Neurobio., Univ. of California San Diego, La Jolla, CA; ³Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ; ⁴Inst. of Neuroscience, CAS, Shanghai, China

Abstract: Cerebral blood flow exhibits dynamics across a wide range of physiologically relevant temporal scales as it perfuses a vascular network spanning several orders of magnitude in spatial scale. The precision and robustness of flow control are strongly constrained by network architectures and physiological fluctuations, yet our systematic understanding of flow regulation has been hindered by the lack of conceptual and technical tools. To address this, we first theoretically studied the non-perturbative, network-level flow response to changes in hydrodynamic resistance in individual vessels. Applying these analytical results to anatomically accurate vascular connectomes revealed a weak, localized, and heterogeneous flow response in adjacent vessels. We then developed a model-based tracking framework that leverages network structure, along with computational tools to analyze blood cell dynamics imaged by confocal light-field microscopy. This experimental approach enabled the reconstruction of a vascular network comprising over 3,000 interconnected vessel segments and the tracking of over one million cell detections at 2- μ m spatial and 14-ms temporal resolution. The resulting trajectories captured single-cell dynamics across three orders of magnitude in speed and revealed substantial low-frequency variations in the flux of red blood cells across microvessels in the resting state. Combining theoretical results and high-throughput measurements, our analysis suggested that contractile pericytes are optimally positioned for regulation of flow within the first three branches of microvessels that emanate from penetrating arterioles, i.e., within the tree-like architecture of the arteriole-to-capillary transition zone. We further uncovered long-range, anti-correlated fluctuations in flux through spatially separated microvascular segments that are fed by a common source. Lastly, we quantified phase separation in the three-dimensional cerebral microvascular network at single-cell resolution. This revealed a stochastic partition ratio with a strong nonlinear dependence on local hemodynamics. Collectively, these results highlight the competition between optimality and complexity in constraining the control of brain microcirculation.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.09/AA4

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant U19 NS123717

Title: Characterizing how intrinsic vasomotion shapes pial arterial dynamics using rhythmic sensory stimulation and genetic perturbation of neurovascular coupling

Authors: *J. DUCKWORTH¹, T. BROGGINI^{3,4}, K. CHHABRIA¹, S. IYENGAR⁵, M. NELSON⁶, D. KLEINFELD^{1,2};

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Cancer Inst., Goethe Univ. Frankfurt, Frankfurt am Main, Germany; ⁵Dept. of Statistics, Univ. of Pittsburgh, Pittsburgh, PA; ⁶Dept. of Pharmacol., Univ. of Vermont, Burlington, VT

Abstract: The pial arteriole network spans the surface of neocortex and dynamically delivers blood to underlying brain cells. These arterioles exhibit intrinsic ~ 0.1 Hz changes in diameter that are modulated through electrical communication from both neighboring arterioles and underlying neurons. Here we test how intrinsic vasomotion shapes the pial arterial representation of spectrally varied neural activity. We used wide-field imaging to simultaneously measure arteriole diameter, via expression of GCaMP8.1 in arterial smooth muscle cells, and pan-neuronal activity, via expression of jRGECO1a in excitatory neurons, across the murine cortical mantle in awake head-fixed mice. We confirm that, during resting-state, pial arterioles undergo broadband oscillations near 0.1 Hz, reflecting intrinsic vasomotion with noisy neural input. During periodic visual and vibrissae sensory stimulation, we found that the neural signal is resonantly amplified by the arterioles at frequencies near the intrinsic vasomotor frequency. Moreover, we find that the phase of the arteriole response is a nonlinear function of frequency and is well described by a forced oscillator model. In additional experiments to explore arteriole resonance, localized optogenetic rostral ventrolateral medulla (RVLM) activation near the intrinsic vasomotor frequency elicited arteriole entrainment with comparable amplitude to that of vibrissae stimulation. When presented in competition with simultaneous vibrissae stimulation, the stimulation closest to the vasomotor frequency entrained preferentially more cortical area. To identify which components of arteriole activity are inherited from underlying neural activity, we inhibited neurovascular communication via inducible genetic knockout of the inward rectifier potassium channel Kir2.1 in vascular endothelial cells. We found that during knockout the arteriole amplitude in response to sensory stimulation is reduced, and its peak time is delayed, consistent with ablation of Kir2.1-mediated regenerating hyperpolarization. Additionally, noise in resting-state vasomotor oscillations is reduced, evidenced by sharpening of the vasomotor spectral peak. More generally, we probe the extent to which resting-state and driven arteriole activity is altered during endothelial Kir2.1 knockout, with the goal of providing mechanistic and quantified bounds for which neural activity can be inferred from vascular signals in the brain.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.10/AA5

Topic: D.02. Ischemia

Support: NIH grant R61NS137365

Title: In vivo chemogenetic control of brain pericytes as a model of cerebral hypoperfusion

Authors: T. MCGILLIS¹, S. XIA³, Z. ZHAO⁴, *A. Y. SHIH²;

¹Ctr. for Developmental Biol. and Regenerative Med., Seattle Children's Res. Inst., Kent, WA;

²Seattle Children's Res. Inst., Seattle, WA; ⁴Physiol. and Neurosci., ³USC, Los Angeles, CA

Abstract: Cerebral hypoperfusion leads to degradation of brain structure and function in Vascular contributions to cognitive impairment and dementia (VCID). Neuropathological studies have shown that brain capillaries, which are lined by pericytes, undergo pathological changes in VCID. However, little is known about how capillary-level changes contribute to tissue pathology or cognitive/behavioral deficits.

There are currently no strategies to promote capillary-level flow resistance that contributes to hypoperfusion. To address this gap, we have developed a new mouse model for chemogenetic stimulation of CNS pericytes. The expression of Gq-DREADD was regulated under the CNS pericyte-specific promoter, Atp13a5. Gq-DREADD activation through acute administration of the potent, blood-brain barrier permeant DREADD agonist, deschloroclozapine (DCZ), resulted in robust capillary constriction. Capillary constriction and blood flow reduction was observed in somatosensory cortex using in vivo two-photon imaging, and was not seen in littermate wild-type controls or in vehicle-treated Gq-DREADD expressing mice.

In ongoing work, we are testing whether chronic administration of DCZ through dosed drinking water models aspects of chronic cerebral hypoperfusion, including long-term blood flow reduction, neuroinflammation, blood-brain barrier disruption, and behavioral or cognitive deficits. DCZ was delivered through water at 0.3 ug/mL, 3 ug/mL or 0 (vehicle). Mice were weighed regularly to ensure DCZ was not associated with adverse health outcomes. Following 2-4 weeks of exposure, mice were transcardially perfused and their brains collected. Tissues were sliced and then stained for markers of microgliosis, astrogliosis and myelin integrity.

Preliminary data suggests that there is an increase in microglial reactivity across the brain in mice exposed to 3 ug/mL DCZ-infused water compared to those exposed to 0.3ug/mL or vehicle. Ongoing studies are being carried out to evaluate the effects of chronic hypoperfusion on mouse behavior. Utilizing Digital Ventilated Cages, numerous behaviors ranging from locomotor to sleep/wake cycles were recorded. Data from these studies will be used to understand the immediate and long-term behavioral changes that may arise from chronic hypoperfusion that are generally missed in standard housing set ups.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.11/AA6

Topic: D.02. Ischemia

Support: NIH R61NS137365-01

Title: Pericyte-induced capillary perfusion deficits reveal regional vulnerability to hypoxia

Authors: *D. T. CHEN^{1,2}, L. T. SULLIVAN¹, C. FOSTER¹, K. ELK³, B. ZIMMERMAN³, B. MARXMILLER³, T. MCGILLIS¹, A. MISHRA³, Z. ZHAO^{4,5}, A. Y. SHIH¹;

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Abstract: Cerebral hypoperfusion, defined as reduced blood flow to the brain, is a hallmark of aging and dementia that compromises oxygen delivery essential for neuroglial function. The resulting tissue hypoxia reflects downstream metabolic stress and serves as a spatially localized marker of microvascular dysfunction. However, the regional determinants of brain hypoxia and their relationship to vascular architecture remains poorly understood. Capillary pericytes, contractile mural cells embedded in 90% of the vascular endothelium, are well positioned to regulate capillary blood flow, yet their contribution to hypoxia remains unclear. To test whether pericyte activation drives tissue hypoxia, we developed a novel chemogenetic mouse line, *Atp13a5*^{CreERT2}-G_q-DREADD, by breeding *Atp13a5*-2A-CreERT2-IRES-tdTomato knock-in mice with mice expressing a floxed G_q-DREADD sequence. This model leverages *Atp13a5*, a CNS-enriched pericyte marker, to genetically target CNS pericytes. Mice received daily tamoxifen injections (70 mg/kg, i.p.) for one week, followed by a two-week recombination period. G_q-DREADD expression was observed in a sparse population of α-SMA-negative capillary pericytes. Adult mice were then injected with pimonidazole hydrochloride (Hypoxyprobe, 70 mg/kg, r.o.), an exogenous hypoxia marker that binds tissue at pO₂ < 10 mmHg. Mice then received deschloroclozapine (DCZ, 10 μg/kg, i.p.) to induce pericyte contraction. Ninety minutes later, brains were collected, sectioned, and immunostained with PAb2627 to detect hypoxia. Z-score normalized pixel intensities were used to map diffuse ($z > 1\sigma$) and focal ($z > 2\sigma$) hypoxia across anatomically defined brain regions. Compared to controls, DCZ-treated mice exhibited increased hypoxic burden in the lateral cortex, ventrolateral brain regions (e.g., piriform cortex, amygdala) and hippocampus. These regions showed a higher density of discrete hypoxic micropockets >1,000 μm². Quantitative analysis confirmed significant regional differences in hypoxic area and micropocket density. Capillary length density, derived from vascular gel-based labeling and serial two-photon microscopy, was inversely correlated with both hypoxia measures, implicating regional capillary topology as a key determinant of susceptibility. These findings demonstrate that acute pericyte activation is sufficient to induce localized hypoxia and suggest that capillary architecture underlies regional vulnerability to perfusion deficits. This work provides a mechanistic framework linking pericyte function, capillary structure, and brain oxygenation.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.12/AA7

Topic: D.02. Ischemia

Support: NIH R01 AG062738

Title: The role of platelet-derived growth factor beta in the structural remodeling of brain pericytes

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Abstract: Pericytes are integral to the maintenance of brain capillary structure, blood-brain barrier integrity, and capillary blood flow. Pericyte loss occurs in aging and Alzheimer's Disease (AD), and experimental approaches to ablate pericytes in mice result in abnormalities in capillary structure and perfusion. Pericytes have demonstrated remodeling capabilities such that after selective pericyte ablation, surrounding pericytes extend their processes to restore endothelial coverage and capillary diameter. The rate of pericyte remodeling is decreased in aging and mouse models of amyloid pathology. However, the mechanisms responsible for pericyte remodeling are still unknown. A target for understanding structural remodeling is the platelet-derived growth factor (PDGFB/PDGFR beta) signaling pathway that is necessary for the recruitment of pericytes to endothelial cells during vascular development and has been shown to be dysregulated in human AD tissue. To understand the role of PDGFR beta in pericyte remodeling, we implanted chronic cranial windows in 3-9 month old PDGFR beta-tdTomato mice that express tdTomato in all mural cells. We administered a PDGFR beta selective inhibitor SU16f at 10 mg/kg per day via oral gavage that was blood-brain barrier permeable and present for >12 hours after administration based on HPLC mass spectrometry. In vivo two-photon microscopy was used to ablate capillary pericytes and measure the average growth rate of pericyte processes following pericyte ablation over 7 days. SU16f did not decrease the rate of pericyte remodeling. However, the average growth rate correlated to baseline length such that pericyte processes less than 25 μ m had an increased growth rate compared to processes exceeding 25 μ m. These unexpected results could be due to compensatory effects of drug administration, but suggest that PDGFR beta is involved in pericyte remodeling. As such, we are currently examining the rate of remodeling in a CNS pericyte specific PDGFR beta conditional knockout mouse model. Understanding the mechanisms of pericyte remodeling is important for developing therapeutic treatments to promote pericyte coverage in aging and disease.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

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Program #/Poster #: PSTR289.13/AA8

Topic: D.02. Ischemia

Support: 5K08NS118138
5R37CA275954
R01AG062738

Title: Heterogenous endothelial activation and capillary architecture shapes specialized inflammatory niches in the brain microvasculature in vivo

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Abstract: Endothelial activation is a hallmark of neuroinflammation in brain diseases. Increased leukocyte adhesion to brain vessels can lead to vascular dysfunction, including disrupted barrier function, dysregulated cerebral blood flow, and capillary flow disturbances. Single-cell transcriptomic studies have revealed endothelial heterogeneity in immune-regulatory gene expression both across and within vascular zones. However, the spatial organization of this heterogeneity and its influence on endothelial-leukocyte interactions within the live 3-D brain vasculature remains largely unknown. To address this, we performed in vivo spatial mapping of inflammatory-responsive endothelial niches in the mouse brain microvasculature utilizing two-photon imaging. Mice with chronic cranial windows received either lipopolysaccharide (LPS) (3 mg/kg, i.p.) or saline two days prior to imaging to induce neuroinflammation or as sham control. Thirty minutes prior to imaging, anti-ICAM-1 or -VCAM-1 antibodies, anti-CD45.2 antibodies and fluorescently conjugated dextran were administered intravenously to label activated endothelium, circulating leukocytes and brain vasculature, respectively. We observed that ICAM-1 and VCAM-1 expression was highly heterogeneous, displaying patchy patterns across and within arteriolar, capillary and venular zones. Both were predominantly expressed in venular and peri-venous capillary zones, and ICAM-1 expression was more often distributed over other capillary zones compared to VCAM-1. The intravenous labeling of ICAM-1 and VCAM-1 in Claudin5-eGFP mice revealed that the patchy patterns resulted from differential upregulation of ICAM-1 and/or VCAM-1 among neighboring individual endothelial cells. Circulating leukocytes appeared to be trapped on a subset of the ICAM-1 or VCAM-1-enriched endothelium, obstructing capillary blood flow (122.25 ± 13.81 capillary stalls/mm³ brain, 8 mice). In addition, the emergence of leukocyte plugging was higher at vascular branching points or within capillaries of a greater tortuosity and longer length, implying that the leukocyte adhesion dynamics is also influenced by local capillary geometry. In sum, our results show that single-cell-level variability in endothelial inflammatory responses shapes spatial heterogeneity of the likelihood of endothelial-leukocyte activity within capillaries, in conjunction with local vascular geometry. We suggest that specialized vascular niches can play differential immunomodulatory roles in impacting brain pathologies.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

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Program #/Poster #: PSTR289.14/AA9

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: R01NS078168
U19NS128613

Title: Visualizing cerebrospinal fluid displacement driven by penetrating arterial dilation

Authors: *H. LEE¹, H. HIRASE², P. J. DREW¹;

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Abstract: Arterial pulsation-driven cerebrospinal fluid (CSF) circulation is thought to be important for preventing the accumulation of metabolic waste in brain. CSF is in contact with brain tissue in the perivascular space (PVS) surrounding penetrating vessels. Prior studies have shown that the PVS can be compressed by arterial dilations, implying that the dilation of arteries can drive CSF displacement; however, whether solutes are displaced with CSF during dilation of penetrating arteriole remains unclear. Technical challenges of introducing extrinsic CSF tracer in PVS of penetrating arterioles have hindered the direct access to solute movement in PVS of penetrating arterioles. Here, we labeled CSF surrounding penetrating vessels using an intrinsically generated fluorescently tagged extracellular protein near the penetrating artery. We transduced cortical cells with AAV9 encoding the immunoglobulin kappa light chain (IgKL) conjugated with the green fluorophore mNeonGreen (mNG). With this method, the PVS around penetrating arterioles were visible. Using two-photon microscopy in unanesthetized, head-fixed mice, we found that CSF surrounding penetrating vessels was displaced during whisker stimulation-evoked arterial dilations. Our finding provides a direct observation of arterial pulsation promoting solute turnover in CSF by displacing it from the perivascular space under physiological conditions.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

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Program #/Poster #: PSTR289.15/AA10

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH R01-AT011429
NIH U19-NS128263

Title: Volitional Respiration Modulates CSF Flow and Autonomic Physiology in the Awake Human Brain

Authors: *M. BANKS^{1,3,4}, H. FISHER^{2,3,4}, D. E. GOMEZ⁵, V. NAPADOW^{6,8,3}, J. R. POLIMENI⁹, L. D. LEWIS^{4,3,7};

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Abstract: Respiration is a known modulator of cerebrospinal fluid (CSF), the critical fluid involved in brain waste clearance. However, prior studies have not systematically examined how different respiratory frequencies—spanning a range of arousal states—influence the coordinated dynamics of CSF flow, cerebral hemodynamics, and autonomic physiology¹⁻⁴. The speed of breathing varies substantially across arousal states and could potentially modulate the consequences for CSF flow dynamics. The mechanisms linking volitional respiration to CSF flow and brain state remain incompletely understood yet are essential to leveraging the breath as a tool for improving brain health. To investigate how breath timing modulates CSF flow, we employed ultra-high field 7 Tesla phase contrast MRI (temporal resolution=903ms) and fluid-sensitive fast fMRI (TR=499ms) to measure CSF flow in the fourth ventricle during a visually guided breathing task in 23 healthy adults. Each trial consisted of 1 minute of free breathing before and after (pre- and post-free) 3 minutes of paced breathing at 0.1, 0.125, 0.17, or 0.25 Hz (corresponding to 10s, 8s, 6s, and 4s breath cycles). These paces span the spectrum of rhythms observed across natural arousal states^{5,6}. During each run, we simultaneously recorded CSF inflow, BOLD signal, heart rate, expired CO₂, and respiratory waveforms. We found that slower paces of breathing—particularly at 0.1 Hz—increased CSF inflow and velocity compared to faster paces. These effects persisted even after controlling for respiratory depth and significant heart rate changes across conditions, suggesting a unique contribution of respiratory timing and autonomic changes. To evaluate the hemodynamic response, we used FSL FEAT to model BOLD changes during paced breathing and contrasts with the flanking free-breathing periods. We observed a widespread decrease in BOLD signal during the paced breathing task, indicating a global modulation of hemodynamics. These results enhance our mechanistic understanding of respiration's role in CSF dynamics and brain physiology, and suggest that volitional breathing may serve as a noninvasive tool to modulate brain state for current (e.g. yoga, meditation) and future interventions targeting cognitive or psychiatric outcomes.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

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Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: JSPS KAKENHI (24K21952, 24K02217 to M.N., 25K10630 to T.S.)
JST CREST (JPMJCR2331 to Y.O.)

Title: Stimulated Raman scattering imaging visualizes the water dynamics in the mouse brain

Authors: *T. SHINOTSUKA¹, Y. OZEKI², M. YASUI¹, M. NURIYA¹;

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Abstract: The flow of fluid is considered to be a part of the clearance system of metabolic wastes in the brain. The cerebrospinal fluid and the interstitial fluid flow in the brain parenchyma, washing the wastes out of the brain. Impaired flow of fluids may lead to abnormal accumulation of wastes such as amyloid β protein and cognitive dysfunctions. Thus, understanding the dynamics of fluids in the brain is expected to shed new light on the mechanisms underlying neurodegenerative disorders. However, in sharp contrast to solutes, the dynamics of water in the brain remain poorly characterized. Here, we apply stimulated Raman scattering (SRS) microscopy to both *ex vivo* and *in vivo* mouse brain to directly assess the dynamics of water in living brain tissues. To investigate dynamics of water, we utilized deuterated water (D_2O) that is biochemically similar to normal water (H_2O) but distinctly detectable by vibration-sensitive Raman scattering as a probe. By continuously observing D_2O and fluorescent dyes perfused into *ex vivo* brain tissues, we successfully visualized the dynamics of D_2O and fluorescent dyes simultaneously, together with structures of specific cell types, at high spatiotemporal resolution. Using this system, we found that D_2O diffuses more rapidly than fluorescent dyes in *ex vivo* mouse brain tissues. Detailed analysis revealed that D_2O diffuses through brain tissues both inside and outside cells, whereas fluorescent dyes diffuse only through extracellular spaces. Furthermore, by characterizing water dynamics under a pathological condition, we found a uniquely robust nature of water inside brain tissues. Recently, we have upgraded the system to visualize the dynamics of water in the *in vivo* mouse brain. The *in vivo* SRS imaging system enables us to visualize the dynamics of D_2O in mouse brain following intravenous administration of the D_2O -based saline solution through the femoral vein. Notably, the dynamics of D_2O differ between arteries, veins and brain parenchyma. Thus, our approach provides new insights into the fundamental properties of water dynamics in the living brain and demonstrates the potential of SRS multimodal microscopy as a powerful tool for studying brain fluid transport under physiological and pathological conditions.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR289.17/AA12

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH 1R01HL162575

Title: Divergent Hypoxia- and Acidosis-Induced Responses in Parenchymal Arterioles of the Supraoptic Nucleus: An Activity-Dependent Neurovascular Mechanism

Authors: *S. AGUS¹, L. SHOOK^{2,3}, J. E. STERN^{2,3}, J. A. FILOSA¹;

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Abstract: Neurovascular coupling (NVC) regulates local blood flow to meet neural metabolic demands. While traditional NVC involves activity-driven vasodilation, recent evidence suggests an inverse NVC—neuronal activity leading to vasoconstriction—in select brain regions, though its functional significance remains unclear. The hypothalamic supraoptic nucleus (SON) regulates water balance through vasopressin (VP) release into the circulation. Our previous work demonstrated that SON arterioles constrict under hyperosmotic stress, inducing local hypoxia. We hypothesize that activity-induced hypoxia enhances VP neuronal activation, while subsequent acidification mediates vasodilation, suppressing neuronal firing and protecting against ischemic injury. Using *ex vivo* brain slices from 7-10-week-old male Wistar rats, we combined video microscopy of SON arterioles and whole-cell patch-clamp recordings of VP neurons under hyperosmotic, hypoxic, or acidotic conditions. Arterioles were preconditioned with U46619 (200 nM) to induce steady-state vascular tone. Hypoxia was evoked by switching the artificial cerebral spinal fluid (aCSF) from 95% to 20% O₂ (5% CO₂, balance N₂), while acidification was induced via hypercapnic acidosis (HA) (15% CO₂). Hypoxia significantly increased vasoconstriction ($p=0.01$, $n=11$) and increased VP neuronal firing ($p=0.0004$, $n=12$), whereas HA induced vasodilation ($p=0.0046$, $n=10$) and suppressed VP activity ($p=0.0222$, $n=16$). To test our working hypothesis, we measured the response of SON arterioles to HA in the presence of 1 μ M VP. VP-induced vasoconstriction ($p=0.0064$, $n=9$) was reversed by HA-mediated vasodilation ($p=0.0006$, $n=9$). These findings suggest that *inverse* NVC within the SON facilitates VP neuronal activation while reinforcing a hypoxic microenvironment. However, the resultant acidification counteracts hypoxia by promoting vasodilation, restoring perfusion, and suppressing neuronal activity. This biphasic vascular response to chemosensitive signals may serve a neuroprotective role, mitigating hypoxia-induced damage or modulating VP responses to hyperosmotic stimuli.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

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Program #/Poster #: PSTR289.18/AA13

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: Turkish Academy of Sciences (GEBIP Program) (Evren Erdener)
Turkish Academy of Sciences (Turgay Dalkara)

Title: Downstream capillary pericytes regulate flow via torsional contraction

Authors: *E. ERDENER¹, G. KÜRELI¹, M. BASARAN¹, M. HADDARA², L. ALARCON-MARTINEZ², T. DALKARA³;

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Abstract: Background: The debate over the contractility and active blood flow regulation by downstream capillary pericytes persists, as their slender, strand-like processes are believed to lack the proper form for effective contraction.

Aim: This study investigates the contractility of downstream pericytes in retinal microcirculation. We compare organization of contractile fibers to discern different contraction phenotypes and assess their potential impact on blood flow regulation.

Method: Whole-mount retinal preparations from Swiss-albino mice were fixed with methanol and were stained with fluorescent lectin and immunolabeled for α -SMA. Intravitreal noradrenaline (NA) and/or blebbistatin was administered before euthanasia, to modulate actomyosin contraction in pericytes. 3D capillary models were generated via segmentation of confocal Z-stack images for subsequent geometric analyses. Another set of mice were injected with FITC-dextran and retinal capillaries were imaged in vivo as line scans interlaced with repetitive Z-stacks with two-photon microscopy (TPM) over 10 min.

Results: While circular pericyte processes resulted in nodal constrictions in NA-treated upstream capillaries, axial and oblique α -SMA fiber bundle orientations in downstream midcapillary pericytes resulted in twisting contractions of the vessel wall. A notable increase in centerline torsion was observed in segments covered by midcapillary pericytes on the 5th and 6th order capillaries from the NA-treated retina compared to the naive group. This increase was prevented by blebbistatin, suggesting that the torsional contraction was mediated by actomyosin coupling. Torsional bends were associated with asymmetric arrangement of pericyte processes surrounding these particular segments. These ex vivo observations were corroborated by retinal imaging in living mice. We detected spontaneous and transient torsional changes in capillary geometries over minutes and we found a significant negative correlation between torsion and red blood cell velocities. Conclusions: These findings provide direct evidence of the functional role of downstream pericytes in blood flow regulation, highlighting their distinctive torsional forces aligned with their helical structure.

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Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

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Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: China Brain Initiative Grants (STI2030-Major Projects 2021ZD0202803)
New Cornerstone Investigator Program
Research Unit of Medical Neurobiology at Chinese Academy of Medical Sciences (2019RU003)
Beijing Municipal Government

Title: G protein-coupled receptor 30 mediates neurovascular coupling

Authors: *H. LI;

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Abstract: The brain's immense computational capacity is energetically supported by a precisely regulated blood supply. This fundamental physiological process, termed neurovascular coupling (NVC), manifests as rapid (within seconds) regional cerebral blood flow (CBF) adjustments that dynamically match neuronal activity fluctuations. While NVC dysfunction has been implicated in various neurological disorders, the mechanistic underpinnings of this critical phenomenon remain incompletely understood. We identified G protein-coupled receptor 30 (GPR30), which had been long misclassified as a G protein-coupled estrogen receptor, as a novel mediator of neurovascular coupling. Through comprehensive approaches, including RNA in situ hybridization and transgenic mouse models, we demonstrated exclusive GPR30 expression in mural cells within the central nervous system. Pharmacological activation of GPR30 induced robust cerebrovascular dilation. Using in vivo two-photon imaging and functional ultrasound imaging, we established that GPR30 knockout mice exhibited significant impairment in neurovascular coupling responses in both sexes. Specifically, GPR30 deficiency attenuated whisker stimulation-evoked local cerebral blood flow (CBF) increases in the barrel cortex, visual stimulation-induced hemodynamic responses in the primary visual cortex, and direct electrical stimulation-triggered CBF augmentation in neural circuits. Our in vitro studies reveal that GPR30 is uniquely activated by bicarbonate (HCO_3^-), a key metabolic byproduct of neuronal activity, rather than by conventional vasodilators such as PGE_2 , ATP, adenosine, or glutamate. In acute brain slice preparations, HCO_3^- similarly evoked mural cell calcium transients in a GPR30-dependent manner. Knockout of GPR30 in mice significantly attenuated brief CO_2 stimulation-induced vasodilation. Furthermore, mutant mice expressing a bicarbonate-insensitive GPR30 variant exhibited impaired neurovascular coupling response. These findings support a novel mechanistic paradigm: mural cell-expressed GPR30 functions as a metabolic sensor that detects activity-dependent local increases in bicarbonate concentration, thereby mediating neuronal activity-evoked vasodilation and subsequent cerebral blood flow augmentation.

Disclosures: H. Li: None.

Poster

PSTR289: Cerebral Blood Flow and Microvasculature

Location: SDCC Halls B-H

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Program #/Poster #: PSTR289.20/AA15

Topic: G.05. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH U19 NS123717

Title: Optogenetically-induced neuronal potassium efflux increases local cortical blood flow

Authors: ***B. HOLLOWAY**¹, A. DESOUZA², B. L. BLOODGOOD⁴, D. KLEINFELD³;

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Abstract: Cerebral blood flow (CBF) is tightly coupled to neuronal activity. Yet the hyperemic response often precedes detectable tissue hypoxia or glucose depletion. This suggests upstream signaling mechanisms independent of metabolic demand. Interstitial potassium (K⁺) has been proposed as a key neurovascular mediator, but neuronal activation releases many vasoactive signals, which complicates efforts to isolate K⁺'s specific role. Here, we developed an optogenetic approach to selectively increase neuronal K⁺ conductance and tested whether this is sufficient to modulate local blood flow. C57BL/6 mice (8-12 weeks) received intracortical injections of an AAV vector (4.6 nL; 250 μm depth, somatosensory cortex) expressing a CaMKIIα-driven photoactivatable adenylyl cyclase (PAC) and cAMP-gated K⁺ channel (SthK), tagged with mCherry. In ex vivo slices, we confirmed PAC-K functionality using whole-cell patch clamp. Photostimulation (445 nm) evoked an outward current with a reversal potential near *E_K* (-93 mV) in voltage clamp, and robust hyperpolarization and spike silencing during current clamp, despite induced 2-10 Hz spiking. For in vivo studies, mice were implanted with cranial windows. Optogenetic stimulation increased capillary red blood cell speed and dilated pial vessels near the injection site, as measured via two-photon linescan microscopy and widefield imaging respectively. In anesthetized mice, we measured interstitial K⁺ concentration using K⁺-selective microelectrodes at multiple distances from the injection site. Light pulses (0.01-1 s) triggered spatially graded increases in extracellular K⁺, which scaled with pulse duration. Light pulses also correlated with vascular responses when recorded simultaneously. These findings demonstrate that selective neuronal K⁺ efflux is sufficient to rapidly increase local CBF in vivo. This supports interstitial K⁺ as a critical, spatially informative signal in neurovascular coupling and validates a novel optogenetic tool for its mechanistic dissection.

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Poster

PSTR290: Physiology and Models Advancing New Theories of Sleep

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR290.01/AA16

Topic: G.07. Biological Rhythms and Sleep

Support: National Natural Science Foundation of China 32325021
National Natural Science Foundation of China 32221003

Title: A time window for memory consolidation during NREM sleep revealed by cAMP oscillation

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Abstract: Memory formation requires specific neural activity in coordination with intracellular signaling mediated by second messengers such as cyclic adenosine monophosphate (cAMP). However, the real-time dynamics of cAMP remain largely unknown. Here, using a genetically encoded cAMP sensor with high temporal resolution, we found neural-activity-dependent rapid cAMP elevation during learning. Interestingly, in slow-wave sleep, during which memory consolidation occurs, the cAMP level in mice was anti-correlated with neural activity and exhibited norepinephrine β 1 receptor-dependent infra-slow oscillations that were synchronized across the hippocampus and cortex. Furthermore, the hippocampal-cortical interactions increased during the narrow time-window of the peak cAMP level; suppressing hippocampal activity specifically during this window impaired spatial memory consolidation. Thus, hippocampal-dependent memory consolidation occurs within a specific time window of high cAMP activity during slow-wave sleep.

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Poster

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Program #/Poster #: PSTR290.02/BB1

Topic: G.07. Biological Rhythms and Sleep

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Title: Biophysical and molecular properties of glutamatergic neurons in the ventral prefrontal cortex that promote recovery sleep-like in mice

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Abstract: With extended periods of wakefulness, rising sleep pressure is reflected by prominent δ oscillations (0.5 - 4Hz) in the electroencephalogram (EEG) originating from the prefrontal cortex (PFC). The neural circuits in PFC that mediate this process remain poorly understood. To gain access to the neuronal populations involved in δ oscillations, we activity-tagged neurons under light anesthetic conditions using TRAP-cre mice. We identified a population of layer 5 (L5) pyramidal neurons enriched in the ventral PFC. Chemogenetic activation of these activity-tagged neurons in the PFC of freely moving mice promoted non-rapid eye movement (NREM) in freely moving mice that was reminiscent of recovery sleep. Increased δ oscillations, NREM bout durations, punctuated by occasional REM episodes and microarousals were followed by consolidated periods of wakefulness in HM3Dq transfected animals (n=15) but not mCherry controls (n=15). Chemogenetic activation of PFC neurons was also associated with $\sim 1-2^{\circ}\text{C}$ drop in body temperature (n=7). To elucidate the molecular and biophysical cell-type identity of these NREM-promoting neurons (NpN), we performed Patch-seq recordings in ex vivo slices. The majority of activity-tagged NpNs had biophysical properties were consistent with L5 extratelencephalic neurons (L5 ET). These NpNs were enriched with δ -frequency resonance subthreshold to action potentials and low input resistance, both features consistent with the presence of h-currents. The proportion of NpN's with L5 ET biophysical properties (23/37) was greatly enriched compared to neighboring L5 pyramidal neurons (3/26). A subset of NpN (19) were sequenced and mapped to molecular neuronal subtypes using correlation-based clustering. The majority of NpNs were molecular defined as L5 ET (13/19). Based on these data, we tested whether we could recapitulate promoting NREM sleep with a transgenic mouse line designed to label L5 ET neurons (Glt25d2-cre_NF107 MMRC: 036504; Colgalt2-cre). Chemogenetic activation of Colgalt2-cre+ neurons in the infralimbic area of PFC (ILA) promotes NREM sleep (n=5). Mapping the long-range projections of Colgalt2-cre+ neurons revealed that they have minimal projections to the hypothalamus, but project strongly to the anterior medial thalamus and claustrum. Combined, this work suggests that NpNs may promote NREM sleep via a novel circuit.

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Poster

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Title: Neocortical long-range inhibition promotes cortical synchrony and sleep

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Abstract: Behavioral states such as sleep and wake are highly correlated with specific patterns of rhythmic activity in the cortex. During low arousal states such as slow wave sleep, the cortex is synchronized and dominated by low frequency rhythms coordinated across multiple regions. Although recent evidence suggests that GABAergic inhibitory neurons are key players in cortical state modulation, the in vivo circuit mechanisms coordinating synchronized activity among local and distant neocortical networks are not well understood. Here, we show that somatostatin and chondrolectin co-expressing cells (Sst-Chodl cells), a sparse and unique class of neocortical inhibitory neurons, are selectively active during low arousal states and are largely silent during periods of high arousal. In contrast to other neocortical inhibitory neurons, we show these neurons have long-range axons that project across neocortical areas. Activation of Sst-Chodl cells is sufficient to promote synchronized cortical states characteristic of low arousal, with increased spike co-firing and low frequency brain rhythms, and to alter behavioral states by promoting sleep. Contrary to the prevailing belief that sleep is exclusively driven by subcortical mechanisms, our findings reveal that these long-range inhibitory neurons not only track changes in behavioral state but are sufficient to induce both sleep-like cortical states and sleep behavior, establishing a crucial circuit component in regulating behavioral states.

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Poster

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Topic: G.07. Biological Rhythms and Sleep

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Title: Prefrontal Cortex Regulation of Pontine Waves

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Abstract: Rapid eye movement (REM) sleep is a heterogeneous state comprising both tonic and phasic substages. Classic hallmarks of phasic REM sleep include rapid eye movements and pontine waves (p-waves). P-waves during REM sleep coincide with a synchronous activation of cortical and subcortical brain areas, potentially providing windows for synaptic plasticity and memory processing. However, the neural mechanisms regulating the induction of p-waves, and their frequency remain largely unknown. In this study, we investigated the role of the medial prefrontal cortex (mPFC) in the regulation of p-waves. Using calcium imaging, we found that neurons in the mPFC exhibit heightened activity during spontaneously occurring p-waves. To further explore the relationship between the mPFC and p-waves, we conducted optogenetic stimulation experiments targeting pyramidal neurons within the infralimbic (IL) region of the mPFC. Single optogenetic pulses in the mPFC reliably triggered p-waves specifically during REM sleep. We confirmed that the optogenetically induced p-waves closely resembled spontaneous p-waves in their waveform and their effect on EEG. Our results reveal a novel cortical top-down mechanism for the regulation of key features of phasic REM sleep, setting the stage for further functional investigation of this substage of REM sleep.

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Poster

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Topic: G.07. Biological Rhythms and Sleep

Support: NIH Grant R01AG077570

Title: Region-specific pericyte Ca²⁺ transient dynamics and capillary hemodynamics during sleep-wake cycle

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Abstract: Brain capillary pericytes are crucial in neurovascular coupling (NVC) as they regulate cerebral blood flow (CBF) in response to changes in local neuronal activity. While patterns of neuronal activity during sleep and waking have been extensively studied, pericyte activity remains less understood. Using an *in-vivo* calcium (Ca^{2+}) imaging tool, we recorded cytosolic Ca^{2+} transient dynamics from brain pericytes across natural sleep-wake cycles in the prefrontal cortex (PFC) and lateral hypothalamus (LH) of mice expressing the Ca^{2+} sensor GCaMP6f specifically in pericytes. Our results show that PFC pericytes exhibit low basal Ca^{2+} transients during non-rapid eye movement sleep (NREMS) and rapid eye movement sleep (REMS), while showing synchronous Ca^{2+} peaks following transitions from sleep to waking. In contrast, LH pericytes display diverse Ca^{2+} transient patterns across the sleep-wake cycle, with 81.3%, 15.6%, and 56.3% presenting Ca^{2+} peaks during NREMS, REMS, and waking, respectively. Hemodynamic recordings in PFC demonstrated more vigorous and dynamic fluctuations in red blood cell (RBC) velocity during waking compared to NREMS, likely driven by periodic pericyte Ca^{2+} peaks during waking. In LH, average capillary diameter and RBC velocity significantly increased during waking compared to NREMS, aligning with the predominantly NREMS-ON pattern of pericyte Ca^{2+} transients and frequent capillary stalling observed during NREMS. These findings underscore the essential role of region-specific pericyte activity patterns in regulating local CBF during natural sleep-wake cycles, emphasizing their importance in maintaining neurovascular function.

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Poster

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Program #/Poster #: PSTR290.06/BB5

Topic: G.07. Biological Rhythms and Sleep

Title: State-dependent coupling to respiration in substantia nigra and motor cortex

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Abstract: Neural oscillations are increasingly recognized to be coupled with respiration rhythms across widespread brain regions. Emerging evidence suggests that such coupling coordinates sleep-related oscillations and supports memory consolidation. While the basal ganglia is increasingly implicated in sleep regulation, respiration-coupled neural dynamics in this region have not been previously reported. Here, we examined state-dependent respiration-neural couplings in the substantia nigra pars reticulata (SNr), a major output nucleus of the basal ganglia, and the primary motor cortex (M1) in mice. Simultaneous recordings of local field

potentials (LFPs) from M1 and SNr, along with diaphragm EMG and ECG, revealed state-dependent, region specific patterns of respiration coupling. Coupling strength in both SNr and M1 was attenuated during non-rapid eye movement (NREM) sleep compared to rapid eye movement (REM) sleep and quiet wake. However, under ketamine/xylazine anesthesia, coupling was markedly enhanced in SNr, but not in M1, indicating region-specific sensitivity to arousal and anesthesia state. Notably, coupling strength was systematically related to delta-band power; stronger respiration coupling was associated with decreased slow delta (0.5-2 Hz) power and increased fast delta (2.5-4 Hz) power. These coupling dynamics were embedded within infraslow fluctuations that also structured other neural and physiological patterns. Together, our results highlight dynamic, state-dependent modulation of respiration-neural couplings in cortico-basal ganglia circuits, suggesting its significant relationship to body-brain interactions during sleep and anesthesia.

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Poster

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Topic: G.07. Biological Rhythms and Sleep

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Title: Sleep-like Replay Enables Continual Learning with Equilibrium Propagation

Authors: *Y. KUBO, E. DELANOIS, M. V. BAZHENOV;
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Abstract: Recurrent neural networks (RNNs) trained using Equilibrium Propagation (EP)—a biologically plausible learning algorithm—have shown strong performance across tasks such as image classification and reinforcement learning and can even rival backpropagation-based neural networks—widely used models that have powered major advances in areas like computer vision, strategic gameplay, and reinforcement learning. However, EP-based RNNs remain vulnerable to catastrophic forgetting in continual learning settings, where learning new tasks disrupts previously acquired knowledge. This contrasts with the human brain’s ability to retain and integrate both old and new information, partly due to memory consolidation during sleep via the replay of learned experiences. To address this challenge, we introduced a **sleep-like replay consolidation (SRC)** algorithm tailored for EP-trained RNNs. SRC significantly enhanced the network’s resistance to forgetting in class-incremental learning. When applied after each new task training, our multilayer RNN trained with EP (MRNN-EP) outperformed feedforward networks equipped with several well-established regularization techniques. Moreover, MRNN-EP with SRC matched or exceeded the performance of RNNs trained with Backpropagation

Through Time (BPTT) across multiple datasets—including MNIST, Fashion-MNIST, Kuzushiji-MNIST, CIFAR-10, and ImageNet. Combining SRC with rehearsal (awake replay) further improved long-term knowledge retention during continual learning. Our findings demonstrate the effectiveness of biologically inspired sleep-like replay in RNNs and highlight the potential of integrating human-like learning dynamics into artificial neural networks.

Disclosures: **Y. Kubo:** A. Employment/Salary (full or part-time): University of California San Diego. **E. Delanois:** A. Employment/Salary (full or part-time): University of California San Diego. **M.V. Bazhenov:** A. Employment/Salary (full or part-time): University of California San Diego.

Poster

PSTR290: Physiology and Models Advancing New Theories of Sleep

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Topic: G.07. Biological Rhythms and Sleep

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Title: Mechanistic insights into age-related changes in sleep slow oscillations through whole-brain modeling

Authors: G. NAVAS, *M. V. BAZHENOV;
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Abstract: Slow-wave sleep (SWS), a dominant brain state during non-rapid eye movement (NREM) sleep, plays a critical role in memory consolidation. A hallmark of SWS is the slow oscillation (SO, <1 Hz), characterized by alternating active (“up”) and silent (“down”) states in the thalamocortical network. With aging, sleep EEG recordings consistently show altered SO properties, including reduced amplitude and density, as well as decoupling from sleep spindles. However, the mechanisms by which age-related large-scale structural brain changes, such as cortical thinning and white matter degradation, impact these dynamics remain poorly understood. To investigate this, we developed a multi-scale, whole-brain thalamocortical network model of SWS that incorporates realistic human cortical connectivity. The model includes 10,242 cortical columns spanning one hemisphere, each comprising six layers with populations of spiking pyramidal (PY) and inhibitory (IN) neurons. It also features a thalamic module with biophysically grounded thalamocortical and reticular neurons. Long-range cortical connectivity is derived from diffusion MRI tractography data from the Human Connectome Project, with distance-dependent synaptic delays. A myelination-based hierarchical organization of connections determines laminar connectivity. We simulated aging as a progressive weakening or loss of synaptic connections. The model recapitulates empirically observed age-related SO

alterations, including reduced amplitude, density, and slope, alongside increased duration. Importantly, these changes were best reproduced when selectively degrading PY-PY, but not PY-IN, connections - suggesting that a shift in excitation-inhibition balance may underlie the aging brain's altered sleep dynamics. These results provide mechanistic insight into how structural brain aging degrades the neural substrate of sleep and suggest specific circuit-level targets for interventions to preserve sleep quality and cognitive function across the lifespan.

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Poster

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Topic: G.07. Biological Rhythms and Sleep

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Title: Thalamic input modulates hippocampal ripple timing during closed-loop sleep stimulation: insights from a biophysical model

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Abstract: Sleep supports memory consolidation through coordinated interactions among brain rhythms, including slow oscillations (SOs), spindles, and sharp wave-ripples (SWRs) in the cortex, thalamus, and hippocampus. Empirical studies across species have shown that the spectral power, density, and temporal coupling of these rhythms positively correlate with improved memory consolidation. Recent studies have demonstrated that closed-loop auditory stimulation (CLAS), precisely timed to the SO up phase, can increase power in the SO and spindle bands and induce SWRs with minimal latency, thereby improving memory performance through enhanced triple-phase locking of the three rhythms. While CLAS provides a powerful tool to control the timing of SWRs relative to the SO phase, the circuit-level mechanisms mediating these effects remain poorly understood.

To address this, we employed a closed-loop biophysical model of the hippocampal-thalamic-cortical loop, incorporating spiking neuron populations in the cortex, thalamus (including the reticular, medial dorsal, and reuniens nuclei), and hippocampal subfields CA1 and CA3. Neural dynamics are modeled using detailed Hodgkin-Huxley neuron models in the thalamus and cortex, and computationally efficient integrate-and-fire models for the hippocampus. The model can reproduce SO, ripple, and SWR events during slow-wave sleep.

Critically, we introduced a reuniens nucleus-to-CA1 projection - an anatomically supported but underexplored pathway. Simulating CLAS by targeting thalamic relay neurons at specific SO phases reproduced key experimental findings: an immediate increase in ripple power following stimulation, followed by sustained ripple suppression. Removal of the thalamus-to-CA1 connection abolished this pattern, highlighting its necessity in coordinating SO-SWR dynamics. Our results suggest that reuniens-mediated input modulates hippocampal responses to sensory stimulation during sleep. Beyond explaining recent experimental results, this model provides a platform for predicting how changes in connectivity within the hippocampal-thalamic-cortical loop affect sleep dynamics and memory consolidation.

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Topic: G.07. Biological Rhythms and Sleep

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Title: Systems consolidation can promote selective consolidation or forgetting depending on whether the cortex is in a slow or delta oscillation regime

Authors: *R. GOLDEN¹, A. MIZRAHI-KLIGER², S. COURY³, K. GANGULY⁴, M. V. BAZHENOV¹;

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Abstract: The role of sleep in memory consolidation has been an increasingly studied topic in neuropsychology since Jenkins and Dallenbach noticed that sleep helped with retention just over a century ago. Since then, research has put forward hypotheses for the roles of individual sleep stages, as well as processes within these stages, with regards to consolidation. Perhaps the most well studied being the roles of low frequency cortical slow-wave activity (SWA) and high frequency hippocampal sharp-wave ripples (SWRs) during slow-wave sleep (SWS). This framework, known as Systems Consolidation (i.e. Complementary Learning Systems) theory, suggests that SWRs replay recently encoded memories which are slowly assimilated into the cortex during SWS. Traditionally, most studies have not differentiated between slow oscillations (SO; 0.2-1 Hz) and delta (δ ; 1-4 Hz) activity during SWS, focusing instead on how memories are stabilized and strengthened. In this study, we leverage a biophysical thalamocortical model capable of learning neural sequence memories during wakefulness and generating spontaneous memory replay during SWS to investigate the distinct roles of SO and δ activity in memory consolidation. Specifically, we implemented a model of hippocampal indexing that simulates the

routing of SWR activity to the cortex to examine how the assimilation of recent memories into a cortex already containing remote memories depends on whether the cortex is in an SO or δ regime. We found that: (1) without hippocampal indexing, only the remote memory is maintained and further consolidated, while the recent memory is not - regardless of the SWA regime; (2) indexing the recent memory during an SO regime led to robust consolidation of both memories; and (3) indexing the recent memory during a δ regime resulted in consolidation of the recent memory while the remote memory was forgotten. Thus, our model predicts that within the Systems Consolidation framework, selective or joint consolidation of recent and remote memories can occur flexibly, depending on the cortical SWA regime and the presence of hippocampal indexing.

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Topic: G.07. Biological Rhythms and Sleep

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Title: Designing non-invasive thalamic stimulation strategies to enhance spindles: insights from a thalamocortical model

Authors: *G. NAVAS¹, T. RAUFEISEN³, I. VIOLANTE⁵, U. BARTSCH⁴, M. V. BAZHENOV²;

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Abstract: The functional significance of sleep oscillations, including their role in memory consolidation, has spurred growing interest in their targeted modulation. Advances in non-invasive brain stimulation (NIBS) now enable access to deep-brain structures such as the thalamus—a key generator of sleep spindles (~9-16Hz waxing-and-waning oscillations). Specifically, temporal interference stimulation (TIS) is a promising NIBS technique that applies two or more high-frequency currents, individually too fast to elicit spiking, but which interact to produce a low-frequency envelope capable of modulating neural activity. TIS allows for deep brain targeting while minimizing unwanted activation of the overlying cortex and nearby structures. However, the stimulation parameter space, encompassing intensity and temporal patterning, is vast and difficult to explore empirically. To address this, we employed a biophysically detailed thalamocortical network model to systematically evaluate stimulation

patterns that bias spindle activity. The model consists of 1,000 pyramidal (PY), 200 inhibitory (IN), 500 thalamocortical (TC), and 500 reticular (RE) neurons with Hodgkin-Huxley-type dynamics. To simulate TIS, we applied sub-threshold sinusoidal input currents to TC and RE neurons at various target envelope frequencies, systematically varying amplitude and frequency. Our simulations show that subthreshold stimulation can elicit spindle activity in a frequency-dependent manner. Notably, the network exhibits Arnold tongue-like dynamics, with maximal spindle responses at 5-6 Hz envelope frequencies. Preliminary human sleep TIS-EEG data supports this prediction. These results suggest that the intrinsic resonance properties of TC relay neurons may allow frequency-selective, cell-type-specific targeting via TIS. This work yields testable predictions for optimizing TIS protocols to modulate sleep spindles, with potential applications in improving sleep quality and continuity, enhancing memory consolidation, and developing treatments for sleep disorders.

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Poster

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Topic: G.07. Biological Rhythms and Sleep

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Title: Neural networks navigate sleep-wake cycles through quasicritical dynamics

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Abstract: Despite over a century of investigation, an understanding of why all animals sleep remains elusive. Despite this challenge, there is a general consensus that sleep subserves some fundamental aspect of homeostasis, as it is almost universally restorative. It is generally understood that any system that drifts or is actively modified (i.e., plasticity) requires homeostatic control for reliable functioning. Yet mechanistic links between plasticity and sleep, and the precise variables that drive sleep-wake switching, remain controversial. Only recently have quantitative approaches suggested a set-point around which sleep modulates neural activity (Xu et al., 2024). Specifically, emerging data suggest that sleep restores an optimal

computational regime that maximizes information processing in neural circuits. This regime is called criticality. The identification of sleep's set-point may represent a unifying feature of brain activity, as sleep is restorative across all brain regions, species, and the span of life. One of the strictest tests of a candidate set-point is to ask whether it can be used to predict future sleep/wake transitions. Specifically, as an animal moves through stereotyped behavioral cycles, underlying neurobiological networks should follow a consistent pathway through various dynamical states. In this context, the pathways between states and the triggers that govern the transitions are unknown. Here, we introduce quasicriticality, a novel framework that extends criticality theory to a broader dynamical landscape, specifically one that accounts for substantial changes in external drive. We map the hysteresis loop traced by neural networks across sleep/wake cycles within this expanded parameter space, unifying disparate aspects of sleep regulation. This framework accounts for state-dependent alterations in neuronal activity, activity variance, and network stability. We validate our framework using chronic, continuous single unit recordings from mouse and rat isocortex, demonstrating that functional connectivity, average activity, and activity variance track the predicted hysteresis trajectories across sleep-wake cycles. Furthermore, we show that these network metrics can serve as predictors of upcoming state transitions.

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Poster

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Title: A unifying theory of sleep: restoration of an optimal computational regime in diverse networks

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Abstract: Sleep is poised to reveal a unifying principle of neurobiological computation, as it is necessary across all animals, brain regions, and phases of life. This universality, and the universal cost of degraded sleep, implies that sleep actively maintains a feature that accounts for

the brain's remarkable computational power and adaptability.

We reasoned that sleep's endpoint must directly explain computational capacity. A plausible solution emerges from statistical mechanics: when neuronal populations operate near criticality—a boundary state with scale-invariant, marginally stable dynamics—they maximize dynamic range, information transmission, learning flexibility, and behavioral adaptability.

Unlike other homeostatic set-points, criticality has direct explanatory power for the flexible computation that characterizes the vast majority of what brains do.

Using chronic electrophysiology in freely-behaving mice, we tested this hypothesis empirically.

We demonstrate that proximity to criticality follows a robust sleep-wake pattern: wake progressively corrupts computational depth, while sleep restores it. Crucially, distance from criticality—unlike any other neural measure—predicts future sleep-wake behavior, revealing bidirectional coupling between sleep regulation and computational optimization.

Building on this empirical foundation, we conducted a comprehensive evaluation of criticality as a unifying principle. Our meta-analysis of 320 studies spanning two decades reveals universal signatures of criticality across species, measurement scales, and analytical approaches. We reconcile apparent contradictions in the literature through methodological insights, showing that temporal coarse-graining explains seeming inconsistencies between early negative and recent positive reports.

The criticality framework generates testable predictions across neuroscience domains. In tauopathy models, criticality deteriorates progressively even when traditional homeostatic markers remain robust. Human neuroimaging confirms that proximity to criticality predicts cognitive performance. Pathological states reveal characteristic deviations: epilepsy exhibits supercriticality, while schizophrenia, autism, and neurodegeneration show subcritical dynamics. This work reveals how an end-point of sleep points toward criticality as evolution's solution to flexible computation, unifying diverse neurobiological phenomena under a single mathematical principle with direct therapeutic implications.

Disclosures: K.B. Hengen: None. W.L. Shew: None.

Poster

PSTR290: Physiology and Models Advancing New Theories of Sleep

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR290.14/BB13

Topic: G.07. Biological Rhythms and Sleep

Support: ERC Grant 101088598
PID2022-139822NB-I00

Title: Decoding Internal Representations During Sleep Via Neural Population Dynamics Across Cortical Regions

Authors: *Y. BOLLMANN¹, A. ESTOP², P. MATEOS-APARICIO¹, J. FERNÁNDEZ DE CASTRO¹, R. F. CORREIA¹, Y. FUENTEALBA¹, A. RANSON¹;

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Abstract: Sleep plays a critical role in cognitive and physiological processes including memory consolidation, immune regulation, and emotional stability. Beyond these foundational functions, recent evidence suggests that sleep—particularly rapid eye movement (REM) sleep—may support generative cognitive phenomena such as planning, problem-solving, and creativity. These may be enabled by internally simulated experiences akin to dreaming, wherein distributed brain regions generate novel representations or replay prior experiences. To investigate how internal representations are structured during sleep, we combined mesoscale two-photon calcium imaging with pupillometry and electrophysiology in mice. We focused on the posterior parietal cortex (PPC) and primary visual cortex (V1), regions implicated in sensory processing, integration and decision-making. We show the similarities and differences between within area correlational structure, and inter-areal communication in different states of arousal by comparing private and shared low-dimensional embeddings. By analysing activity of axons providing input to V1 from retrosplenial cortex, we additionally propose a potential top-down route through which internally generated activity in V1 is produced/modulated during sleep. Using pupil diameter and movement as markers of sleep state, we found that pupil dynamics reliably predict transitions between wakefulness, non-REM, and REM states. Notably, REM-associated eye movements were aligned to neural activity in many V1 neurons, raising the possibility that these movements reflect active exploration of internally generated visual scenes. Building on these insights, we implemented deep learning-based encoding models to decode internal visual representations from large populations of V1 neurons. These models integrate both sensory and non-sensory variables (e.g., eye position, locomotion speed) to predict neuronal responses, and here we apply them to reconstruct visual representations during sleep. We also evaluate how stimulus design and model generalization across animals affect decoding fidelity. Together, our findings suggest that eye movements during REM may represent active sampling of internal experiences during “dream-like” states. By combining neural recordings with computational modeling, this work opens the door to empirically accessing, modifying and reconstructing the content of internally generated experiences during sleep.

Disclosures: Y. Bollmann: None. A. Estop: None. P. Mateos-Aparicio: None. J. Fernández de Castro: None. R.F. Correia: None. Y. Fuentealba: None. A. Ranson: None.

Poster

PSTR290: Physiology and Models Advancing New Theories of Sleep

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR290.15/BB14

Topic: G.07. Biological Rhythms and Sleep

Support: ASAP Grant 025185

Title: Dopaminergic signaling and sleep homeostasis

Authors: *X. L. HUANG, X. TU, Y. DAN;
Univ. of California Berkeley, Berkeley, CA

Abstract: Dopamine is widely regarded as a wake-promoting neurotransmitter. Intuitively, this suggests that dopamine loss would lead to reduced wakefulness and increased sleep. However, sleep loss is frequently observed in dopamine-deficient conditions such as Parkinson's disease, depression, autism spectrum disorder, and attention-deficit/hyperactivity disorder. To resolve this paradox and clarify the precise role of dopamine in sleep-wake regulation, we employed a combination of approaches in mouse models, including optogenetics, fiber photometry with genetically encoded fluorescent sensors, and simultaneous EEG/EMG/video recordings. Our findings reveal that dopamine does not merely promote wakefulness; it also plays a critical role in regulating sleep homeostasis—the balance between sleep and wakefulness. Notably, dopamine supports this regulation through two distinct, time-dependent mechanisms.

Disclosures: X.L. Huang: None. X. Tu: None. Y. Dan: None.

Poster

PSTR290: Physiology and Models Advancing New Theories of Sleep

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR290.16/BB15

Topic: G.07. Biological Rhythms and Sleep

Title: Sleep Regularity As A Mediator Between Caffeine Consumption and Pain Reports

Authors: *E. SKAER¹, M. KELLEY¹, A. SABBAGH¹, M. IMAN¹, Y. SHEN¹, W. J. HORTON²;

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Abstract: An estimated 85% of the U.S. population consumes caffeine on a daily basis, and chronic pain is one of the most pervasive public health issues in the nation. Numerous studies have shown an association between caffeine intake and self-reported pain levels. However, the directionality of this association is conflicting, and the underlying mechanism has yet to be fully elucidated. We proposed sleep as a novel mediator that explains the relationship between caffeine intake and subjective pain. Further, we hypothesized that increased caffeine intake leads to increased sleep irregularity, which in turn increases reported pain levels. To test this, daily sleep measurements were collected on participants aged 18-24, over a three-week period (n=32). Sleep data was collected via actigraphy (Actiwatch Spectrum; Philips Respironics, Murrysville, PA), and sleep regularity was calculated using the sleep regularity index (SRI), which quantified the consistency of a person's sleep-wake patterns over a week period. Caffeine intake was a self-reported measure, using a daily survey where participants listed beverages and medications containing caffeine. Caffeine content (in mg) was calculated using a detailed reference sheet and treated as a continuous variable. Pain was a self-reported measure, using a daily survey where participants reported their pain according to a numerical scale (1-100) reflecting pain

experienced on that day. Initial Pearson correlation coefficients were used to assess relationships between caffeine intake, sleep regularity, and pain averaged over a week's period for each participant. Preliminary analyses showed a significant relationship between caffeine and pain ($r = .167$, $p < .001$), however has yet to indicate an association between caffeine intake and sleep regularity ($r = 0.314$, $p = 0.22$). The absence of statistical significance may reflect limited statistical power due to the preliminary sample size; full analyses are currently underway and will be presented. Results from this study allow a further explanation of the complex relationship between caffeine consumption, subjective pain, and sleep quality. Understanding of this interplay contributes to a better understanding of how daily behaviors influence overall health.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.01/CC1

Topic: H.02. Reward and Appetitive Learning and Memory

Title: Neural correlates of hierarchical contextual control in associative reward prediction

Authors: *A. L. MAHERAS¹, J. SULLIVAN², R. KEIFLIN²;

¹Molecular, Cell. and Developmental Biol., ²Psychological and Brain Sci., Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: Reward-predictive cues exert strong control over behavior, but their motivational significance is often ambiguous and context-dependent (e.g. a “shot” has a different meaning in a bar vs a vaccine clinic). To resolve this ambiguity, animals can use contextual cues as hierarchical “gatekeepers” (or occasion-setters) that guide the interpretation of ambiguous by cues by selecting the appropriate associative memories. Behavioral studies and theoretical models suggest that the motivational “eliciting” and hierarchical “gatekeeping” properties of cues are functionally and neurobiologically dissociable. However, the neural circuits supporting this hierarchical contextual gating—and how they differ from those involved in simpler cue-elicited behaviors—remain elusive. To identify the functional networks underlying contextual gating, we performed brain-wide c-Fos activity mapping in rats trained on one of four Pavlovian discrimination tasks. Each task involved two auditory cues (X, Y) presented in two visual contexts (LIGHT, DARK) and was designed to engage distinct associative architectures: (1) Ctx-dep. O1: cue X was rewarded only in LIGHT, cue Y only in DARK, both yielding the same outcome—critically engaging contextual gating; (2) Ctx-dep. O1/O2: similar reward contingencies as Ctx-dep. O1, but with differential outcomes (O1 for LIGHT:X, O2 for DARK:Y), allowing for non-hierarchical solutions to this discrimination problem; (3) Simple Discrimination: cue X always rewarded, Y never rewarded, regardless of context; (4) No Discrimination: both cues probabilistically rewarded in both contexts. The last two tasks did not

engage contextual gating. Following stable task performance, brains were processed for c-Fos immunofluorescence and c-Fos⁺ neurons were quantified across 48 brain regions. Interregional correlation matrices were generated to characterize functional networks associated with each task. Representational similarity analysis revealed that Simple Discrimination and No Discrimination tasks produced relatively similar c-Fos correlation patterns, whereas Ctx-dep. O1 engaged a distinct functional network. The Ctx-dep. O1/O2 condition shared features with both Ctx-dep. O1 and the simpler tasks, consistent with its hybrid associative demands. Notably, network analyses identified two regions—the claustrum and pulvinar—as key functional hubs uniquely recruited in the Ctx-dep. O1 task. These regions, traditionally implicated in large-scale cortical coordination, emerge here as candidate substrates for the hierarchical contextual gating in reward prediction.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.02/CC2

Topic: H.02. Reward and Appetitive Learning and Memory

Title: Reward memories bias instrumental rule selection via orbitofrontal projections to the secondary motor cortex

Authors: *M. S. LE¹, A. MAHERAS², S. PETERSON¹, R. KEIFLIN¹;

¹Psychological and Brain Sci., ²Molecular, Cellular, and Developmental Biol., Univ. of California Santa Barbara, Goleta, CA

Abstract: Reward-predictive cues can influence decision-making and promote the pursuit of specific outcomes. This influence of cued reward memories on decision-making is classically studied using the specific Pavlovian-to-Instrumental Transfer (PIT) paradigm. In this paradigm, stimuli established as Pavlovian predictors of specific outcomes can bias action selection and promote the instrumental action (often a left or right lever press) directed toward the cued outcome. However, the traditional PIT paradigm fails to account for the fact that, beyond specific discrete actions, reward pursuit typically involves multiple actions organized into logical sequences and orchestrated by goal-specific abstract rules. The influence of cued reward memories on rule selection has received less attention, and the neural circuits mediating this effect remains largely unknown. Here we adapted the PIT paradigm to investigate how Pavlovian reward cues bias instrumental rule selection. The implication of the orbitofrontal cortex (OFC) projections to the secondary motor cortex (M2) to this effect was tested using pathway-specific chemogenetic silencing. Rats expressing hM4Di or mCherry in OFC→M2 projection neurons were first trained on a 3-step instrumental sequence task (sample lever - nose poke - choice lever). In alternating blocks, rats learned that two different rules could be used to complete the instrumental sequence, each rule corresponding to a different outcome. Specifically, in “delayed

non-match to sample” blocks, rats learned to select the non-sampled lever (and disregard a visual cue information) to obtain outcome 1 (dNMTS-O1). Conversely, in “visual” blocks, rats learned to select the visually cued lever (and disregard the previous sampling information) to obtain outcome 2 (VIS-O2). In a second phase, two distinct auditory cues were established as Pavlovian predictors of the two outcomes (S1-O1; S2-O2). Finally, in non rewarded probe tests preceded by DCZ injections, we measured how the presentation of these Pavlovian cues biased instrumental choices and promoted the adoption of an abstract rule. In control (mCherry) rats, Pavlovian cues promoted the adoption of the rule corresponding to the cued outcome (S1 promoted dNMTS and S2 promoted a visual rule). In hM4Di rats, DCZ-mediated inactivation of OFC→M2 projection neurons abolished this effect. Importantly, while OFC→M2 silencing abolished cue-based (i.e. memory-based) rule selection, it spared outcome-based (i.e. evidence-based) rule selection.

Disclosures: M.S. Le: None. A. Maheras: None. S. Peterson: None. R. Keiflin: None.

Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.03/CC3

Topic: H.02. Reward and Appetitive Learning and Memory

Support: Hellman Fellowship to R. Keiflin

Title: Cue- and Pathway- specific involvement of the Orbitofrontal cortex in context-gated reward predictions.

Authors: *R. KEIFLIN¹, S. PETERSON²;

¹Psychological & Brain Sci., Univ. of California, Santa Barbara, Santa Barbara, CA;

²Psychological & Brain Sci., Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: Reward cues can be powerful motivational triggers, but their meaning is often ambiguous and context-dependent (what is good in one context is not necessarily good in another context). To resolve this ambiguity, animals form hierarchical associations in which the context gates the retrieval of appropriate cue-evoked memories. The orbitofrontal cortex (OFC) is thought to support such context-dependent hierarchical predictions, and prior work implicates the lateral OFC in the contextual regulation of cue-elicited reward seeking. However, the specific OFC output pathways that mediate this top-down contextual control remain unclear. Here, we used a pathway-specific optogenetic approach to silence OFC terminals in either the central dorsal striatum (cDS) or mediodorsal thalamus (MDT) while rats performed a context-dependent Pavlovian discrimination task. Rats expressing the inhibitory opsin eNpHR3.0 (or the control transgene mCherry) in OFC neurons were trained on a task in which a visual contextual cue (A) informed the predictive value of two auditory target cues (X and Y): one cue was rewarded only in the presence of A, and the other only in its absence (A:X+ / X- / A:Y- / Y+). Once rats

reached stable discrimination performance, we tested the effect of OFC pathway silencing during cue presentation. Silencing OFC terminals in the cDS profoundly disrupted the contextual regulation of reward seeking. In contrast, silencing OFC terminals in the MDT had no significant overall effect. Further analysis revealed that the effects of OFC pathway silencing were highly cue-specific. Indeed, OFC→cDS silencing completely abolished the contextual regulation of cue Y responses (negative gating) but had only modest impact on responses to cue X (positive gating). Conversely, OFC→MDT silencing slightly reduced the regulation of cue Y responses, but, surprisingly, improved the contextual regulation of cue X responses. Together, these findings identify functionally distinct roles for OFC projections in the contextual regulation of reward seeking. The results also suggest that positive and negative contextual gating may rely on separate associative and neural architectures, offering new insight into how the brain flexibly integrates context to guide behavior.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

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Program #/Poster #: PSTR291.04/CC4

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NSTC 112-2628-B-002-011
NSTC 113-2628-B-002-002

Title: Cerebellum-vta gabaergic projections sculpt choice-outcome coupling in reward-based learning

Authors: *P. CHEN¹, L.-Y. LU^{2,3}, M.-K. PAN⁴;

¹Grad. Inst. of Pharmacology, Natl. Taiwan Univ. Col. of Med., Taipei, Taiwan; ²Inst. of Biomed. Sci., Academia Sinica, Taipei City, Taiwan; ³Dept. of Med. Research, Natl. Taiwan Univ. Hosp., Taipei, Taiwan; ⁴Inst. of Pharmacol., Natl. Taiwan Univ. Col. of Med., Taipei, Taiwan

Abstract: Reward-based learning, a critical ability to let us adapt in the changing world, based on a fundamental process of coupling our choices and behavioral outcomes. Despite a wealth of evidence underpinning of reward outcome processing, it is not clear how the formation of choice-reward coupling arises in the brain.

Numerous studies identified cerebellum has the direct excitatory projection to ventral tegmental area (VTA) from deep cerebellar nucleus (DCN), which supports dopamine neurons in the VTA to optimize computing reward prediction error (RPE). Interestingly, our anatomical tracing and functional assays reveals GABAergic cerebellar nuclei neurons also provide projection to VTA. Here, we report that GABAergic DCN-to-VTA projections regulate reward-based learning by coupling choices and outcomes. VTA-innervating GABAergic DCN neurons show sustained

activity during the period of choice and reward. The neuronal activity does not report anticipation and is unrelated to rewarded timing and history, supported by both single-unit and fiber photometry recordings. Disruption of axonal activities of choice-outcome coupling not only impaired behavioral learning, but also led to unlearning and choice difficulties in well-learned mice. In contrast, manipulating these GABAergic neurons during the period of reward consumption did not change the behaviors, while the same manipulation to glutamatergic neurons leads to rewarding behaviors. The GABAergic DCN neurons not only coupling the choice-outcome events but also the timing. Optogenetically prolonged or shortening the cell-firing duration the coupling period results in time shifting of conditioning reward licking. Taken together, these results extend our understanding of the cerebellum dynamically constructs choice-reward coupling, suggesting a potential mechanism in the acquisition of choice and reward outcome association.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

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Program #/Poster #: PSTR291.05/CC5

Topic: H.02. Reward and Appetitive Learning and Memory

Support: ERC (funded by UKRI, EP/X026655/1)
Royal Society (NIF\R1\232039)

Title: Dopamine neurons show diverse integration of sensory evidence and reward value during decision-making

Authors: *J. MORETTI¹, S. LIEBANA GARCIA², A. LAK¹;

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Abstract: Making decisions among stimuli requires integration of available sensory evidence and the reward values that stimuli predict. Past studies show that midbrain dopamine neurons encode these variables, integrating them into reward prediction errors to guide decision-making. However, these studies have two shortcomings. First, they used sensory decision tasks in which reward values were changed in blocks of trials (asymmetric sized rewards for left and right choices), rather than using stimulus features to indicate sensory evidence and reward value in a trial-by-trial fashion. Second, they either recorded the spiking activity of one dopamine neuron at a time, or imaged bulk dopamine activity. As such, the extent to which different dopamine neurons integrate available sensory evidence and reward value remains unclear. To address these, we designed a visual decision task for head-fixed mice and used two-photon imaging through an implanted GRIN lens to simultaneously record activity of multiple individual midbrain dopamine neurons. In each trial, a sinusoidal grating appeared on the left or right side of the screen, the contrast of which varied to parametrically change available sensory evidence in

different trials. Moreover, in each trial, the orientation of the grating indicated whether the correct choice yielded a small or large water reward. Mice turned a wheel to report the position of the stimulus, and received the corresponding reward for a correct choice. Mice performed the task with high accuracy, and their choices reflected both the trial-by-trial level of sensory evidence and the size of pending reward. Choice accuracies increased as a function of grating contrast, and were higher for gratings predicting larger rewards. Dopamine neurons showed substantial activity during the task that diversely reflected various task variables such as sensory evidence, reward value, and stimulus laterality. Regression analyses demonstrated that stimulus-locked neuronal responses significantly encoded one, or a combination, of these variables. Neurons representing both sensory evidence and stimulus laterality were the most common, followed by neurons that integrated sensory evidence, reward value, and stimulus laterality. Our ongoing analyses seek to further examine these neuronal signals and investigate the relationship between these signals and behavioral measures using reinforcement learning modelling. Our work addresses how sensory evidence and reward value drives decision making, and how different dopamine neurons integrate these variables differently to guide decisions.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

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Topic: H.02. Reward and Appetitive Learning and Memory

Support: ERC (funded by UKRI, EP/X026655/1)
Wellcome Trust (222807/Z/21/Z)
A*STAR (National Science Scholarship)

Title: Dorsomedial frontal cortical neuronal responses during a visually-guided probabilistic reward task

Authors: *S. TAN, M. TVRDIC, H. ATILGAN, A. LAK;
Univ. of Oxford, Oxford, United Kingdom

Abstract: Dorsomedial frontal cortex (dmFC) plays crucial roles in shaping diverse aspects of behavior including sensory processing, reward expectation, and action planning. Several studies have shown that dmFC neurons encode the expected value of stimuli and action plans to obtain rewards. However, the relative contribution of these variables in shaping dmFC neuronal responses remains unclear. Moreover, it remains unknown whether different populations of dmFC neurons, defined by their striatal projections, differentially encode these variables. To address these questions, we trained mice in a Pavlovian conditioning task in which different visual stimuli were associated with different probabilities of reward (0, 50, or 100%). We used two-photon calcium imaging to record the activity of all dmFC excitatory neurons, and a subset

of them projecting to the dorsal striatum during the task. Mice showed predictive licks that scaled with the cued probability of reward, indicating that they learned the task. As expected, the magnitude of the stimulus-locked dmFC population neuronal responses increased as a function of reward probability. Our analyses so far have uncovered two characteristics of these neuronal responses. First, in trials without predictive licks, dmFC neuronal responses to stimuli strongly reflected reward probabilities. Second, in trials with the same stimuli (e.g. 50% reward), neuronal stimulus-locked responses differed in a trial-by-trial manner, depending on whether the animal licked or not. Thus, the results so far indicate that dmFC neurons reflect the expected value of stimuli, and are also modulated by actions to obtain rewards. Our ongoing analyses suggest that dmPFC neuronal responses better reflect reward expectation than action planning. We continue to further analyse these, and also investigate whether these neuronal representations depend on the striatal projection targets. Together, these experiments will provide insights into frontal neuronal representations linking stimuli, actions, and reward, and whether these representations are mapped onto distinct frontal-striatal circuits.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

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Program #/Poster #: PSTR291.07/CC7

Topic: H.02. Reward and Appetitive Learning and Memory

Support: ÚNKP-23-3-II-SE-24
EFOP-3.6.3-VEKOP-16-2017-00009
Gedeon Richter Excellence PhD Scholarship
AMAT Award for Young Investigators

Title: Novel brainstem reward center inhibits lateral habenula and induces reward-seeking behavior

Authors: *K. ZICHÓ^{1,4}, B. BALOG^{1,5}, R. SEBESTÉNY¹, J. BRUNNER², V. TAKACS¹, A. M. BARTH¹, C. SENG⁶, Á. OROSZ¹, M. ALICZKI³, H. SEBOK¹, E. MIKICS³, C. FOLDY⁶, J. SZABADICS², G. NYIRI¹;

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Abstract: The lateral habenula (LHb) regulates behavioral flexibility that is essential for effective decision-making. LHb activity can facilitate aversion or reward-seeking behavior as a function of its excitatory/inhibitory inputs. However, despite LHb abundant connections with

hindbrain areas, the role of brainstem inputs is still unclear. Using viral tract tracing, in vivo electrophysiology, single-cell sequencing, and optogenetic behavioral experiments in transgenic mice, we investigated a previously unrecognized gamma-aminobutyric acid (GABA)-ergic cell population in the pontine central grey, named subventricular tegmental nucleus (SVTg), and its role in reward-seeking behavior. We found that their stimulation induced place preference. Moreover, mice self-stimulate their SVTg, which can also be activated directly by the orbitofrontal cortex, resulting in fast and effective inhibition of the LHb. Conversely, inhibition of SVTg caused place avoidance and disrupted reward location memory formation. By mapping the genetic profile of SVTg neurons and identifying their specific markers (*Satb1*, *Pax6*, *Reln*), we successfully identified them in the brainstem of both primates and humans. Our results suggest that a novel GABAergic pontine nucleus that innervates the LHb has a major role in processing positive valence and reward-seeking behavior and it might have a role in mood-related disorders. **Funding:** This project was supported by the ÚNKP-23-3-II-SE-24 New National Excellence Program of the Ministry of Innovation, by the EFOP-3.6.3-VEKOP-16-2017-00009 Semmelweis 250+ Excellence PhD Fellowship, sponsored by Gedeon Richter Talentum Foundation in the framework of Gedeon Richter Excellence PhD Scholarship of Gedeon Richter and by the Association of Hungarian American Academicians.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.08/CC8

Topic: H.02. Reward and Appetitive Learning and Memory

Title: Novel brainstem reward center activates during reward and balances negative experiences

Authors: *B. BALOG^{1,2}, K. ZICHÓ¹, R. SEBESTÉNY¹, J. BRUNNER³, V. T. TAKACS⁵, A. M. BARTH⁶, C. SENG⁷, Á. OROSZ³, M. ALICZKI³, H. SEBOK¹, E. MIKICS⁴, C. FOLDY⁸, J. SZABADICS⁹, G. NYIRI¹;

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Abstract: The lateral habenula (LHb) is an evolutionarily well-preserved brain structure involved in motivation and cognition. Using viral gene delivery in transgenic mice, we identified a novel inhibitory pontine cell population that projects to the LHb, which we named the

subventricular tegmental nucleus (SVTg). Fiber photometry revealed that rewarding and reward-predicting cues increased SVTg activity. Optogenetic stimulation of the SVTg induced dopamine (DA) release in the nucleus accumbens, further suggesting a role in reward processing. Surprisingly, aversive airpuffs also activated the SVTg, raising the question of whether it plays a broader role in emotional regulation. Indeed, optogenetic inhibition of the SVTg during fear memory acquisition led to an increase in contextual and generalized fear. Additionally, SVTg activation decreased anxiety-related behavior in the elevated plus maze. In vitro electrophysiological recordings showed uniform spontaneous excitatory postsynaptic potentials (EPSPs) and burst firing across all SVTg neurons, with serotonin further exciting them. These findings suggest that the SVTg modulates both rewarding and aversive experiences, potentially acting as a buffer against excessive negative emotional states. Given this role, dysfunction in the SVTg may contribute to mood disorders such as depression and anxiety.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

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Topic: H.02. Reward and Appetitive Learning and Memory

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NIH Grant F31MH135679
Brain and Behavior Research Foundation NARSAD Young Investigator Award 27735
Klingenstein-Simons Fellowship Award

Title: Tachykinin 1 neurons in the habenula signal reward prediction error

Authors: ***K. SUZUKI**¹, T. SEAGOE¹, B. HOLCOMB², J. KUYAT¹, E. L. SYLWESTRAK¹;
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Abstract: Evaluating outcomes to accurately predict which actions lead to reward is crucial to survival. In several reward-related brain regions, neuronal activity reflects the discrepancy between predicted outcomes and realized outcomes, termed reward prediction error (RPE). The lateral habenula (LHb) plays a well-established role in reward-guided decision-making and contains a subset of neurons that encode negative RPE: they are excited by worse-than-expected outcomes and inhibited by better-than-expected outcomes. The transcriptomic identity of RPE encoding neurons is unclear, limiting the use of genetic approaches to perform anatomical

tracing or causal manipulations to understand their specific role in outcome evaluation. Using cell type-specific recording of habenula neurons in mice performing a reward-guided task, we show that the gene tachykinin 1 marks a population of neurons (LHb^{Tac1}) that is excited by reward omissions. We found that reward outcome responses were sensitive to task variables that altered both the predicted value (reward probability and recent reward history) and the realized value (reward size), and scaled with the magnitude of the difference. These data suggest that LHb^{Tac1} neurons encode reward prediction error on a population level and point to a potential role for LHb^{Tac1} neurons in integrating past success or failure to drive dynamic reward-guided behavior. Additionally, LHb^{Tac1} neurons showed very little activity modulation to other aspects of the task or the presentation of innately aversive stimuli, suggesting LHb^{Tac1} neurons may represent a genetically defined subpopulation of habenular neurons that preferentially encode RPE. Together, these data demonstrate the cell type-specific contributions of habenular neurons to reward prediction error encoding and open avenues for a more precise investigation of neural circuitry underlying reward-guided behavior.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.10/CC10

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIH 2RO1 DA047870
NSF GRFP

Title: Probing anterior cingulate cortex-ventral tegmental area pathway contributions to reward learning and decision making

Authors: *M. E. ORTEGA¹, L. MERGELIAN², A. IZQUIERDO²;

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Abstract: Ventral tegmental area (VTA) neurons of the midbrain and their diverse projections enable actions, support cue-based learning, and modulate executive functions. However, comparatively less is known about how frontocortical input to VTA support these behaviors. Previous studies have shown that there are robust afferents originating from PFC subregions like anterior cingulate cortex (ACC) into VTA (Souza et al. 2022). ACC projects to VTA, and while less dense than those sent from orbitofrontal cortex (Watabe-Uchida et al. 2012; Souza et al. 2022) ACC - VTA projections have been previously found to support cue-motivated reward-seeking (McKendrick et al. 2022). Moreover, ACC has been found to generate reward prediction errors, like VTA, which support learning and decision making. To probe this circuit, we first sought to virally trace the ACC-VTA pathway and their collaterals in rats using a dual-virus

chemogenetic approach. Rats were prepared with bilateral cre-dependent DREADDs under a hSyn promoter in ACC and retrograde cre in VTA (N=4, n=2 with 6-weeks and n=2 with 9 weeks expression time). We were able to visualize the DREADDs in deep layer 5 of ACC and terminals in VTA following 9-weeks and found that the ACC-VTA pathway reliably expressed collaterals in the retrosplenial cortex and subiculum. We then prepared (N=7) rats (4 females) with the same combination of viruses, this time testing them on the influence that Pavlovian cues exert over instrumental responding, probed with Pavlovian-to-instrumental transfer (PIT). We found that systemic pathway inactivation during testing but not learning resulted in attenuated outcome-specific responding, suggesting a role of this pathway in regulating cue-motivated reward-seeking. With some evidence of necessity, we next probed this circuit with increased pathway specificity by preparing rats (N=6, 3 females) with inhibitory DREADDs in ACC and chronic indwelling cannulae in VTA. Rats underwent PIT training and testing with pathway inhibition at test. Our preliminary findings further support that selective inhibition of the ACC - VTA pathway results in attenuated, but not blocked, outcome-specific responding. Finally, we studied this pathway with more temporal specificity, using optogenetics to selectively silence ACC - VTA across cue presentation during PIT testing (N=8, 5 females). Our preliminary optogenetic results show that ACC - VTA inhibition decreased responding for a less preferred outcome but remained intact for preferred outcomes. Our ongoing work seeks to elucidate the function of ACC - VTA projections in guiding decisions towards rewarding outcomes when rewards are equally valued.

Disclosures: M.E. Ortega: None. L. Mergelian: None. A. Izquierdo: None.

Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

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Topic: I.03. Decision Making

Support: NIMH T32 MH119049
Canadian Institute for Advanced Research (Fellowship in Brain, Mind, & Consciousness)
Air Force Office of Scientific Research (FA9550-20-1-0106)

Title: Precision-weighted integration explains dynamic effects of expectations on perceptual decisions

Authors: *A. KHOUDARY, M. A. PETERS, A. BORNSTEIN;
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Abstract: How do expectations support efficient decision-making in noisy, uncertain environments? The standard approach posits that expectations-observers' knowledge of the prior probability of choice outcomes-bias the starting point of an evidence accumulation process

toward the expected outcome. This approach is statistically powerful, supported by empirical data, and sometimes optimal, but rests on a critical assumption: the effect of expectations is constant within a single decision. Recent neural and behavioral evidence challenges this assumption, finding effects of expectations change over the course of a decision (Hanks et al., 2011; Bornstein et al., 2023). These effects were observed by different researchers using distinct tasks, species (humans vs non-human primates), and neural measures, and thus were explained using different evidence accumulation models. We present simulation results showing that these findings can be unified by a single model along with experimental results supporting the model's core theoretical predictions. Our model posits that expectations are a dynamic evidence source that is sampled in parallel to vision, and integration into the decision process according to the time-varying relative precision of each evidence source. The core prediction is that expectations should dominate the decision process specifically at moments of low sensory precision. We tested this prediction in a novel task performed by healthy adults (n=44; male=7, female=34, nonbinary=3; mean age=20.64 years). Observers learned probabilistic cue-image associations (e.g., that a red border predicts Scene A 80% of the time), which we then used to manipulate their expectations trial-by-trial while they decided which of two perceptually similar scenes dominated a rapidly-alternating (60Hz) visual evidence stream. Crucially, on each trial, we interrupted the visual stream with stochastic epochs of pure visual noise. Our model predicts that drift rates of the accumulation process should be non-zero during these noise periods, with magnitudes scaled by the cue's predictiveness on each trial. Conversely, the standard model predicts no impact of noise periods on drift rates. Formal model comparisons found that accuracy and reaction times were better explained by our model than the standard model ($\Delta\text{BIC}=-195$) despite our model being ~2x as complex (14 vs 7 free parameters). Further, we found that noise period drift rates were positively modulated by cue predictiveness ($t_{83}=2.022$, $p=0.046$). Taken together, these findings suggest that precision-weighted integrated offers a unifying explanation for dynamic effects of expectations on perceptual decisions.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

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Program #/Poster #: PSTR291.12/CC12

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIMH R01 MH118545
NIMH R01 MH096478
N01HD13334
N01HD63258

Title: Prenatal maternal inflammation and offspring white matter connectivity: implications in reward circuitry in late midlife

Authors: ***R. MOPURU**¹, B. L. ELLIOTT², L. HOFFMAN², N. TAN², A. KRING⁶, E. BREEN⁷, B. COHN⁸, P. CIRILLO⁸, N. KRIGBAUM⁸, M. D'ESPOSITO⁶, A. COGAN⁶, B. PATWARDHAN⁶, T. OLINO³, I. R. OLSON⁴, L. ELLMAN⁵;

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Abstract: Background: Prenatal maternal inflammation (PNMI) has been associated with a range of neurodevelopmental changes in human offspring. Preclinical work suggests that dopaminergic signaling is impaired in offspring as a consequence of PNMI. The ventral tegmental area (VTA), a key nucleus in the mesolimbic dopamine system, primarily projects to the limbic striatum, and more sparsely to the hippocampus. Given hippocampal vulnerability to inflammatory insult, with PNMI being associated with reduced hippocampal neurite density in a previous study, VTA-hippocampal connectivity was investigated in the current study. Methods: Participants included 89 mother-offspring dyads from the Healthy Brains Project cohort with inflammatory biomarker data [interleukin (IL)-6, IL-8, IL-1 receptor antagonist (IL-1RA), and soluble TNF receptor-II (sTNF-RII)] from first (T1) and second trimester (T2) maternal sera. Diffusion MRI scans were collected in offspring aged 57-63. Probabilistic tractography was used to delineate mesolimbic VTA tracts to the hippocampus and limbic striatum independently. For macrostructural analyses, hierarchical linear regressions were conducted to assess the effects of PNMI on offspring streamline density. To assess microstructural integrity, neurite density was estimated using the neurite orientation dispersion and density imaging (NODDI) model. Each tract was split into 100 nodes, and neurite density was extracted at each node. A nodewise permutation model with linear regressions was used to identify consecutive nodes that were associated with PNMI, allowing for spatial insight into where along the tract PNMI correlated with tract microstructure. Analyses controlled for intracranial volume and maternal education. Results: Higher levels of PNMI (T1 IL-1RA & sTNF-RII, and T2 IL-8) was associated with decreased streamline density in the right VTA-Limbic striatum tract. In microstructural models, higher T1 IL-8 was associated with reduced neurite density in significant clusters within the left VTA-hippocampus tract closer to the hippocampus, and in the right and left VTA-Limbic striatum tracts closer to the VTA. Higher T2 IL-6 was positively associated with neurite density within a significant cluster in the right VTA-hippocampus tract towards the end of the tract. Conclusions: These findings highlight the association of PNMI with late midlife offspring reward circuitry changes with implications for understanding how prenatal exposure may contribute to offspring brain programming, and in turn, affective, motivational, and learning deficits across the lifespan.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.13/CC13

Topic: H.02. Reward and Appetitive Learning and Memory

Support: McDonnell Center for Systems Neuroscience

Title: Cortical neural landscape captures mouse-to-mouse variability in anticipatory vs. inattentive decision making.

Authors: *C. YIN¹, N. HIRATANI²;

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Abstract: Understanding individual variability in behavior is crucial for both basic and clinical neuroscience, yet it remains challenging to study in traditional single laboratory experiments with small sample size. Leveraging standardized behavioral and neural datasets from the International Brain Laboratory, comprising approximately 100 mice trained on a visual decision-making task, we investigated the structure and neural correlates of inter-animal behavioral variability. Using reaction time analysis and a deep learning-based embedding of individual animals, we uncovered large but low-dimensional differences in behavioral traits. Some mice consistently exhibited anticipatory responses, marked by fast reaction times, while others showed slower, more disengaged behavior. These behavioral profiles were consistent across sessions and laboratories, with female mice tending to show more anticipatory behavior than males. We hypothesized that this behavioral spectrum reflects differences in the depth of underlying cortical states, reflected in the temporal dynamics of neural activity. Supporting this idea, we found that the characteristic timescale of population activity, measured during both inter-trial intervals and spontaneous periods, correlated with an animal's anticipatory tendency across cortical areas, especially in medial visual areas. These findings suggest that individual differences in the cortical dynamics may underlie distinct decision-making strategies.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

Location: SDCC Halls B-H

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Topic: H.04. Emotion

Support: NIH Grant RF1AG076821
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Title: Mitochondrial Energy Transformation Capacity Influences Brain Activation During Sensory, Affective, and Cognitive Tasks

Authors: *K. BO¹, C. KELLY³, M. PICARD⁴, T. D. WAGER²;

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³Columbia Univ. Irving Med. Ctr. (CUIMC), New York City, NY; ⁴Columbia Univ., New York, NY

Abstract: Brain function relies on energy supplied by mitochondria through cellular energy transformation, but how cellular energetics constrain neurological function and cognition remains poorly understood. Genetic defects in mitochondrial DNA cause rare mitochondrial diseases (MitoD) that offer a unique window into the energetic foundations of cognition, shedding light on the neural processes that are most energetically constrained. In this study, we assessed functional magnetic resonance imaging (fMRI) on 29 participants with MitoD and 62 matched controls during resting state and tasks probing cognitive (N-back task), affective (cold pain), and sensory (multisensory visual and auditory perception) functions. MitoD individuals exhibited significant cognitive deficits across a range of functions, including executive function and working memory, mental and physical fatigue, low exercise tolerance, and low mood. These deficits were accompanied by markedly elevated blood levels of metabolic stress markers, including GDF15 and FGF21. Surprisingly, overall BOLD fMRI activity and connectivity largely intact across all tasks in MitoD individuals. However, those with more severe cognitive impairment and higher GDF15 levels showed reduced working memory-related activity, which in turn mediated poorer task performance. Conversely, individuals with relatively preserved cognitive function exhibited hyperactivation in working memory regions and working memory performance compared to controls, suggesting compensatory engagement of cortical systems in high-functioning MitoD individuals. These effects were weaker in the sensory domain and absent during affective (cold pain) processing, suggesting an energy hierarchy in the brain that prioritizes essential functions such as affective responses while downregulating more energy-demanding, complex cognitive processes when resources are limited.

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Poster

PSTR291: Neural Correlates of Reward-Based Decision Making and Learning

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR291.15/CC15

Topic: H.04. Emotion

Title: Prefrontal EEG Complexity Reveals Interactive Effects of Emotional Valence and Perceived Control

Authors: *W. CREEL, R. HARTMAN;
Loma Linda Univ., Loma Linda, CA

Abstract: Neural complexity, as measured by EEG entropy, reflects the brain's adaptive flexibility. Although emotional valence and arousal have been widely studied, the role of perceived control (dominance) in shaping neural complexity remains underexplored. We examined how emotion type and dominance interact to influence prefrontal EEG complexity during emotional film clips. Using the DREAMER dataset (n = 23), EEG was analyzed from AF3 and AF4 during 18 video trials per participant. Valence was categorized based on target emotion (positive vs. negative), and dominance was defined from self-reports (≤ 2 = low, ≥ 4 = high). Multiscale entropy was computed across 20 scales and averaged across frontal channels. A linear mixed-effects model tested main and interaction effects of valence and dominance on standardized SampEn, with subject as a random factor. Positive valence and low control were each associated with higher entropy ($\beta = +0.40$ SD, $p = .014$; $\beta = +0.46$ SD, $p = .049$). However, a robust interaction ($\beta = -0.73$ SD, $p = .016$) showed that low control increased entropy for negative stimuli, while high control increased entropy for positive stimuli. High entropy under low-control negative states may reflect emotional dysregulation and regulatory effort. In contrast, high control over negative stimuli may reduce threat appraisal, requiring fewer cognitive resources. For positive stimuli, high control may signal opportunity and promote engagement, while low control elicits less intensive processing. These findings suggest EEG entropy reflects not just valence but the contextual influence of perceived control, supporting complexity as a dynamic marker of emotional adaptability.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Program #/Poster #: PSTR292.01/DD1

Topic: H.04. Emotion

Support: R01MH127214
R01MH137461
Vallee Scholars Award
F32MH133387
ARCS Foundation Fellowship

Title: Directed cell-type recruitment during consolidation of a remote memory

Authors: *Z. ZEIDLER¹, A. YU², L. A. DENARDO²;
²Physiol., ¹UCLA, Los Angeles, CA

Abstract: Memories are consolidated into a distributed neocortical network for long-term storage. Long-term memory retrieval relies on cells that are active during learning and undergo

necessary plasticity. However, remote memory retrieval activates a broader circuit, with learning-activated cells representing only a small subset. What are the rules and cell-types governing memory trace reorganization? We identified a class of prefrontal projection neurons that are gradually recruited to a memory trace through synaptic activity of learning-activated cells. This population, which projects to the temporal association area (TEa), progressively strengthens its encoding of memory-induced behaviors, mirroring increases in TEa activity. Notably, the prefrontal-TEa pathway is required for remote but not recent memory retrieval. Our findings reveal a cell type-specific mechanism underlying memory trace reorganization during consolidation.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Program #/Poster #: PSTR292.02/DD2

Topic: I.03. Decision Making

Support: ARO MURI W911NF2110328
Tianqiao & Chrissy Chen Center for social and decision neuroscience

Title: A neurocomputational account of hierarchical goal selection in humans

Authors: *S. AENUGU, J. P. O'DOHERTY;
Caltech, Pasadena, CA

Abstract: Goals that humans pursue are often hierarchical and temporally-extended in nature. In the presence of multiple such competing goals in dynamic environments, adaptive goal control necessitates evaluating the viability of various goals and their constituent subgoals to decide whether to commit to a specific goal or switch to an alternative goal. In this study, we investigated the mechanistic and computational basis of adaptive goal control in humans by employing a hierarchical goal selection paradigm inspired by board games. In the game, participants can freely choose to undertake one of three projects at any given moment (building a spaceship, house, or observatory) for which they need to collect different resources (stone, glass, metal, concrete) in specific proportions. The availability of different resources was changed across the experiment creating differences in the probability of achieving particular goals over time. To perform well on the task, participants need to adjust their building goals across time depending on resource availability. We analyzed human performance in the task (online: n=30; fMRI: n=26). We found evidence that one key strategy that participants relied on to solve this task is “goal momentum”, a model-free estimation of the speed of goal progress, which takes into account both prior progress and an estimate of current progress rate. Using neural data analyses, we show that the ventromedial prefrontal cortex (vmPFC) tracks goal momentum. Higher vmPFC activity is also observed when participants chose to stay with a previous goal,

while activity in frontopolar cortex and cerebellum are correlated specifically with switching of higher-level goals and not their constituent subgoals. Our work lends insight into mechanistic processes in the brain that encode, sustain, and adapt value representations to facilitate flexible goal pursuit in humans.

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Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

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Program #/Poster #: PSTR292.03/DD3

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIDA grant #R15DA051795

Title: Additive summation of same or different reward representations - neural correlates across Pavlovian and instrumental conditioning procedures

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³Psychology, Brooklyn Col. - CUNY, Brooklyn, NY

Abstract: When two reinforcing stimuli are presented in compound, reward expectations “summate,” prompting more reward-seeking than individual stimuli (e.g., Weiss, 1964). Models of associative learning (e.g., Rescorla and Wagner, 1972) appeal to this but do not explicitly address the finding that outcome-selective reward identity plays a role in learning (e.g., Corbit and Balleine, 2005). Research on whether summation is outcome-selective has been mixed (Rescorla, 1999, Watt and Honey, 1997) and none have investigated its neural basis. Experiment 1 replicated this procedure by comparing behavior in the presence of stimuli with that in the presence of their compounds, in a within-subject Pavlovian task. Compound stimuli were paired with either the same (“same” compounds) or different (“different” compounds) rewards. This revealed summation, marked by greater responding to the compound over individual stimuli. Responding was greatest for compounds of stimuli paired with the same reward, a result that would only occur if outcome-selective learning played a role. For Experiment 2, we searched for the integrative center of reward expectations using activity-dependent proteins following between-subject presentation of “same” and “different” compounds and individual stimuli. “Different” compounds decreased activity in the orbitofrontal cortex (OFC) and retrosplenial cortex (RSC). Compounds increased cells activated in the lateral hypothalamus and decreased their firing rate (Bertran-Gonzalez et al, 2012), paralleling summation. This did not reveal summation in behavior, however, suggesting these neural effects occurred below the behavioral threshold and begging the question of whether similar effects would be observed when behavioral summation is evident. Experiment 3 made reward delivery contingent on approach, revealing selective summation marked by greatest responding during “same” compounds in a

between-subject comparison amenable to neuroimaging. Experiment 4 therefore assessed neural activity in this procedure and revealed transient behavioral summation. “Different” compounds revealed a selective decrease in firing rate in the granular RSC, and a selective increase in cells activated in the OFC. Activation of these areas during “different” compounds in both Experiments 2 and 4 suggests interference between conflicting reward representations in these regions. “Different” compounds also resulted in a reduction in activation of central amygdala cells with dopamine projections. “Same” compounds, “different” compounds, and stimulus elements also exhibited different patterns of network connectivity.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Program #/Poster #: PSTR292.04/DD4

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIH OD011132
NIH R37 MH117103

Title: Projections from the orbitofrontal cortex to the premotor cortex drive exploitation learning

Authors: *L. BAHENA, S. L. GOURLEY;
Emory Univ., Atlanta, GA

Abstract: One key challenge we face in day-to-day life is the explore/exploit dilemma: deciding whether to try new methods (exploration) or stick with proven strategies (exploitation). The orbitofrontal cortex (OFC) and premotor cortex (M2) appear to individually influence the balance between exploration and exploitation; however, it is unclear whether or how these brain regions interact to coordinate this behavior. In the current study, mice were trained on and tested with three operant conditioning tasks designed to reflect exploration, exploitation, or neither strategy. We found that mice with OFC-to-M2 projection-specific inactivation were unable to learn, and therefore use, optimal strategies, resulting in a failure to engage in exploitative behavior. However, this manipulation had no consequences during tasks that required exploration or neither strategy. Our findings suggest that OFC-to-M2 manipulation does not broadly interfere with the mice’s ability to use adaptive strategies across different contexts, and instead selectively affects the learning of optimal strategies in particular circumstances requiring response exploitation.

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Poster

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Program #/Poster #: PSTR292.05/DD5

Topic: H.02. Reward and Appetitive Learning and Memory

Support: DA052385

Title: Ventral Tegmental Area to Nucleus Accumbens circuitry mediates memory reconsolidation induced by negative prediction error

Authors: *B. CHO¹, I. ACHESON¹, H. SANCHEZ², R. J. DILEONE³, J. TAYLOR⁴;

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Abstract: Drug-associated cues, or conditioned stimuli (CS), can elicit craving and trigger relapse even after long periods of abstinence. A promising strategy to mitigate drug-seeking and taking behaviors involves disrupting cue-associated memories through interference with the memory reconsolidation process. Critically, memory destabilization during reconsolidation is thought to require the induction of prediction error (PE). In this study, we investigated the role of negative PE in modulating drug-seeking behavior and related neural circuits. Rats were infused with a Cre-dependent inhibitory DREADD virus into the ventral tegmental area (VTA) and retrograde Cre recombinase into the nucleus accumbens (NAc). Rats were trained to self-administer (SA) cocaine: Lever pressing for cocaine was associated with two separate cues - tone (CS1) or light (CS2). For memory reactivation, rats were assigned to two groups: one group received a single presentation of CS1 with a smaller dose of cocaine to induce a negative PE, while the other group received a single CS1 presentation with the same dose of cocaine as they received during SA, which would not induce PE (no PE). Immediately afterwards, they received either CNO or vehicle to inhibit neural activity in VTA-to-NAc. The next day, they were tested for cue-induced reinstatement; rats that received CNO after a negative PE showed a significant decrease in lever presses for both CS1 and CS2 compared to vehicle group. Whereas, the rats in the no PE (CNO/vehicle) did not show any differences in cue-induced reinstatement. These data suggest that the cue-drug memory only became labile due to a negative PE that was sensitive to disruption of memory restabilization. These results also demonstrate that inhibition of VTA-to-NAc circuits after a negative PE-induced memory reactivation significantly decreased cue-induced cocaine-seeking behavior toward multiple cues. To further refine our understanding of VTA-to-NAc circuit in memory destabilization, a separate cohort of rats received CNO 30-min prior to the negative PE-induced memory reactivation session, thereby inhibiting the VTA-to-NAc circuit during the actual memory destabilization phase. Rats that received a CNO injection also showed a reduction in cue-induced reinstatement. These data suggest that the VTA-to-NAc circuit is involved selectively in the restabilization rather than the destabilization phase. Overall, our results indicate the specific role of VTA-to-NAc projections in the reconsolidation process initiated by negative PE, and give insights into the behavioral and neural mechanisms by which negative PE influences relapse-like behaviors.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Program #/Poster #: PSTR292.06/DD6

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIDA R01DA054967

Title: A novel task to probe the neural circuits involved in model-based learning

Authors: *A. DAWSON, J. LEAKE, A. ABIERO, M. SHARPE;
Sch. of Psychology, The Univ. of Sydney, Sydney, Australia

Abstract: Model-based learning refers to the development of knowledge that defines how specific cues and outcomes are related. This knowledge is then integrated into broader cognitive maps of the environment that allows us to make good decisions. Work from our lab has recently revealed that some neural populations bias model-based learning toward proximal cues that are most predictive of rewards, while actively biasing learning away from distal predictors of rewards. This reveals added complexity to model-based learning. Investigating the role of different circuits in proximal and distal model-based learning has been stunted by the lack of behavioural designs that can differentiate the roles of each neural population in these different aspects of model-based behaviour. The current study sought to overcome this limitation by developing a novel Pavlovian biconditional discrimination that is amenable to optogenetic dissection of the circuits involved in model-based learning of proximal and distal predictors of rewards. The task involves the presentation of one of two distal cues, followed by one of two proximal cues, where the relationship between the proximal cue and food is dependent on the preceding distal cue (e.g., click-->tone = reward // noise-->tone = no reward; click-->siren = no reward // noise-->siren = reward). In Experiment 1, we built on prior work to compare learning between rats that received distinct food outcomes following different trial types (i.e., differential reinforcement; sucrose vs. pellets) and rats that received the same food across rewarded trials (i.e., non-differential reinforcement; pellets). We demonstrated that differential outcomes facilitated learning of the biconditional discrimination. Next, in Experiment 2, we showed that optogenetic inhibition of neurons in the lateral orbitofrontal cortex (IOFC) across the proximal cue - but not the distal cue - disrupted successful prediction of reward. Given the IOFC is thought to play a role in navigating model-based cognitive maps, these results suggest our task involves such a structure. Finally, in Experiment 3, we confirmed the model-based nature of our task by showing that rats trained on the biconditional discrimination could transfer this learning to new cues using model-based inference. Together, these results establish a novel model-based task that allows dissociable investigation into the circuits that uniquely regulate learning about distal and proximal cues. Now, we are investigating the role of other neuronal populations in this task in the hopes of revealing how distinct neural circuits contribute in unique ways to model-based learning.

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Poster

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Program #/Poster #: PSTR292.07/DD7

Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIDA R01 054967
Uehara Memorial Life Science Foundation Research Fellowship

Title: A behavioural task to probe the balance of learning between distal and proximal cues to rewards

Authors: *M. TAIRA¹, A. DAWSON², A. ABIERO², K. GANESAN², J. LEAKE², M. G. MATTAR³, M. SHARPE²;

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Abstract: Learning associations between sensory stimuli and rewards enables reward predictions based on learned associations or cognitive maps. On some occasions, the sensory events that are predictive of reward have a different relative distance to those rewards. Some sensory events can be proximal predictors of rewards (e.g. a ramen stall selling your favourite ramen). However, at other times, distal cues can predict rewards via the association between itself and a proximal predictor (e.g. a trumpet sound signals the ramen stall is approaching). We have shown that GABAergic neurons in the lateral hypothalamus (LH_{GABA} neurons) differentially regulate learning about reward cues depending on the relative distance of the cues to rewards. This led to our hypothesis that LH_{GABA} neurons bias behaviours toward cues closest to reward and away from distal cues. To formally investigate this in rats, we adapted the “Daw two-step task”, which quantifies the ability of human subjects to use complex task structure to predict rewards. This task enables us to measure the weights of distal and proximal cues on future choices. Rats first receive one of two distal cues that are further from reward followed by presentation of two levers. Rats press one of the levers and then receive one of two proximal cues. The distal cues inform different state transitions from the levers to the proximal cues. In turn, the proximal cues inform the fluctuating reward probabilities (high/low). We found that rats can guide their choices by using the transitional structure of the task. We also found that using different rewarding outcomes following the distal cues allowed rats to better use the structure of the task to influence choice. Lastly, using optogenetics, we inhibited lateral orbitofrontal cortex (IOFC) in the different components of the task - specifically during the distal cue, proximal cue, and outcome delivery timepoints. Optogenetic inhibition of IOFC during the distal cue and the outcome delivery- but not the proximal cue- reduced the ability of rats to use the task structure to influence choice. Overall, these results demonstrate that our task can isolate roles for distinct neural populations in choice behaviour as dictated by model-based learning of distal and

proximal transition structures. Next, we will investigate the contribution of LH_{GABA} neurons to the different components of this task.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Topic: H.02. Reward and Appetitive Learning and Memory

Support: Wellcome Trust (213465/Z/18/Z)
Wellcome Trust (204651/Z/16/Z)
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Title: Reliable sequences of neuronal activity propagating through cortex and subcortex underlie perception

Authors: *B. RUSSELL, M. LYNN, E. NEWNHAM, A. M. PACKER, A. LAK;
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Abstract: Perception is inherently variable; the same external stimulus might be detected or missed in different instances. Previous studies have shown that perceptual variability could be partially explained by whether neuronal signals propagate from primary sensory cortices to higher cortical regions. In addition to these cortico-cortico projections, primary sensory regions also form substantial subcortical projections, e.g. to thalamus and striatum. However, it remains unknown how perceptual information is communicated between primary sensory cortices and their subcortical outputs. To investigate this, we trained mice to report optogenetic stimulation of primary somatosensory cortex (S1) by turning a wheel to receive a water reward. By requiring the animal to use direct activation of S1 to guide behaviour, the task dictates the directional flow of perceptual information by circumventing fanout at pre-cortical processing stages. We used Neuropixels to record electrophysiological neuronal activity from key S1 outputs, including secondary somatosensory cortex (S2), thalamus, and dorsolateral striatum (DLS), during expert behavior. We found that stimulation-evoked neuronal responses propagated with similar temporal delays from S1 to S2 and DLS, and subsequently reached thalamus ~100ms later. S2, thalamus, and DLS contained subpopulations of rate-coding neurons that encoded hit vs. miss trials in their stimulus-driven activity. These stimulus-driven activities propagated among neurons sequentially, forming a temporal neuronal code underlying perception. These sequences often originated in the somatosensory cortex and later spanned into DLS and thalamus, were not present on miss trials, and were not temporally correlated with choice-related movement. Our current experiments focus on two questions: first, to understand how the structure of early cortical activity governs subcortical neuronal signals and perception, we use targeted two-photon

stimulation of projection-specific, whisker-responsive, or reward-responsive S1 neurons whilst recording subcortical neuronal activity in trained mice. Second, to investigate whether distinct S1 subcortical projections have different roles in learning, we compare task learning across mice that selectively expressed opsin in either thalamus- or DLS-projecting S1 neurons. Overall, our results indicate that perception emerges from reliable sequences of neuronal activity propagating through cortical and subcortical brain regions.

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Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

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Program #/Poster #: PSTR292.09/DD9

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UCLA Graduate Research Mentorship Award
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Title: Gabaergic neurons dynamically promote consumption according to food value

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¹UCLA, Los Angeles, CA; ²NYU, New York, NY; ³Federal Univ. of ABC (UFABC), São Paulo, Brazil; ⁴CSULB, Lakewood, CA

Abstract: Adaptive feeding depends on the integration of internal satiety states and food value, yet the underlying neural circuits remain poorly defined. Recent work has identified GABAergic neurons in the lateral and ventrolateral periaqueductal gray (l/vIPAG) as key regulators of food-seeking and consumption. Here, we extend this framework by investigating how l/vIPAG vgat-expressing neurons regulate feeding in relation to food value and hunger state. Using optogenetic stimulation, we show that activation of l/vIPAG GABAergic neurons in vgat-cre mice robustly increases consumption of both low- and high-palatable foods. Notably, optogenetically-induced food consumption is greater for palatable caloric foods compared to vegetables, suggesting that these neurons promote feeding in proportion to perceived food value. Indeed, devaluation of a particular food source abolishes the effect of l/vIPAG activation, indicating that their influence on consumption is contingent on current food value. To examine endogenous activity, we performed fiber photometry and found that l/vIPAG vgat neuron activity increases prior to eating and is suppressed during consumption. Interestingly, systemic administration of ghrelin and leptin does not alter activity of these neurons, suggesting a dissociation between classic

hormonal hunger signals and PAG-mediated feeding. In agreement with these findings, only a small fraction (<9%) of l/vIPAG vgat neurons express either the leptin or ghrelin receptors. Together, our findings support a model in which l/vIPAG GABAergic neurons modulate food-seeking dynamically according to food value.

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Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

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Topic: I.03. Decision Making

Support: NIMH IRP ZIAMH002983
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Title: Heterogeneous neural encoding of expected delay to and amount of reward in the VS during an odor guided choice task

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Abstract: The ventral striatum (VS) is a critical node in the brain circuitry that supports reward learning. Neural activity in the VS has been shown to correlate with the value of expected rewards during reward-guided tasks, connecting directly to the dopamine neurons of the midbrain that are thought to compute reward prediction error signals that drive learning. Intact ventral striatal activity is also specifically implicated in learning about the timing of rewards, suggesting it may be uniquely important for this dimension of reward expectation. Here, we assessed the dynamic neural representation of the components of reward expectation in the VS, using single unit recordings from rats as they performed an odor-guided choice task in which the delay to and amount of reward were manipulated block-wise. We fit a series of generalized-linear encoding models (GLMs) to the spike counts of this VS ensemble at key points of a trial to test for separable encoding of delay and amount of reward in VS neural activity during learning. We found that a ‘delay-amount’ GLM outperformed a ‘value’-GLM at the level of the ensemble, specifically in the time period after reward delivery. A subset of individual neurons were better accounted for by the delay-amount GLM at all timepoints, suggesting a VS sub-population selective for the temporal features of expected reward. Neurons displayed marked heterogeneity in their dependence on trial-level reward conditions, including response direction-, delay- and

amount-selectivity. Further, we also found systematic changes in the configuration of VS neural activity that parallels the hidden block structure in the task. Together, these modeling results indicate that neural representations of predicted reward in the VS contain distinct dimensions of the expected outcome, allowing for a specific role for the VS in regulating the temporal dimension of reward prediction.

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Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

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Topic: I.03. Decision Making

Support: NIMH IRP ZIAMH002983
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Title: Previous exposure to cocaine alters behavioral and dopaminergic signatures of learning in rats

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Abstract: Chronic exposure to drugs of abuse can significantly impair learning about rewarding outcomes. The specific nature of this disruption to learning is unclear, and is hypothesized to either alter choice preferences for rewards, disrupt the update of those preferences following rewarding outcomes, or both. These changes to learning are thought to derive from drug-induced alterations to the dopaminergic circuitry that mediates reward learning in the brain. Here, we explored how choice preference during learning is impacted by previous exposure to cocaine. We analyzed choice patterns of rats that had previously undergone a cocaine self-administration procedure, along with controls, as they performed an odor-guided choice task in which delay to or amount of reward was systematically manipulated across blocks. On average, choice behavior across both groups was remarkably similar, with subtle perturbations to the evolution of choice preference induced in the cocaine-experienced rats. To further assess the effect of cocaine exposure, we fit trial-by-trial choices of rats to a series of reinforcement learning models. Across all rats, we found evidence that choice preference is differentially updated for a change in the timing of the reward compared to a change in the reward amount, and that this differentiation is specifically altered in the cocaine group. This is consistent with a selective effect of cocaine

exposure on learning in the presence of delays to reward. Cocaine-experienced rats also displayed fewer 'lapse' choices, indicating their preferences more stably followed previously learned values of the options. Using VTA dopamine recordings from each group acquired on the same task, we relate these group differences in choice and learning to the degradation of the phasic dopamine response induced by cocaine exposure.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Georgia Tech Julian T. Hightower Chair

Title: Evidence of Metabolic Cost Monitoring in the Human Anterior Cingulate Cortex

Authors: *T. ALBARRAN¹, C. MAY¹, X. VELEZ¹, K. KERR², K. HERRIN³, J. LEESTMA³, T. HARVEY³, R. HERSHENBERG⁴, A. YOUNG³, G. S. SAWICKI³, C. ROZELL¹, S. ALAGAPAN¹;

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Abstract: The dorsal anterior cingulate cortex (dACC) is hypothesized to play a central role in effort-based decision-making, with previous studies demonstrating that its neural activity tracks physical effort costs^{1,2}. However, most human studies have evaluated dACC function using stationary tasks (e.g., button pressing), leaving it unclear whether dACC activity encodes objective metabolic costs, subjective perceptions of effort, or changes in gait during naturalistic scenarios. To address this gap, we developed an incline walking task with synchronous brain-body data from 64-channel mobile electroencephalography (EEG), indirect calorimetry, treadmill force plates, and ratings of perceived exertion (Borg 6-20).

Healthy adults (N=15, 6 females) participated in six 6-minute walking trials, completing two trials for each incline (0°, 3°, 6°). EEG data were source-localized to the dACC (13 participants) and processed by removing aperiodic neural activity (1/f), extracting the canonical frequency bands, and Z-scoring. Weight normalized energy expenditure derived from indirect calorimetry and the resulting source features were averaged over the latter portion of each trial, i.e., metabolic steady state.

Linear mixed-effects models revealed that dACC theta ($\beta = 0.337$, $p = 0.008$) and high-beta ($\beta = -0.305$, $p = 0.023$) power were significantly associated with metabolic cost, whereas no significant relationships were observed with the Borg RPE measures. To assess the potential confounding effects of gait or neural control of movement, we performed an event-related spectral perturbation analysis synchronized to toe-off and heel-strike events. Although neural oscillations varied significantly across gait events ($p < 0.001$) primarily in the theta band, this gait-modulated neural activity did not significantly differ across treadmill incline.

By parametrically varying treadmill incline, we found that the dACC selectively tracked metabolic cost, potentially supporting its role as an integrator of lower-level physiological information to inform valuations of effortful actions. In addition, this study establishes an ecologically valid framework for comparing effort valuation in healthy individuals and clinical populations, which may inform future clinical interventions.

1. Porter BS, Hillman KL, Bilkey DK. Anterior cingulate cortex encoding of effortful behavior. *J Neurophysiol.* 2019 Feb 1;121(2):701-714. doi: 10.1152/jn.00654.2018. Epub 2019 Jan 9. PMID: 30625016.

2. Bradford, J. C., Lukos, J. R. & Ferris, D. P. Electrocortical activity distinguishes between uphill and level walking in humans. *Journal of Neurophysiology* **115**, 958-966 (2016).

Disclosures: T. Albarran: None. C. May: None. X. Velez: None. K. Kerr: None. K. Herrin: None. J. Leestma: None. T. Harvey: None. R. Hershenberg: None. A. Young: None. G.S. Sawicki: None. C. Rozell: F. Consulting Fees (e.g., advisory boards); Motif Neurotech, Inc.. S. Alagapan: None.

Poster

PSTR292: Reward Learning and Primary Rewards

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Topic: H.02. Reward and Appetitive Learning and Memory

Support: NIH BBQS - R61MH138966
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Title: The Role of Muscle Fatigue in the Perception of Effort During Incline Walking

Authors: *C. A. MAY¹, T. ALBARRAN², T. HARVEY³, K. HERRIN³, J. LEESTMA³, A. YOUNG³, G. S. SAWICKI³, C. ROZELL⁴, S. ALAGAPAN¹;

²Electrical Engin., ³Mechanical Engin., ⁴Electrical and Computer Engin., ¹Georgia Inst. of Technol., Atlanta, GA

Abstract: Fatigue is a highly impairing symptom of major depressive disorder (MDD) frequently reported by patients¹. However, fatigue is often a subjective phenomenon that may stem from multiple components, including peripheral muscle fatigue, energy inefficiency, and

altered effort perception. While energy expenditure (EE) tends to scale predictably with increasing incline, subjective ratings of effort (e.g., Borg RPE) do not always align with objective effort costs. We hypothesized that peripheral muscle fatigue, indexed via median frequency (MDF) of EMG signals, may help explain this discrepancy. To test this, we examined how MDF, EE, and perceived exertion, along with baseline fatigue (Multidimensional Fatigue Inventory [MFI]), interact in a healthy cohort.

Fifteen participants completed a treadmill walking task consisting of six trials at varying inclines (0°, 3°, and 6°). Trials were grouped into two blocks: Block 1 included the first three trials (one at each incline), and Block 2 repeated the same inclines in a different order. EMG signals were recorded from eight muscles (Soleus, Gastrocnemius Medialis, Tibialis Anterior, Vastus Medialis, Rectus Femoris, Biceps Femoris, Gluteus Medius, and Gluteus Maximus) and processed to extract z-scored MDF. EE was measured via indirect calorimetry. Subjective exertion was recorded post-trial using the Borg RPE scale, and MFI scores were collected pre-experiment.

In Block 1, MDF significantly decreased with increasing effort in the Soleus (Friedman $p = .001$), with post-hoc comparisons indicating significant changes between 3°→6° ($p = .003$) and 0°→6° ($p = .002$). Additionally, average MDF in Block 1 correlated with MFI physical fatigue scores ($r = -.82$, $p = .0002$) and Borg RPE ($r = -.57$, $p = .028$). Mediation analysis revealed that MDF significantly mediated the relationship between EE and perceived effort in both Block 1 (indirect effect $ab = 1.225$, $p < .001$) and Block 2 ($ab = 1.226$, $p = .002$).

These results suggest that muscle fatigue, as indexed by MDF, may contribute to perceived effort beyond what is explained by metabolic cost alone. This framework offers a promising approach for disentangling the physiological and perceptual components of fatigue and may be especially informative for understanding psychiatric populations, such as MDD.

References: 1. Gao K, Calabrese JR. Prevalence and factors associated with fatigue in patients with major depressive disorder or bipolar disorder. *J Affect Disord.* 2024 Oct 1;362:493-501. doi: 10.1016/j.jad.2024.07.016. Epub 2024 Jul 14. PMID: 39009311.

Disclosures: C.A. May: None. T. Albarran: None. T. Harvey: None. K. Herrin: None. J. Leestma: None. A. Young: None. G.S. Sawicki: None. C. Rozell: F. Consulting Fees (e.g., advisory boards); CJR is a consultant to Motif Neurotech, Inc.. S. Alagapan: None.

Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

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Title: Hunger accelerates reward learning through a movement specific dopamine signal

Authors: *M. CHEVEE¹, M. E. ALTEMUS¹, S. D. EMERSON¹, J. TAT¹, H. YOON¹, N. CROW¹, C. KIM¹, T. CHALASANI¹, S. ROBERTS¹, S. CAJIGAS GABRIEL¹, M. Z. LEONARD¹, A. KONOMI PILKATI², O. MURFIELD¹, E. S. CALIPARI¹;

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Abstract: Hunger is a need state that drives food seeking and also potentiates learning. We show that food restriction enhances performance only when learning requires actions and those actions are reinforced by appetitive outcomes - both caloric and non-caloric. This is mediated by the potentiation of a dopamine signal at movement offset that integrates information about actions and the spatial location of rewards. Hunger increases dopamine release far from locations with high reward probability to drive future approach - attracting mice toward the goal and increasing opportunities to engage. Hunger-modulated dopamine transients are specific to the dorsomedial striatum and are not a general amplification of reward-related signaling. Rather, they only emerge in contexts where reward locations are known from prior experience. These findings reveal a dopamine-based mechanism through which hunger biases spatially guided approach towards rewards, thereby accelerating reinforcement learning.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Topic: H.02. Reward and Appetitive Learning and Memory

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Title: Optogenetic stimulation of DRD1 cells in the main intercalated nucleus of the amygdala during extinction.

Authors: *N. ARABSHAHI¹, J. TINNING², E. S. KIM²;

¹Psychology, ²George Fox Univ., Newberg, OR

Abstract: Emerging evidence from animal studies highlights the critical role of dopaminergic pathways connecting the prefrontal cortex to the amygdala in the regulation of emotion. The intercalated cell masses (ITCs) of the amygdala, particularly the main intercalated nucleus,

function as a key modulatory hub by integrating excitatory inputs from the prefrontal cortex and relaying inhibitory outputs within the amygdala (Asede et al. 2022). In the present study, we targeted dopamine D1 receptor (DRD1)-expressing neurons within the ITCs using a Cre-dependent AAV-CAG-ChR2-GFP viral vector in D1-Cre transgenic Long-Evans rats. Following viral expression, animals underwent a modified real-time conditioned place preference (CPP) task to assess baseline preference. In the second phase, animals were trained on a variable ratio 5 (VR5) reinforcement schedule, receiving optogenetic stimulation (10 mA, 20 Hz, 1 ms square pulse) contingent upon lever pressing. After establishing a stable baseline across three days of stimulation, extinction sessions were introduced while maintaining stimulation. A final real-time CPP test was conducted post-extinction. Initial stimulation of DRD1 ITC neurons did not elicit a significant real-time place preference, nor were there differences between stimulation and non-stimulation periods at baseline. However, during extinction, stimulation elicited persistent and reversible operant responding that was significantly elevated during stimulation epochs. Post-extinction testing revealed a clear real-time place preference for the previously stimulated side. These findings suggest that DRD1-expressing neurons in the intercalated nucleus of the amygdala contribute to reward learning through associative mechanisms and may play a broader role in modulating reinforcement-driven behavior.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Topic: H.02. Reward and Appetitive Learning and Memory

Support: 2R01 DA047870

Title: Contributions of the basolateral amygdala to anterior cingulate cortex pathway in learning under unexpected uncertainty

Authors: *C. G. AGUIRRE, L. ALHABBAL, J. L. ROMERO, A. IZQUIERDO;
Dept. of Psychology, UCLA, Los Angeles, CA

Abstract: The Anterior Cingulate Cortex (ACC) and Basolateral Amygdala (BLA) have both been implicated in flexible reward learning individually, yet it is not well understood whether their communication is necessary to guide choice behavior in this context. We have previously shown the BLA is recruited under uncertain reward probability conditions, specifically the first probabilistic (90/10) reversal, but not under deterministic (100/0) conditions. However, whether this information is conveyed to the ACC to causally contribute to learning is unknown. Here we examined the contribution of the BLA-to-ACC (BLA-ACC) pathway on a spatially-based flexible learning task with increasingly uncertain reward probabilities using a chemogenetic pathway-specific approach. Male and female Long-Evans rats (N=14) were stereotaxically

prepared with a virus containing inhibitory hM4Di DREADDs on a CaMKII promoter bilaterally in the BLA and bilateral guide cannulae above the ACC in the same surgery. Rats were then tested in a touchscreen-outfitted chamber, on a flexible learning task in which they were required to initiate each trial by nosepoking a center stimulus, make a choice by nosepoking a stimulus on either the left or right side of the screen, and obtain a sucrose pellet on rewarded trials. Within a given session, each rat was tested on one of three possible reward schedules progressing from less to more reward uncertainty (100/0 to 70/30), each consisting of 3 blocks of 75 trials each, with reward contingencies reversing on every block and increasing in uncertainty across blocks. Five minutes prior to each reward schedule rats received a 0.5 uL microinfusion of either clozapine-N-oxide or artificial cerebrospinal fluid solution, using a between-subject design. Generalized Linear Models were conducted to test the effect of drug, schedule, and block, on several outcome variables. We found a significant drug*schedule*block interaction for high probability choice in the first reward schedule, such that BLA-ACC pathway inhibition facilitated learning in blocks 1 (100/0) and 2 (0/100), but impaired learning in block 3 (90/10), compared to non-inhibited control animals. Critically, the 3rd block was the first combination of a probabilistic schedule and reversal, i.e. unexpected uncertainty. Similarly, in the second reward schedule, BLA-ACC inhibition also facilitated learning in blocks 1 (90/10) and 2 (10/90) compared to non-inhibited control animals. These findings reveal a novel function of the BLA-ACC pathway in either facilitating or impairing flexible learning depending on both the level of uncertainty in the environment as well as prior experience with uncertainty.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Program #/Poster #: PSTR292.17/EE1

Topic: H.02. Reward and Appetitive Learning and Memory

Support: University of Vermont Department of Psychological Science

Title: Inactivation of the nucleus reuniens, but not the mediodorsal nucleus of the thalamus, affects context-dependent operant responding

Authors: E. W. HARLAN¹, *J. T. GREEN²;

¹Psychological Sci. and Neurosci. Grad. Program, ²Univ. of Vermont, Burlington, VT

Abstract: Contextual cues, background physical (e.g., olfactory, visual, auditory, and tactile sensations) and internal (e.g., hunger, stress) stimuli, modulate learned behaviors (Bouton, 1993, 2019). For example, rats may learn that an operant response leads to an outcome in one context (Context A), then, in a second context (Context B), the response no longer produces the outcome and the response extinguishes. When the rat is returned to Context A, the extinguished response

returns (ABA renewal) (Bouton & Bolles, 1979; Bouton et al., 2011). We have previously shown that pharmacological inactivation of a region of the medial prefrontal cortex, the prelimbic cortex (PL), selectively affects operant responding in Context A. Many thalamic nuclei are directly connected with the PL and modulate its function. Here, we investigated two thalamic nuclei, the nucleus reuniens (RE) and the mediodorsal nucleus (MD), that have strong reciprocal connections with the PL (Vertes et al., 2015). We predicted that these regions would also be important for Context A operant responding. In the first experiment, we examined the RE. Previous work by others has revealed a role for the RE in extinction of Pavlovian fear conditioning (Ramanathan et al., 2018; Totty et al., 2023). Male Wistar rats underwent daily training sessions in Context A where lever pressing was reinforced, and were exposed to a second, untrained context (Context B). Rats were then tested in Contexts A and B after an intra-RE infusion of either muscimol, a GABA_A receptor agonist, or vehicle. RE inactivation reduced responding in Context A but did not affect responding in Context B. On the following days, rats underwent extinction in Context B and were exposed to Context A (no lever present). After extinction training, rats received a second test in Contexts A and B. During the second test, RE inactivation again reduced responding in Context A without affecting responding in Context B. A second experiment followed an identical procedure, except that guide cannulae were bilaterally implanted into the MD. MD inactivation had no effect on responding in either context. Our results suggest that the RE (but not the MD) may be a part of a larger circuit (including the PL) involved in contextual modulation of operant responding.

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Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR292.18/EE2

Topic: H.04. Emotion

Support: Johns Hopkins University Discovery Award
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Title: Effects of psilocybin on brain representations of and memory for audiovisual narratives

Authors: *B. WINSTON¹, G. LOFLAND³, J. CHEN², F. S. BARRETT⁴;
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Abstract: Background The classic psychedelic drug psilocybin is known to perturb the functional landscape of the human brain during unconstrained perception and cognition. However, the degree to which psilocybin alters processing of and memory for naturalistic, multi-modal stimuli across the cortical hierarchy is not well characterized. Methods 16 participants (of a planned 30) watched nine short audiovisual movies totaling one hour while undergoing functional MRI during the acute effects of 10mg/70kg psilocybin and placebo administered at separate visits. Movies were segmented into events by human raters. To examine how psilocybin influences brain representations of movie content across the cortical hierarchy, we used intersubject pattern correlation (pISC), which quantifies the similarity of neural patterns between individuals, isolating the stimulus-driven signal (Chen et. al, 2017). pISC was calculated and averaged across events and movies in primary auditory and visual cortices, posterior-medial cortex, medial prefrontal cortex and angular gyrus and compared between drug conditions using two-tailed t-tests. To examine how psilocybin during movie watching affected later memory for movies, participants recalled movies in as much detail as possible six hours after watching (by which point drug effects had subsided.) Whisper v3 was used to transcribe recall, and GPT-4o was used to determine which events were remembered. Proportion of events remembered and intersubject correlation (ISC) of *which* events were remembered were calculated for each drug condition. Results pISC was reduced in all ROIs in the psilocybin condition after Bonferroni correction ($p < 0.01$). There was no significant difference in the proportion of events remembered between the placebo (37%) and psilocybin (34%) conditions ($p=0.25$), nor the ISC ($r=0.44$ placebo; $r=0.43$ psilocybin; $p=0.6$). Conclusions pISC was reduced in both sensory and high-order brain regions, suggesting that altered stimulus processing as early as primary sensory cortex could contribute to psilocybin-induced perceptual changes. Memory strength was equal across drug conditions, indicating no impairment in comprehension nor memory encoding of the stimulus under acute effects of psilocybin. Follow-up analyses will investigate variables that could drive changes in pISC, including attentional dynamics, gaze position, large-scale neuromodulatory effects, and the relative influence of internal representations and external stimuli on naturalistic perception.

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Poster

PSTR292: Reward Learning and Primary Rewards

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR292.19/EE3

Topic: I.03. Decision Making

Support: National Research Foundation of Korea (Grant No. RS-2025-00516410)
Seoul National University Creative Pioneering Researchers Program

Title: Investigating the effects of environmental stochasticity on real-time decision-making in a naturalistic driving task using inverse reinforcement learning

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Abstract: Naturalistic tasks provide expansive state-action spaces and incorporate ecologically valid stimuli, allowing researchers to observe decision-making in contexts that closely resemble real-world environments. This approach addresses key limitations of traditional behavioral paradigms, which often rely on simplified stimuli and constrained actions that often fail to capture the complexity of human decision-making. In our previous study, we developed a naturalistic real-time driving task-the (static) highway task-where participants controlled a car to drive as fast as possible without crashing into the other cars to achieve a high score. We used an inverse reinforcement learning (IRL) algorithm as a modeling framework and showed that the inferred reward functions as indicators for impulsivity. In a follow-up study, IRL reward trajectories showed strong correlation with BOLD signals in the reward circuit, indicating that they might indeed reflect value representations in the brain. In the present study, we extend the highway task (the dynamic highway task) by introducing stochastic lane changes by non-player vehicles, increasing environmental stochasticity. This study aims to investigate the effects of environmental stochasticity on real-time driving behavior, and how these differences are reflected in reward trajectories inferred through IRL. A total of 28 participants visited the lab and performed both static and dynamic highway tasks, the order of which was counterbalanced. We focused on two salient types of events during the task: cut-ins and overtaking. Preliminary results revealed that IRL reward trajectories varied across the levels of impulsivity. In cut-in situations, the high impulsivity group showed an early IRL reward peak at lane-change onset, followed by a rapid decline. In contrast, the low impulsivity group showed a delayed peak, occurring at the moment the intruding vehicle entered their lane. This suggests that the individuals with high impulsivity assign reward value more reactively and immediately, whereas those with low impulsivity demonstrate more temporally extended reward evaluation. In overtaking, the low impulsivity group showed greater reward increases prior to passing maneuver. Given that successful overtaking may reduce the likelihood of future cut-ins in a dynamic environment, this may reflect anticipation of such benefits. By introducing environmental stochasticity, this study enabled the investigation of how the inferred reward varies in real-time with impulsivity during uncertain decision-making.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Topic: H.04. Emotion

Support: KAKENHI 20J13905
US Brain Initiative 1U01NS122123-01

Title: A functional input-output organization in the amygdala for emotional learning and memory

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Abstract: Major advances in understanding how neural circuits encode emotion have been made using viral-based optogenetics and calcium imaging, particularly in the lateral and basal nuclei of the amygdala (LA/B) - a hub for integrating sensory, salience, and valence (aversive/rewarding) information to guide adaptive emotional behaviors. Prominent theories conceptualized emotions within a 2-dimensional framework of valence and salience. While LA/B neurons are known to encode these dimensions, it remains unclear whether anatomical connectivity defined LA/B cell types exhibit functional specialization for aversive, rewarding, or salience processing. Here, we employed a new dual-recombinase viral strategy (PINCER) to target LA/B neurons defined by both their inputs from an aversive signaling midbrain region (the cuneiform nucleus, CnF), and outputs to the central amygdala (CeA). We expressed calcium indicators and optogenetic tools in three cell groups: those defined by CnF input alone, CeA output alone, or both. We recorded and manipulated activity during aversive and appetitive Pavlovian conditioning and a salience-dependent latent inhibition task. Calcium imaging revealed that input-alone and output-alone LA/B populations responded to both conditioned stimuli (CS) and unconditioned stimuli (US) in both aversive and appetitive contexts. Optogenetic inhibition during US presentation impaired aversive memory formation. These populations also showed reduced CS responses after repeated unreinforced CS presentations, and disrupting their activity during this phase attenuated latent inhibition, indicating a causal role in salience-based learning. Strikingly, neurons defined by both CnF input and CeA output selectively responded to aversive US during conditioning and developed a response to the sensory CS only at aversive memory retrieval. Optogenetic inhibition of these cells impaired aversive memory without affecting reward or salience learning, suggesting a specific role in encoding aversive value. These results reveal a functional architecture in the amygdala in which LA/B neurons defined by input-alone or output-alone connectivity broadly support salience and valence processing, while subpopulations defined by both input and output are selectively engaged in aversive learning.

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Poster

PSTR292: Reward Learning and Primary Rewards

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Topic: H.04. Emotion

Support: NIDA K01DA053438

Title: Brain connectivity and representations encode positive affect in dynamic experiences

Authors: ***T. SAVALIA**¹, S. MARTIN², S. DEVARAYAPURAM RAMAKRISHNAN⁴, A. O. COHEN³;

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Abstract: Emotions are thought to be represented across the human brain. Experimental research often assumes that different types of aversive and appetitive experiences evoke similar affective states and often uses static reinforcers that bear little resemblance to real-world experiences of affect. Recent work used predictive modeling to show that different types of aversive experiences are encoded via both modality-specific and modality-general neural representations (Ceko et al. (2022)). It is not known whether dynamic appetitive experiences are represented in similar distributed signatures that can be used to predict positive affect. In an fMRI scanner, 52 adult participants aged 18-25 completed a personalized appetitive experiences task. Participants were presented with 4 types of dynamic appetitive stimuli selected because they are encountered in everyday life and can be readily used in a variety of settings: monetary rewards, social media clips from preferred categories, music clips from preferred genres, and positive autobiographical memories. Participants provided a subjective rating about each experience, indicating how much they wanted to have that experience again. We find that winning money and recalling past positive autobiographical memories are rated as more positive experiences than participant-preferred social media and music clips. We employ a predictive modeling pipeline including univariate feature selection and partial least squares regression to assess brain activation patterns and functional connectivity encoding participants' subjective experiences. We find that, unlike aversive experiences, multivariate representations are not sufficient to decode dynamic appetitive experiences. In order to capture positive affect across our dynamic stimuli, we show the need to incorporate task-based functional connectivity in predictive models to successfully decode subjective affect. Finally, using a permutation importance approach, we identify multivariate representational features across the brain, including the medial frontal regions, the inferior temporal gyri, the precuneus, and the insula, along with widespread connectivity, including from the Cingulate gyrus, OFC, and the temporal cortex that capture positive affect across different modalities. We also identify features that capture positive affect specific to each modality. Our work signifies that positive affect in dynamic experiences is encoded in both distributed multivariate patterns and putative communication between regions across the brain.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR293.01/EE6

Topic: H.03. Motivation

Support: DA007359

Title: Modulating levels of Δ FOSB alters nucleus accumbens medium spiny neuron activity

Authors: *T. MARKOVIC¹, L. HOLT², A. M. MINIER-TORIBIO³, E. M. PARISE⁵, A. GODINO³, V. KONDEV⁴, Y. YIM³, E. J. NESTLER⁶;

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Abstract: FOSB is a key transcription factor that mediates gene expression changes in the nucleus accumbens (NAc) and other brain regions in response to chronic stimuli. The NAc is composed of GABAergic medium spiny neurons (MSNs) that express either dopamine receptor 1 (D1) or D2. Previous work in rodents showed that cocaine induces Δ FOSB in D1 MSNs, chronic stress induces the protein in D2 MSNs in stress-susceptible but in D1 MSNs in stress-resilient animals, while opioids and natural rewards induce Δ FOSB in both. This cell-type-specific regulation of Δ FOSB expression in NAc correlates with its different effects on synaptic properties of MSNs: Δ FOSB decreases excitatory synaptic strength and increases silent synapses onto D1 MSNs, with opposite effects seen for D2 MSNs. However, no studies have investigated how changes in Δ FOSB expression in NAc alter the in vivo activity of MSNs. To address this, we injected D1-Cre and D2-Cre mice with Cre-dependent AAVs that express a calcium sensor and epigenome-editing tools to either induce or repress endogenous Δ FOSB in NAc. We recorded in vivo activity of D1 and D2 MSNs using fiber photometry in response to social reward, saccharin reward, foot shock, and drug rewards. We found that manipulation of Δ FOSB primarily altered MSN responses to salient stimuli such as foot shock and cocaine conditioned place preference (CPP). In fact, decreasing Δ FOSB in D1 MSNs attenuated foot shock-induced calcium transients, while decreasing Δ FOSB in D2 MSNs enhanced them. Similarly, in a cell-specific manner, decreasing Δ FOSB in D1 MSNs and increasing Δ FOSB in D2 MSNs decreases social interaction. Decreasing Δ FOSB in D1 MSNs - but not D2 MSNs - blocks cocaine CPP and attenuates neuronal activity aligned with entrance to the cocaine-paired side. These findings of opposite in vivo modulation of D1 vs. D2 MSN activity in vivo by Δ FOSB demonstrate how Δ FOSB influences circuit activity and shed light on cell-autonomous mechanisms controlling behavioral responses. Ongoing electrophysiology experiments are assessing how changing endogenous Δ FOSB levels affects baseline excitability of D1 and D2 MSNs. Lastly, to assess the translational potential of Δ FOSB, we utilized novel lipid nanoparticle (LNP) gene therapy technology to systemically deliver Δ Fosb mRNA during cocaine CPP. Using this approach, we

found that LNP-driven overexpression of Δ FOSB augmented drug seeking behavior of normally non-reinforcing doses of cocaine. Taken together these findings demonstrate Δ FOSB's bidirectional modulation of behavioral responses to rewarding and aversive stimuli and its potential for clinical translation.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Topic: H.03. Motivation

Support: NIH Grant K99 DA056574
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Title: Nucleus accumbens shell *Pdyn* neurons modulate dopamine release to control prosocial behavior

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Abstract: The nucleus accumbens (NAc) integrates circuits involved in motivation and reward, where dopamine (DA) release is known to motivate appetitive behaviors. DA modulates D1 or D2 receptors expressed by NAc medium spiny neurons, a process thought to control appetitive behaviors. However, emerging evidence points to a more complex relationship: not only are these neurons influenced by DA, they may also regulate DA release themselves. Characterizing this bidirectional relationship is a critical step in understanding how mesolimbic circuits control motivated behavior and mediate the development of motivational disorders, such as addiction and depression. To determine whether NAc spiny neurons modulate DA release to control behavior, we focused on D1 neurons in the medial shell. Analysis of single-cell RNA seq data shows that the majority of NAc D1 neurons express the neuropeptide dynorphin (*Pdyn*), consistent with previous studies, and tracing from NAc *Pdyn* neurons shows a dense projection to the ventral tegmental area (VTA). Stimulation of NAc *Pdyn* neurons with red-shifted ChRmine in *Pdyn*-Cre mice triggered large increases in DA release locally in the NAc medial shell and this stimulation was reinforcing in a real time place preference test. On the other hand, stimulation of VTA DA inputs in the NAc in *Pdyn*-Cre:DAT-Flp mice had an inconsistent effect on *Pdyn* neuronal activity. We next prevented transmitter release from NAc *Pdyn* neurons by expressing Cre-dependent short hairpin RNAs (shRNAs) targeting *Vgat* or *Pdyn*, both of which

significantly blunted ChRmine-evoked DA release. To assess whether this treatment affected DA release triggered by naturally rewarding stimuli, we recorded DA release in response to social interaction. Mice expressing either shRNA displayed a significantly blunted increase in DA during social interaction, which was paralleled by a reduction in social behavior. We next simultaneously recorded DA release and NAc *Pdyn* neuron activity during social interactions and found that during interaction bouts, both DA release and NAc *Pdyn* activity increased, with *Pdyn* activity consistently preceding DA release. Cross-correlation revealed a lag time of ~100 msec. hM4Di-mediated inhibition of NAc *Pdyn* neurons blunted socially-tuned DA release and reduced interaction time. Together, these data suggest that NAc *Pdyn* neurons release both GABA and dynorphin to modulate DA release and promote social behavior. Future studies are investigating the functional anatomy underlying NAc *Pdyn* neuron control of DA release.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: H.03. Motivation

Support: NSF Grant 2135305

Title: Dopamine's role in modulating exploratory activity in *Drosophila melanogaster*

Authors: *T. SAQUIB¹, G. W. ROMAN²;

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Abstract: Exploratory behaviors are driven by neophilia in many species. In neophilia, novel stimuli arouse the animals' attention, which drives exploratory behaviors. This response to novelty is susceptible to changes in the animal's behavioral state, e.g., environmental stressors or hunger can lead to changes in the behavioral response to novelty. While the central reward system is known to modify novelty-seeking behavior, the underlying neural mechanisms remain unclear. In *Drosophila melanogaster*, dopamine signaling regulates arousal, attention, and learning. We hypothesized that dopamine also modulates behavioral responsiveness to novelty during exploratory activity. Using an open-field arena paradigm, we employed genetic and pharmacological approaches to dissect the dopaminergic contributions to exploration. Our findings demonstrated that the *Dop1R1* receptor function is critical for regulating exploration. *Dop1R1*^{f02676} mutants and flies with pan-neuronal *Dop1R1*-RNAi expression showed increased exploratory activity. Pharmacologically increasing brain dopamine levels via Levodopa and Carbidopa administration suppressed exploration in wild-type flies. This inhibitory effect of

increased dopamine was reduced in *Dop1R1*^{J02676} mutants, indicating dopamine's suppression of exploration requires, in part, *Dop1R1* activity. We further examined the *Dop1R2* receptor activity and found that, in contrast to *Dop1R1*, *Dop1R2* has an activating role in exploratory behavior. *Dop1R2*^{MB05108} mutants and pan-neuronal inhibition of *Dop1R2* resulted in significantly reduced exploratory behavior. This suggests a bimodal dopaminergic regulation of exploration, with *Dop1R1* mediating inhibition and *Dop1R2* mediating activation of exploratory activity. We have further examined the role of environmental stress in exploration. Confinement stress significantly suppresses exploratory activity compared to normal-housed controls. *Dop1R1*^{J02676} mutants exhibited significantly reduced sensitivity to confinement stress. This suggests that the stress-induced suppression of exploration is partially mediated by dopamine signaling through *Dop1R1*. Together, these findings suggest a complex, receptor-specific modulation of novelty-driven exploration in *Drosophila*, shaped by both intrinsic dopaminergic signaling and extrinsic stress factors.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Title: The encoding of interoceptive-based predictions by the paraventricular nucleus of the thalamus D2+ neurons

Authors: *B. MACHEN¹, S. MILLER¹, C. LAMPERT¹, L. ASSAF², J. TUCKER³, S. BEAS¹;
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Abstract: Interoceptive predictions are internal models the brain uses to anticipate bodily states based on past experiences and environmental cues. Under normal conditions, these predictions help maintain homeostasis by guiding behavior in anticipation of physiological needs. However, they can become maladaptive, with certain cues triggering exaggerated physiological responses (e.g., fear cues inducing stress). While progress has been made in identifying the neural substrates of interoceptive predictions, the specific circuits involved remain unclear. The paraventricular nucleus of the thalamus (PVT) receives input from regions processing both interoceptive and exteroceptive signals, positioning it to integrate these domains and influence

behavior. The PVT is functionally diverse, and our previous work (Beas et al., 2024) showed that this heterogeneity has behavioral relevance. Specifically, we found that a subset of PVT neurons expressing dopamine receptor 2 (PVTD2+) are sensitive to physiological state changes, suggesting a role in encoding interoceptive predictions. In this study, we used fiber photometry to monitor PVTD2+ activity in mice performing a linear maze task for food rewards. Mice initiated trials by entering a trigger zone, then navigated a linear track to a reward zone following a cue. We found that PVTD2+ photometric responses increased during reward approach and delivery, with higher responses when mice were hungrier or when non-caloric rewards were provided. These neurons also responded more strongly when the reward matched the physiological need (e.g., water when thirsty vs. water when hungry). The responses of these neurons were reduced when cues were absent, indicating sensitivity to reward expectation. Interestingly, PVTD2+ signal decreased during return trips to the trigger zone, and these responses were still modulated by state and outcome, further showing modulation by expectation. We also found that these approach and return responses were learned over time, and chronic inhibition of PVTD2+ neurons impaired task performance. Our findings strongly suggest that PVTD2+ neurons play a critical role in computing interoceptive predictions by integrating physiological signals, external cues, and prior experiences to optimize goal-directed behavior.

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Poster

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Topic: H.03. Motivation

Support: State of Washington

Title: A simplified rat environmental enrichment procedure to combine with detection of dopamine using matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS)

Authors: ***J. W. GRIMM**, A. O'SHEA, M. GARCIA, T. TRICKETT, E. VAN DEN DUNGEN, A. VON HAGEL, B. OLIVER, A. CORTEZ-TINAJERO, R. BARCOTT, S. HOHNDEL;
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Abstract: We developed a simplified environmental enrichment (EE) procedure to assess the effects of EE on motivation for sucrose by adult rats. Briefly, rats reared on auto-watering were first acclimatized to water bottles for two days. Rats were then placed into a 24h test condition assigned as a cross between being provided with water or 10% sucrose and remaining in the home cage or being placed in EE. Male and female rats in EE consumed significantly less

sucrose, but not less water, than rats in control housing. To better understand the relationship between brain dopamine and the ability of EE to reduce motivation for sucrose, our goal is to examine dopamine levels across several brain regions from subjects exposed to EE and/or sucrose using matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS). The MALDI method under development utilizes 10 micrometer slices of a flash-frozen whole brain. Several slices are placed on an indium tin oxide coated slide with reference spots of dopamine and dopamine added to brain tissue homogenate. A-cyano-4-hydroxycinnamic acid (HCCA) containing matrix solution is then sprayed onto the slide. A slide is imaged using a Bruker Autoflex Max MALDI-TOF MS apparatus. Laser shots (5 micrometer spatial resolution) across each slice produce spectra representing several dozen molecules. We have identified in various brain regions peaks that correspond to dopamine + H (154.4 +/- .2 m/z), dopamine + Na (176 +/- .5 m/z), and dopamine + K (192 +/- .5 m/z). Next steps include correlating dopamine signal in motivation-relevant brain regions (e.g., nucleus accumbens) with sucrose intake during EE.

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PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Topic: H.02. Reward and Appetitive Learning and Memory

Support: HHMI Hannah Gray Fellowship

Title: Spatiotemporal DA Coordination as a Target of Antipsychotic Action

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Abstract: Dopaminergic dysfunction in the dorsal striatum is a hallmark of psychosis spectrum disorders, including schizophrenia. While hyperdopaminergia in the associative caudate is a well-replicated biomarker, its consequences on the spatiotemporal organization of dopamine (DA) dynamics and the implications for disrupted cognition—remain poorly understood. Our group recently discovered a fundamental organizing principle of DA activity in the dorsal striatum: structured, wave-like dynamics that precisely coordinate task-related changes across caudate-putamen. These DA waves are sensitive to task demands that require state inference and action-outcome learning, implicating them in cognitive processes often impaired in psychosis. Here, we show that pharmacologically induced hyperdopaminergia disrupts the spatial and temporal precision of DA wave motifs, degrading region-specific DA coordination. Using

high-resolution optical imaging of genetically encoded DA sensors across dorsal striatum, we quantify how DA wave features—such as propagation direction, velocity, and opto stimulation-evoked dynamics—are altered under psychotomimetic conditions. We further test whether various antipsychotic medications (including Haloperidol, Clozapine, Xanomeline) restores normal DA dynamics that are essential for behavioral flexibility. Ultimately, our findings suggest that DA wave coordination is a vulnerable substrate for cognitive dysfunction in psychosis and a functional readout for evaluating the efficacy of antipsychotic interventions. This framework shifts focus from bulk DA levels in restricted striatal domains towards studying the complexity of spatiotemporal organization of DA signals in the striatum, offering a novel window into the circuit-level mechanisms through which novel antipsychotic medications alleviate classes of cognitive symptoms.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Title: Not all dopamine neurons are created equal: Exploring the differences between dopamine and glutamate-dopamine co-transmitting VTA neurons

Authors: *E. D. PRÉVOST¹, L. A. WARD², D. ALAS¹, G. A. AIMALE¹, S. IKENBERRY¹, J. PELLETIER¹, K. PRICE³, A. M. POLTER³, D. H. ROOT¹;

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Abstract: For the last half-century, dopaminergic neurons in the ventral tegmental area (VTA) have been of great interest for their roles in addiction, motivation, learning, and psychiatric disorders. However, a subset of VTA dopamine neurons is also capable of releasing glutamate. It is unclear how this molecular heterogeneity contributes to the wide range of functions ascribed to VTA dopamine neurons. Moreover, the individual contributions of glutamate release and dopamine release by glutamate-dopamine co-transmitting neurons on learning to attain rewards or avoid aversive stimuli are unknown. Further, because VTA glutamate neurons can co-transmit glutamate with dopamine or co-transmit glutamate with GABA, it has been challenging to untangle glutamate and dopamine function of VTA neurons, especially the unknown functions of

neurons that release *only* glutamate. Here, we parsed glutamate-dopamine neurons from dopamine-only and glutamate-only neurons, as well as identified the roles of glutamate and dopamine release from co-transmitting neurons toward reward- and aversion-motivated behaviors. Our strategy involved combining intersectional and subtractive genetics with ex vivo whole-cell recordings, in vivo calcium recordings, optogenetics, in vivo dopamine sensors, and cell-type specific and neurotransmitter-specific knockdown. We found that these subpopulations exhibit dissociable—and sometimes opposite—electrical properties and motivation-related activity patterns, and that dopaminergic subpopulations have similar but not identical accumbal dopamine release dynamics. Recording all dopamine neurons without considering glutamate co-transmission showed mixed results that obscured subpopulation function, complicating the interpretation of decades of studies that have assessed all dopamine neurons together. Additionally, we found that dopamine and glutamate released from co-transmitting neurons contribute to independent components of motivated behavior. We found that dopamine was necessary for associative learning in aversive contexts, while glutamate was necessary for appropriate locomotor responding in rewarding, aversive, or neutral contexts. Our results are in accordance with prior investigations of these neurons and provide additional context and novel insights into the specific behaviors they regulate. Taken together, these findings demonstrate that molecular heterogeneity in dopaminergic subtypes can partially explain the abundance of hypotheses surrounding VTA dopamine neurons.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Program #/Poster #: PSTR293.08/FF5

Topic: H.08. Drugs of Abuse and Addiction

Support: R01AA021505
R01AA027768
U01AA025932

Title: Direct pathway medium spiny neuron-induced acetylcholine pause-rebound encodes action-outcome contingency

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Abstract: Instrumental learning enables adaptive behavior by integrating sensory, motor, and reward-related information, a process in which the striatum plays a central role. However, the circuit-level interactions between distinct striatal neuronal populations during contingency

learning remain poorly understood. Here, we employed genetically encoded sensors, fiber photometry, ex vivo electrophysiology, and in vivo optogenetics to investigate acetylcholine (ACh) dynamics in the dorsal striatum during instrumental learning in mice. We observed a distinct ACh dip-rebound pattern that emerged in parallel with increased dopamine (DA) release, elevated activity of direct pathway medium spiny neurons (dMSNs), and suppressed activity of indirect pathway MSNs (iMSNs) once action-outcome (A-O) contingency was established. Using decoupled contingency schedules, we found that while DA release tracked reward delivery, the dMSN activation and ACh dip-rebound occurred independently of reward, instead aligning with whether actions reliably predicted outcomes. This suggests that ACh and DA serve dissociable roles in encoding contingency versus reward. To test causality, we optogenetically activated dMSNs in vivo and ex vivo, which elicited pause-rebound firing in cholinergic interneurons (CINs), confirmed by both electrophysiology and photometry. Conversely, artificially disrupting the ACh dip via CIN activation accelerated extinction learning, supporting a functional role for ACh rebound in sustaining learned A-O associations. These findings establish dMSN-driven ACh pause-rebound dynamics as a circuit-level correlate of contingency, revealing a previously unrecognized striatal mechanism critical for flexible goal-directed behavior.

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Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

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Program #/Poster #: PSTR293.09/FF6

Topic: H.03. Motivation

Title: Associative mechanisms of midbrain acetylcholine release

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Abstract: Dopamine release arising from midbrain neurons in the ventral tegmental area is thought to underlie the attribution of motivational value to cues paired with rewarding and aversive outcomes. The regulation of dopamine neuron activity, thus, is paramount to associating and regulating behavioral responses to reward- and fear-paired stimuli. The activity of VTA dopamine neurons is tightly modulated with limited sources of direct excitation onto these neurons. However, acetylcholine release arising from the brainstem cholinergic neurons in groups C5 and C6 provide a direct excitatory input to VTA dopamine neurons. It has been established that these inputs generally control rudimentary features of reward, yet we fail to understand the psychological consequences of midbrain acetylcholine release. To better understand the function of midbrain acetylcholine release we implanted rats with cannula

targeting the VTA and assessed acetylcholine signaling in Pavlovian fear and reward conditioning. In fear conditioning, Long-Evans rats (n=32, 16 male 16 female; 8 per group) were infused with 300 nL of saline, mecamylamine (a nicotinic receptor antagonist), scopolamine (a muscarinic receptor antagonist), or physostigmine (an inhibitor of acetylcholine clearance, which locally elevates acetylcholine levels) into the VTA prior to fear conditioning. For reward conditioning (n=24 rats, 12 male, 12 female) we assessed 1) consumption of different rewarding concentrations of sucrose 2) Pavlovian conditioned responding, 3) contextual renewal, and 4) sensory-specific satiety following intra-VTA saline, mecamylamine, scopolamine, and physostigmine. Preliminary data suggest that antagonism of midbrain acetylcholine signaling blunts the acquisition of fear conditioning and suppresses responding to reward-paired cues. Increasing local VTA acetylcholine release had divergent effects depending on fear versus reward conditioning. Ongoing experiments are making use of fiber photometry to record intra-VTA acetylcholine release, as well as combining photometry and pharmacology to understand the net effects of VTA acetylcholine release on dopamine output in the striatum. Collectively we aim to provide a comprehensive account of the function of midbrain acetylcholine release in the acquisition, expression, and modification of cue-generated expectations.

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Poster

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Topic: H.03. Motivation

Support: Agence Nationale de la Recherche
Fondation de France
ED CSV
France Parkinson

Title: miR-10a-5p Modulates Impulsive Choice in Rats: Insights into the Role of PI3K and the AKT/MTOR Pathway

Authors: *Y. V. LAGES, T. DUFOURD, M. ROUX, M. BARTOLOMUCCI, S. CARNICELLA;
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Abstract: Impulsivity is a core symptom of various neuropsychiatric disorders, including Attention-Deficit Hyperactivity Disorder (ADHD) and impulse control disorders (ICDs), also called behavioral addictions. ICDs are exacerbated by dopamine replacement therapies in disorders such as Parkinson's disease or hyperprolactinemia, and are associated with a high impulsive choice—characterized by a preference for smaller, immediate rewards over larger,

delayed ones. In this study, we applied the Delay Discounting Task (DDTs), a well-established method for measuring impulsive choice in humans and rodents, to investigate the role of miR-10a-5p in the development of cognitive impulsive behaviors. MicroRNAs (miRNAs) are small, non-coding RNA molecules that regulate gene expression primarily through post-transcriptional mechanisms, influencing the stability and translation of target messenger RNAs (mRNAs). Results obtained from NGS sequencing of miRNA-enriched fractions show that under sub chronic administration of pramipexole, a drug used for the treatment of human PD, the expression of miR-10a-5p is increased in the dorsal medial striatum and the nucleus accumbens (NAc) of highly impulsive rats in DDT. To investigate whether miR-10a-5p overexpression would increase impulsivity, we induced its overexpression in the NAc of low- and moderate-impulsive rats using viral infection. Our results show that increased miR-10a-5p expression, as measured by RT-qPCR, led to a heightened preference for larger rewards, particularly in the moderate-impulsive group, indicating increased impulsive behavior. Additionally, we have analyzed whether the expression of one of the miR-10a-5p's predicted target, PI3K, was altered as well as the expression of mRNAs for proteins of the AKT/MTOR pathway, in which PI3K is a key component. The results revealed reduced PI3K mRNA expression in the NAc, but no changes in the expression of AKT1, MTOR, or PTEN mRNAs. Nonetheless, Western-blot results indicate that the expression and phosphorylation of proteins of the AKT/MTOR pathway are altered. In summary, our findings suggest that miR-10a-5p directly promotes impulsive behaviors, potentially through its interaction with PI3K. Future studies will investigate other molecular targets, as well as the role of miR-10a-5p in compulsive behaviors.

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Poster

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Topic: H.03. Motivation

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Title: Paraventricular Thalamic Neuronal Ensembles Activated by Early-Life Adversity Shape Sex-Dependent Alterations in Adult Reward Behavior

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Abstract: Rationale

Early-life adversity (ELA) increases vulnerability to psychiatric disorders such as major depressive disorder and substance use disorders, both of which are characterized by disrupted reward behavior. While the association between adversity in childhood and later life mental health outcomes has been studied extensively, the underlying neurobiological mechanisms remain unclear.

Methods & Results

In a mouse model of ELA (limited bedding and nesting), we observe persistent, sex-specific alterations in reward-related behavior: males exhibit anhedonia, whereas females show increased reward-seeking and consumption. To identify brain regions encoding adverse early-life experiences, we used TRAP2 mice to permanently tag neurons engaged during either ELA or standard rearing. This approach revealed selective, robust, sex-specific activation of the paraventricular thalamic nucleus (PVT) during ELA. In adulthood, these same early-life-engaged (TRAP) PVT neurons become reactivated during reward behaviors. Region-specific chemogenetic manipulation demonstrates that TRAP neurons in the anterior PVT contribute to ELA-induced anhedonia in males, whereas TRAP neurons in the posterior PVT promote heightened reward behavior in females.

Conclusions

ELA engages the PVT in a sex- and region-specific manner. PVT neurons that are active during early-life contribute to reward behavior in adulthood. These findings suggest that 1) there are underlying sex differences in the activity of the PVT neurons during the early-life epoch affected by ELA, and 2) PVT neurons activated by early adversity encode long-lasting, sex-specific vulnerability to reward disruptions.

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Poster

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Topic: H.03. Motivation

Support: Jim and Betty Ann Rodgers' Chair Fund
The Florida Department of Health James and Esther King Biomedical Research Program

Title: Long-term consequences of early-life nicotine exposure on the mesolimbic system

Authors: *M. X. TRUPIANO, D. M. MC CARTHY, D. L. GRAHAM, C. VIED, P. G. BHIDE;
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Abstract: Smoking during pregnancy is a major public health concern due to its potential adverse effects on both the mother and her offspring. Beyond the immediate perinatal risks, there is growing recognition of the long-term neurodevelopmental consequences of prenatal nicotine exposure. These effects are predominantly neuropsychiatric, including alterations in motivation and emotional regulation, which may increase susceptibility to substance use disorders later in life. However, it remains unclear whether there is an association between early-life exposure to nicotine and dysregulated motivated behavior in children and if so whether changes in neurotransmitter signaling mechanisms within the mesolimbic pathways may underlie the behavioral changes. We found robust changes in motivated behaviors (nest building, sucrose preference, and novelty suppressed feeding) in early-life nicotine exposed mice. RNA sequencing of the ventral striatum followed by KEGG biological pathway analysis of differentially expressed genes showed significant enrichment of addiction pathways and associated neurotransmitter systems. Further, immunohistochemical analysis showed that early-life nicotine exposure produced significant deficits in striatal parvalbumin neuron densities. Taken together, our data show that early-life nicotine exposure produces significant changes in multiple measures of motivated behaviors and expression of genes associated with reward mechanisms in the mesolimbic system.

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Poster

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Program #/Poster #: PSTR293.13/FF10

Topic: H.03. Motivation

Title: Plastic changes in dendritic spines induced by distinct copulatory patterns in the medial prefrontal cortex and nucleus accumbens of male rats

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Abstract: Sexual experience of three mating sessions to ejaculation enhances copulatory performance in male rats. This improvement is associated with plastic changes in the dendritic

spines of neurons in the medial prefrontal cortex (mPFC) and nucleus accumbens (NAc), brain regions involved in regulating and expressing sexual behavior. It has been proposed that changes in the density and morphology of dendritic spines within these regions are critical for the consolidation of the association between stimuli and sexual reward (ejaculation). Moreover, it is suggested that this neuroplasticity is driven by the rewarding properties of ejaculation, which represents the most rewarding event within the copulatory behavior. Nevertheless, other components of copulation, such as mounting and intromission, also possess rewarding properties, although to a lesser extent than ejaculation. Although the ejaculatory sexual experience has previously been linked to dendritic spine density in the brain, the flexibility of these spines concerning the distinct copulatory patterns remains unknown. Hence, this study aims to determine the effect of different copulatory patterns on the density of dendritic spines in the mPFC and NAc of male rats after the third mating sexual test. Wistar adult male rats were divided into four groups, each undergoing three behavioral sessions: (1) a group exposed to a sexually receptive female without direct physical contact ($n = 8$), (2) a group that only performed mounts ($n = 8$), (3) a group that copulated until achieving six intromissions ($n = 8$), and (4) a group that copulated to ejaculation ($n = 8$). The brain tissue was then extracted and fixed for analysis. Applying a modified Golgi method, the dendritic spine density in the mPFC and NAc were quantified and categorized. Rats in the ejaculation group showed greater copulatory efficiency. Additionally, this group exhibited a lower proportion of thin spines and a higher proportion of mushroom spines in both structures compared to the other groups. These results show that ejaculation is the copulatory pattern that induces major plastic changes in dendritic spines in mPFC and NAc, which is associated with greater copulatory efficiency. The differences in the proportion of mushroom spines between groups that only had mounts, mounts more intromissions, or the three copulatory responses could indicate a) the degree of sexual reward achieved with each copulatory pattern or b) a process of major consolidation of the relation stimulus-reward sexual.

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Poster

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Topic: H.03. Motivation

Support: Howard Hughes Medical Institute
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Title: Neural Mechanisms of Hedonic Eating

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Abstract: Hedonic eating is defined as food consumption driven by palatability without physiological need. However, neural control of palatable food intake is poorly understood. We discovered that hedonic eating is controlled by a neural pathway from the peri-locus ceruleus to the ventral tegmental area (VTA). Using photometry-calibrated optogenetics, we found that VTA dopamine (VTA DA) neurons encode palatability to bidirectionally regulate hedonic food consumption. VTA DA neuron responsiveness was suppressed during food consumption by semaglutide, a glucagon-like peptide receptor 1 (GLP-1R) agonist used as an anti-obesity drug. Mice recovered palatable food appetite and VTA DA neuron activity during repeated semaglutide treatment, which was reversed by consumption-triggered VTA DA neuron inhibition. Thus, hedonic food intake activates VTA DA neurons, which sustain further consumption, a mechanism that opposes appetite reduction by semaglutide.<!--EndFragment-->

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Poster

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Topic: H.03. Motivation

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Title: Meal Initiation is a Stochastic Process Regulated by Motivational State

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Abstract: Motivation transforms need into action. Hunger, for example, links energy deficit to the search for and consumption of food. However, the neural substrates linking drive state to behavioral initiation are poorly understood. Using eating as a prototypical motivated behavior, we begin to describe where and how this transformation takes place. We find that a set of brainstem GABA cells (expressing *Vgat*, referred to as DR^{VGAT}), link hunger drive to the acute decision to eat. DR^{VGAT} neurons are anatomically well-positioned to initiate feeding, integrating diverse lines of information to encode and control the rapid onset of a caloric meal. These neurons induce the effortful search for food and promote feeding through negative reinforcement. DR^{VGAT} then tune the animal's likelihood of initiating a meal through two simple parameters, intensity and duration of action. Consistent with their specific role in meal initiation,

DR^{VGAT} act independently of potent anorectic cues, such as GLP1R signaling and naturalistic satiety, and they communicate with motor and exocrine systems critical to starting a meal. Together, this work identifies important principles linking energy balance to the acute decision to feed and introduces a probabilistic framework to better understand the initiation of diverse motivated behaviors.

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Poster

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Topic: H.03. Motivation

Support: Project FONDECYT #1190833 (P.R.M.)
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Title: Behavioral and proteomic characterization of a mouse model of Binge eating disorder: Intermittent palatable food exposure enhances compulsive-like behavior

Authors: *P. A. ACEVEDO¹, P. R. MOYA²;

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Abstract: Background: Binge eating disorder (BED) is a condition characterized by recurrent episodes of unusually large amounts of food consumption. The global prevalence of BED increased after the COVID-19 pandemic. This increase has been linked to a rise in anxious behaviors associated with food consumption and the growing availability of palatable foods. A significant proportion of the population remains undiagnosed, a situation that, in combination with the ineffectiveness of current pharmacological therapies, results in considerable deterioration in their quality of life. Research on the neurobiology of BED remains limited, underscoring the necessity for further investigation. The objective is to explore potential altered molecular pathways in the hypothalamus of a mouse model that could be implicated in the predisposition to BED development.

Methods: Female C57BL6J Wildtype mice (Day 30-40) were used, which were subdivided into 3 groups: Control Group without exposure (Control S/E), Control group with continuous exposure to appetizing food (Control E/ C) and Group of intermittent exposure to appetizing food (Binge). After 30 days of exposure to palatable food for 2 hours every other day, behavioral tests associated with anxiety and compulsivity (marble burying, plus maze test, light-dark test, grooming and t maze) were carried out. Whole hypothalamus were obtained from Control E/C and Binge animals and to explore molecular alteration using proteomics analysis. Samples were processed using liquid chromatography-tandem mass spectrometry coupled with an Evosep Biosystems system, and protein identification was performed using MSFragger v4.1. Statistical analysis was conducted in GraphPad Prism 9.

Results: Palatable food consumption during the 2-hour access period was higher in the Binge group vs Control E/C ($p < 0,0001$). In the behavioral tests, significant differences were observed: Increased time spent in the center ($p < 0,05$ -elevated plus maze), reduced time in the light zone ($p < 0,05$ -light-dark box test), longer average decision time to choose an arm ($p < 0,05$ - T-maze), and increased burying ($p < 0,01$ -marble burying test) were found in Binge group vs Control E/C and S/E. The proteomic study identified 5939 proteins, of which 173 proteins with significant differences ($p < 0,05$) between the Control E/C and Binge groups, which are associated with GABAergic pathways and processes such as inflammation and metabolism, among others. Conclusion: Intermittent exposure to palatable food increases anxiety-like and compulsive-like behaviors. Proteomic analysis suggest specific alterations in the hypothalamus, underscoring the need for further research.

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Poster

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Title: A striosomal accumbens pathway drives compulsive seeking behaviors through an aversive *Esr1*+ hypothalamic-habenula circuit.

Authors: ***T. CONTESSE**¹, M. GRAZIANO², C. FORASTIERI², A. CONTESTABILE², F. JUNG², I. NIKOLAKOPOULOU², E. RUBINO³, X. CAO², V. SKARA², I. MANTAS², S. GIATRELLIS⁴, M. CARLEN², D. CALVIGIONI², K. MELETIS²;
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Abstract: The lateral hypothalamic area (LHA) integrates external stimuli with internal states to drive the choice between competing innate or value-driven motivated behaviors. Projections from the LHA to the lateral habenula (LHb) shape internal states, with excitatory estrogen receptor 1-expressing (*Esr1*+) LHA-LHb neurons driving aversive responses and sustained negative states. Here, we identify and functionally characterize a specific projection from the nucleus accumbens (ACB) that targets *Esr1*+ LHA-LHb neurons. Using cell-type-specific

tracing of monosynaptic inputs, single-nucleus RNA sequencing, and neuroanatomical mapping, we demonstrate that the Esr1+ LHA-LHb pathway receives a major input from a striosomal Tac1+/Tshz1+/Oprm1+ ACB neuron subtype. Intersectional cell-type-specific and input-output defined optogenetic manipulation of this ACB-LHA-LHb pathway revealed its role in signaling aversion after repeated activation, with the negative behavioral state being dependent on recruitment of Esr1+ LHA-LHb neurons. Importantly, we found that either activation of the D1+ ACB-LHA projection or inhibition of LHA-LHb neurons defined by ACB inputs can drive reward-independent compulsive-like seeking behaviors that generalize across contexts. We found that these complex yet stereotyped behaviors compete with highly motivated states and can override the drive for natural rewards or social interactions. Our findings reveal a discrete Tac1+ striosomal ACB projection targeting the aversive Esr1+ LHA-LHb pathway as a key circuit that promotes compulsive seeking behaviors over goal-directed actions.

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Poster

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Program #/Poster #: PSTR293.18/GG3

Topic: I.08. Learning and Memory

Title: Do conditioned stimuli and the rewards they predict activate the same internal representation?

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Abstract: If an auditory conditioned stimulus (CS) has repeatedly been paired with a food reward, a rat is said to “predict” the arrival of food in the presence of the CS. But does that CS activate the same neurocognitive representation as the food (unconditioned stimulus, US) itself? If the neurons activated by the food are the same as those activated by the cue, then the cue should be able to replace or substitute for the food itself. We are investigating this question using an operant biconditional task in which rats learned to choose one response lever (R1) after non-contingent presentation of a pellet outcome (O1) but a different lever (R2) after sucrose (O2). In different groups each correct response was rewarded with the opposite outcome type (i.e., O1: R1-O2 vs R2-, O2: R1- vs R2-O1) either immediately or after a 5s delay. After rats learned this task, they were given differential Pavlovian training with two 2-min auditory cues, a 1500 Hz tone (CS1) and a white noise (CS2) each paired with one of the outcomes (CS1-O1, CS2-O2), before testing whether brief presentation of these cues could substitute for the Os in non-rewarded probe trials initiated by the CSs in place of the Os (CS1: R1 vs R2, CS2: R1 vs R2).

These test trials were inserted into training sessions in which normal rewarded instrumental trials also occurred. Our results suggest that training with immediate rewards tend to promote a Pavlovian-instrumental transfer (PIT) effect, while delayed rewards tend to promote PIT or substitution effects in different subsets of animals. Evaluation of individual rats' data identified two subgroups: one showing a bias toward pressing the lever reinforced with the same outcome that was paired with the trial-initiating CS (e.g., CS1: R1 < R2, PIT effect), and one with a bias toward the lever correctly chosen after a specific trial-initiating outcome (e.g., CS1: R1 > R2, substitution effect). More individuals in the delayed reward group appeared to show the substitution-like effect than in the immediate reward group. We interpret this pattern to mean that CS-activated reward representations can sometimes resemble those representations activated by the reward itself, particularly when control by response-outcome associations have been weakened and control by outcome-response associations have been strengthened by increasing a response-reward delay. We plan to present additional data at SfN 2025 that further explores these effects using tests where control by differential S-O or R-O associations is weakened, as well as data that directly assesses with double-labeling immunohistochemistry whether CSs and USs activate the same neural representations in key reward processing areas.

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Poster

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Program #/Poster #: PSTR293.19/GG4

Topic: H.03. Motivation

Title: Effects of Insular Cortex Damage on Motivation and Cognition

Authors: R. FREMOUW, A. BERTHOLD, G. NASSIVERA, W. LEWISON, K. DAWSON, W. LAFAVE, L. BIER, M. DAVIS, N. CHESTOJANOVA, *W. E. DECOTEAU;
St. Lawrence Univ., Canton, NY

Abstract: This study examined the effect neurotoxic lesions of the right insular cortex have on motivation, anxiety, and cognition. Ten adult male Long Evans rats served as subjects. Animals were maintained at 85% of their free-feeding weight. Their baseline level of motivation was measured in an operant chamber using a progressive ratio 3 (PR3) schedule of reinforcement. Following ten days of PR3 training, all rats received under deep anesthesia stereotaxic injections (0.8 µl @ 0.2 µl/min) targeting the right insular cortex (AP +1.4; ML +5.2; DV -6.8 from bregma). Half the rats were infused with the neurotoxin quinolinic acid (10 mg/kg) dissolved in phosphate buffered saline (pH 7.4) while the other half received vehicle control injections. Surgery groups were assigned in a counterbalanced manner based on pre-surgery PR3 performance. Following a 2-week recovery from surgery, animals were retested on the PR3 task using regular food pellets as well as sucrose and chocolate pellets. Anxiety-like behaviors were also measured using two standard paradigms: the open-field test and the elevated plus maze.

Learning and memory were assessed using the Morris water maze task and, finally, social behavior was evaluated using the 3-chamber social perception task. We found that neurotoxic lesions targeting the right insular cortex increased operant chamber motivation relative to controls. This effect was not influenced by changing to the reinforcer to either sweet (sucrose) or chocolate flavored food pellets. To our surprise, insular lesions induced anxiogenic behavior by way of both decreased time in open areas of the testing apparatuses and increased defecation. Rats with insular lesions learned the standard version of the Morris water maze at a normal rate but showed a mild impairment on a post-acquisition retention probe trial. In terms of social perception, insular lesions disrupted rats' ability to distinguish between a novel and familiar conspecific. In summary, we found that targeted disruption of a small area of the right insular cortex led to sweeping behavioral and cognitive alterations in the rat. These findings suggest this relatively understudied brain region plays a central modulatory role in both fundamental and higher-order behavioral processes.

Disclosures: R. Fremouw: None. A. Berthold: None. G. Nassivera: None. W. Lewison: None. K. Dawson: None. W. LaFave: None. L. Bier: None. M. Davis: None. N. Chestojanova: None. W.E. DeCoteau: None.

Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR293.20/GG5

Topic: H.03. Motivation

Support: JSPS KAKENHI JP25H01365

Title: Modeling the emergence of intrinsic motivation using a chaotic recurrent neural network

Authors: *W. MIYAMORI^{1,2}, M. INOUE³, K. HOSODA²;

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Abstract: Intrinsic motivation, in contrast to extrinsic motivation driven by external rewards and punishments, is characterized by spontaneous engagement in behavior for its own sake.

According to self-determination theory (SDT), a framework developed in psychology, these two forms of motivation are situated on a continuum, and recent neuroscience research has increasingly identified their distinct neural correlates. However, the neurodynamic mechanisms linking intrinsic and extrinsic motivation remain poorly understood. In this study, we computationally investigated how intrinsic motivation might arise during the optimization of externally driven behavior using a neurodynamic modeling framework. We implemented a conventional rate-coded recurrent neural network (RNN) in which each unit's activity was updated over time through recurrent connections and external inputs. The model included three key parameters: a synaptic weight matrix (J), the global strength of neural interaction (β), and

sensitivity to environmental stimuli (γ), which are tentatively associated with the SDT's basic psychological needs of competence, autonomy, and relatedness, respectively. External task performance was defined as the accuracy in producing target outputs in response to inputs, starting from randomized neuronal initial states. A genetic algorithm was used to optimize J , β , and γ solely for external task performance. After training, intrinsic motivation was evaluated as the level of spontaneous activity observed in the absence of input stimuli. We found that, although the optimization exclusively focused on external performance, spontaneous activity emerged robustly in most of the tasks we tested, often exhibiting chaotic dynamics. In contrast, some task conditions demanding high-resolution output tended to suppress spontaneous activity. Furthermore, in conditions where changes in neural activity incurred a cost or large output errors were heavily penalized, spontaneous activity disappeared early in training but later reappeared. These findings are consistent with SDT, suggesting that enhanced external motivation quality can facilitate the emergence of intrinsic motivation, while punitive task structures may suppress it. Our study offers a novel neurodynamic framework that grounds motivational phenomena in biologically inspired computational principles, which complements conventional models that rely on detailed psychological constructs. This framework, while still in an early stage of development, may contribute to a deeper theoretical understanding of the computational principles underlying motivation.

Disclosures: W. Miyamori: None. M. Inoue: None. K. Hosoda: None.

Poster

PSTR293: Neural Mechanisms of Motivation and Reward-Related Behaviors

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR293.21/GG6

Topic: H.03. Motivation

Support: NSF CAREER Award 204611
NIH R01 MH124849
Brown OVPR Seed Award
NIH T32 MH126388
NIH R25 NS124530
NIH K99 MH133912

Title: Distinct neurocomputational signatures of mental effort when motivated by success versus failure

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Abstract: The effort a person puts into a task reflects a combination of their motivations to perform well and to avoid performing poorly. These sources of motivation jointly determine the amount and type of effort people invest, but are typically conflated within research on mental effort. We sought to disentangle the mechanisms underpinning these distinct forms of motivation, by separately varying the valence of expected outcomes (positive vs. negative) and whether these outcomes resulted from good performance (reinforcement) or bad performance (punishment). Across three fMRI studies, participants (total N=172) performed a cognitively demanding task under varying levels of positive reinforcement (monetary reward per correct response) and punishment (monetary loss for performing poorly). Participants were informed of their incentives prior to each trial, allowing us to distinguish neural signatures of incentive processing in anticipation of the task versus during task execution. Using a previously validated computational model of motivated control, we extracted behavioral signatures of reinforcement-driven motivation (reflecting more efficient responding with higher levels of reinforcement) and punishment-driven motivation (reflecting more cautious responding with higher levels of punishment). We found that these two forms of motivation were linked to distinct neural circuits. The motivation to achieve positive reinforcement was associated with activity in ventral striatum in anticipation of performing the task, and with activity in anterior insula (AI) and caudal regions of dorsal anterior cingulate cortex (dACC) both during anticipation and execution of the task. Punishment-related motivation, by contrast, was associated with activity in AI and rostral regions of dACC during anticipation and execution, as well as with inferior frontal gyrus during execution. As independent validation, we found that a previously described multivariate signature of reward anticipation was selectively predictive of positive reinforcement but not punishment-related motivation. By additionally varying negative reinforcement (monetary loss for failing to perform well) in one of the studies (N=37), we further tested whether its neural signatures more closely resembled positive reinforcement or punishment. We found that negative reinforcement shared signatures with positive reinforcement more so than with punishment, both during anticipation and execution. Together, these findings provide novel evidence that effort investment arises from a confluence of distinct motives (related to success vs. failure) underpinned by dissociable neural circuits.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.01/GG7

Topic: H.05. Mood Disorders

Support: NIH Grant R15 MH125306

Title: Selective orexin receptor cross-over (SORCO) treatment produces resilience

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Abstract: Balancing stress-responsiveness includes signaling between complex microcircuits in brain regions associated with pro-stress behavioral outputs, such as the anterior region of basolateral amygdala (aBLA). Modifications to this neurocircuitry, through experience or treatment, shift behavioral and molecular phenotypes of stress resilience and vulnerability. In aBLA, stress-associated genetic marker *Rspo2* (R-Spondin 2) found on excitatory (expressing *CamkIIa*) glutamatergic neurons marks circuit elements modulated by shifts in signaling responsible for enhanced stress-responsivity via orexin type I (Orx₁R, *Hcrtr1*) receptors. This pro-stress system is balanced by GABAergic neurons, stimulated by type II (Orx₂R, *Hcrtr2*) receptors that reduce pro-stress, and thus create anti-stress responses. Modifying this circuitry via icv or intra-BLA inhibition of Orx₁R or stimulation of Orx₂R signaling results in decreased anxious and depressive behavior, as well as neural responses. Thus, we hypothesized that a combination of inhibition of Orx₁R (SB-674042) together with activation of Orx₂R (YNT-185) would produce enhanced reduction of stress-induced affective behaviors. To examine this hypothesis, we used systemic brain delivery (icv) of the selective orexin receptor cross-over (SORCO; SB674042 + YNT-185) to target both pro-stress (inhibition) and anti-stress (stimulation) neurocircuitry and subjected the mice to aggressive social interaction. The Stress Alternatives Model (SAM) is a four-day social defeat/avoidance model that produces two stress-related phenotypes, Escape (resilient) and Stay (stress-vulnerable) that allow for measurable behavioral outcomes of phenotype development, including anxiety, motivation, fear, and socio-spatial learning. Treatment with SORCO produces phenotypic reversal (Stay to Escape) by increasing Escape behavior during SAM interactions, more preference during Social Interaction/Preference testing, and less freezing during the Conditioned Response (CR) test. These behavioral adaptations coincide with an increase in *Hcrtr1*, but not *Hcrtr2* mRNA in *Rspo2*-positive cells of pro-stress circuitry in aBLA.

Disclosures: M.M. John: None. J.D. Yaeger: None. C.H. Summers: None.

Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.02/GG8

Topic: H.05. Mood Disorders

Support: NIH Grant R15 MH125306

Title: Aggression as a contributing factor to social defeat, stress vulnerability and resilience

Authors: W. J. KORZAN¹, M. JOHN², M. A. PRATT³, J. D. YAEGER⁴, R. P. WATERS⁵, P. J. RONAN⁶, *C. H. SUMMERS²;

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Abstract: The Stress Alternatives Model (SAM) is a social defeat/avoidance paradigm developed in our lab that reveals evolutionarily conserved escape responses in fish, hamsters, rats, and mice. During social interactions in a neutral arena, available escape routes sized exclusively for smaller test animals allow for avoidance of a social aggressor. This 4-day social interaction protocol pairs C57BL/6N test mice and a larger, novel, aggressive CD1 mouse each day. Although escape portals are available, and the CD1 aggressor is unremittingly antagonistic, only half of the mice tested utilize the escape tunnels, while escape latency dramatically decreases over time in mice that escape. We sought to determine whether aggression provided the trigger of two stress-related phenotypes that are produced by the SAM. The results suggest threat of aggression, determined by the first attack, is necessary for phenotype development, but the intensity of aggression over time does not determine which phenotype is chosen. Phenotypes are determined by responsiveness and counterbalanced neurocircuits that promote stress-resilience or vulnerability. These stress neurocircuits are modulated by orexins, through orexin 1 and 2 receptors (Orx₁, Orx₂), which promote pro-stress behaviors. In the primary pro-stress neurocircuitry of the aBLA, we examined *Akt* and *Mtor* gene expression in stress-resilient (Escape) and -vulnerable (Stay) mice. The quantity of *Hcrtr1* mRNA/cell was elevated in Stay mice, as were the mRNA/cell numbers for *Mtor*. However, the increase of *Akt2* and *Mtor* mRNA/cell was not evident specifically in *Hcrtr1* expressing cells, suggesting these molecular markers of neuroplasticity are not being activated by Orx₁ receptors.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.03/GG9

Topic: H.05. Mood Disorders

Support: H2020-ITN-MSCA Grant agreement 955684

Title: Diurnal regulation of rapid-acting antidepressant targets and clock gene modulation by nitrous oxide

Authors: *G. GONZÁLEZ HERNÁNDEZ^{1,2}, S. TORRES-SANCHEZ², S. V. ROZOV¹, E. BERROCOSO², T. RANTAMAKI¹;

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Abstract: Depression and the circadian system are closely linked, with disrupted circadian rhythms contributing to depressive states, and their restoration playing a key role in antidepressant effects. Rapid-acting antidepressants (RAADs) like ketamine and nitrous oxide (N₂O), appear to influence neuroplasticity and circadian pathways in mood-related brain regions, including the prefrontal cortex (PFC) and hippocampus (HC), yet this interplay remains relatively unexplored. Here, we investigated the diurnal expression and phosphorylation patterns of RAAD-associated molecular targets—such as Dual-Specificity Phosphatases (DUSP), GSK3B, and TrkB—in male and female naïve mice (C57BL/6J), using qPCR and Western blot. These targets exhibited diurnal oscillations in males, with most transcripts peaking during the dark phase (ZT15-18). Females showed similar oscillations in HC, but lower amplitude rhythmicity in the PFC. Phosphorylation patterns were similar between PFC and HC, with TrkB and GSK3B displaying higher phosphorylation levels during daytime. These data informed the design of chronotherapeutic studies exploring time-of-day-dependent responses to N₂O. In follow-up experiments, we assessed N₂O's impact at ZT3 vs ZT15 in two preclinical models of depression: a chronic corticosterone (CORT) model and the chronic constriction injury (CCI) model of neuropathic pain. Both models confirmed the significant impact of circadian time and N₂O treatment on the selected molecular targets, although no single time-point consistently optimized the molecular response across markers. Additionally, N₂O significantly modulated circadian clock genes expression (e.g. Per1, Per2) in the PFC, supporting its potential role in circadian resynchronization. This work demonstrates that molecular targets potentially implicated in antidepressant responses are under circadian regulation. It further shows that RAADs modulate core clock genes and neuroplasticity-related markers in naïve animals as well as in preclinical models of depression, underscoring the circadian system's potential as a therapeutic target. Together, these results provide a foundation for the development of novel chronotherapeutic strategies for the treatment of mood disorders and their comorbidities, although further research is needed to define optimal intervention windows. Moreover, region- and sex-specific molecular dynamics were observed, emphasizing the complexity of these mechanisms and pointing to critical biological variables that may inform individualized treatment strategies.

Disclosures: G. González Hernández: None. S. Torres-Sanchez: None. S.V. Rozov: None. E. Berrocso: None. T. Rantamaki: None.

Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.04/GG10

Topic: H.05. Mood Disorders

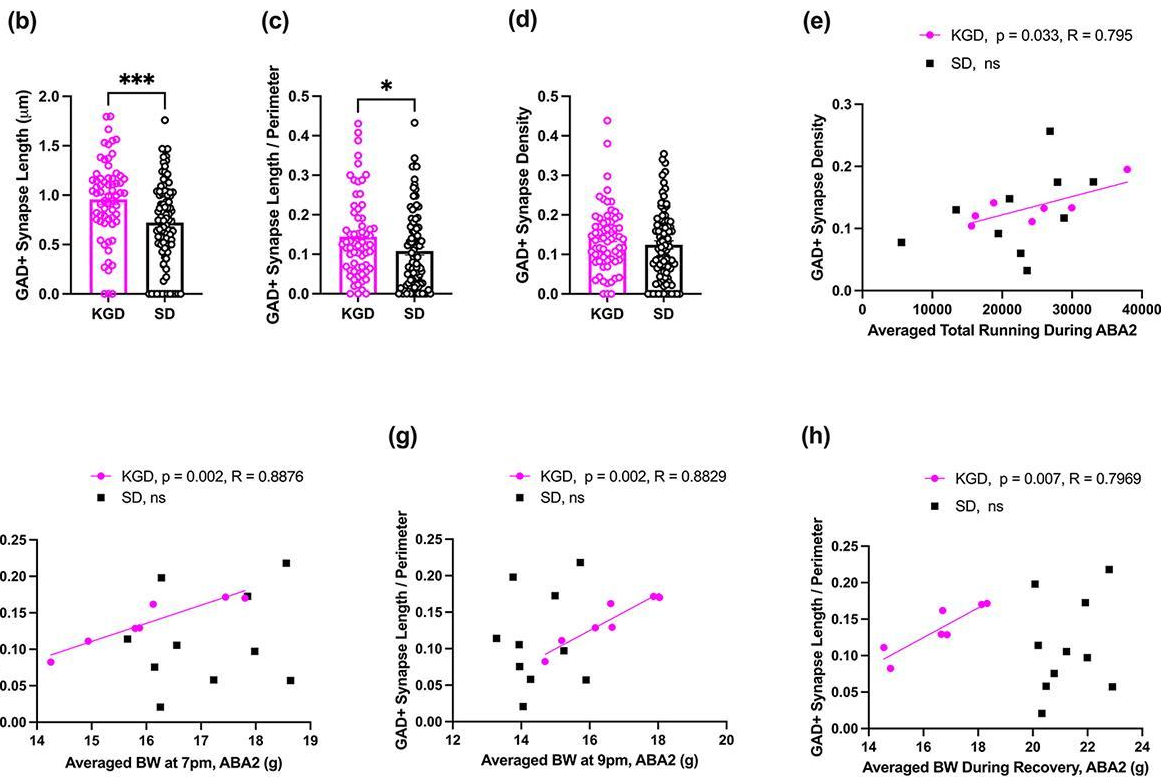
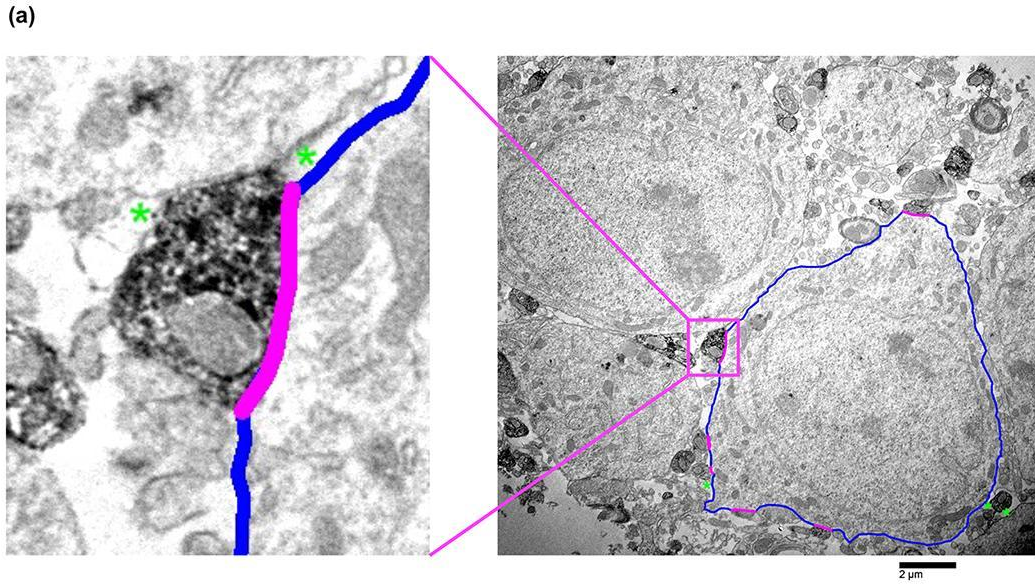
Support: Vulnerable Brain Project Grant
Baszucki Group Grant
NYU DURF grant

Title: Ketamine sustains and enhances the protective effects of ketogenic diet against relapses in a mouse model of anorexia nervosa: examination of GABAergic synaptic plasticity in the hippocampus by electron microscopic immunocytochemistry

Authors: Y. DONG^{1,3}, J. WANG¹, O. GE¹, Y. LIN¹, *C. J. AOKI^{2,4,1};

¹Ctr. for Neural Sci., ²New York Univ., New York, NY; ³Neurosci., Univ. of Chicago, Chicago, IL; ⁴Neurosci. Inst., NYU Langone Sch. of Med., New York, NY

Abstract: Anorexia nervosa (AN) is a mental disorder with high mortality and relapse rates, yet no accepted pharmacotherapies. Reports on six individuals indicate that ketogenic diet (KGD) combined with sub-anesthetic ketamine (KET) may be an effective intervention for AN relapses. Activity-based anorexia (ABA) is an animal model that captures AN's maladaptive behaviors of voluntary food restriction, excessive exercise, severe weight loss and heightened anxiety. Using ABA, we tested (1) whether KGD, alone, can protect against relapses after a severe anorexia-like experience; (2) whether KGD must be maintained to prevent relapses; and (3) whether KET combined with KGD is more ameliorative than KGD alone. KGD's potential for protecting against relapses was tested by imposing ABA induction twice (ABA1, ABA2), with 10 days of recovery in between. Animals were fed a standard diet (SD) of pellet plus wet food during ABA1 and KGD during ABA2 (SD-to-KGD). ABA vulnerability, measured by food restriction-evoked hyperactivity and weight loss, was severe during ABA1 but significantly less during ABA2, compared to those fed SD throughout ABA1 and ABA2 (SD-to-SD). Conversely, when KGD was withdrawn during ABA2 (KGD-to-SD), vulnerability became as severe as for the SD-to-SD. Thus, KGD must be maintained to be protective. We then tested whether KET plus KGD during ABA1 can protect against ABA2 relapse in the absence of KGD (KGD+KET-to-SD). During ABA2, KGD+KET-to-SD exhibited similarly low maladaptive wheel running and greater food consumption compared to animals maintained on KGD throughout ABA1 and ABA2 without the ketamine treatment. Thus, KET sustains and boosts KGD's benefits for >10 days. Electron microscopic analysis revealed that KGD, more than SD, increases lengths and proportions of hippocampal pyramidal cell membranes receiving GABAergic inhibitory synapses, and that GABAergic synapse density correlates with reduction of ABA vulnerability. These results suggest that short-term KGD+KET offers a durable, translationally relevant strategy for reducing AN relapses.



Disclosures: Y. Dong: None. J. Wang: None. O. Ge: None. Y. Lin: None. C.J. Aoki: None.

Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.05/GG11

Topic: H.05. Mood Disorders

Support: Vulnerable Brain Project Grant
Baszucki Group Grant
NYU DURF grant

Title: Ketogenic Diet Enhances Resilience to Anorexia-like Behavior in Rodent Model via Bi-directional GABAergic Interneuron Synaptic Plasticity in the Hippocampus: an Electron Microscope-Behavior Study.

Authors: *E. KIM^{1,3}, Y. DONG^{2,4}, J. WANG², C. J. AOKI^{1,3};

¹Ctr. for Neural Sci., ²New York Univ., New York, NY; ³Neurosci., NYU Langone Sch. of Med., New York, NY; ⁴Neurosci., Univ. of Chicago, Chicago, IL

Abstract: Anorexia nervosa (AN) is an eating disorder with the second-highest mortality rate and high relapse rates (~25%), yet no FDA-approved pharmacologic treatment at present. The Activity-Based Anorexia (ABA) rodent model captures key maladaptive behaviors of AN: voluntary food restriction, hunger-evoked hyperactivity, and heightened anxiety, which can collectively cause weight loss so severe as to cause mortality. ABA is induced by combining wheel access with restricted food availability. Recent findings from the Aoki Lab showed that ketogenic diet (KD) reduces vulnerability of adult mice to AN-like relapses, as measured by the suppression of excessive wheel running and attenuated weight loss during the food restriction period. However, the precise neurobiological mechanisms underlying KD's protective effects remain poorly understood, with the exception of recent evidence suggesting that KD enhances GABAergic inhibition at cell bodies of CA1 pyramidal neurons. I hypothesize that KD-induced bi-directional GABAergic modulation (inhibitory versus disinhibitory) onto CA1 pyramidal cell dendrites may also contribute toward the amelioration of AN relapse severity. To test this, electron microscopic immunocytochemistry was employed to systematically quantify two forms of GABAergic synaptic contacts in stratum radiatum and stratum lacunosum-moleculare of the dorsal CA1 of ABA mice fed either KD or standard diet (SD): (1) GABAergic inputs onto pyramidal neurons, which suppress hippocampal excitability, and (2) GABAergic inputs onto GABAergic interneurons, which promote disinhibition and enhance excitability. Compared to SD-fed mice, KD-fed mice had significantly less GABA-to-GABA disinhibition ($p = 0.0089$, unpaired two-tailed t-test) and greater GABAergic inhibition onto pyramidal cell dendrites ($p = 0.0089$). These results support the idea that KD-mediated gain of resilience against ABA relapse is driven by reduced disinhibition and enhanced GABAergic inhibition, together suppressing hippocampal excitability. Thus, KD can be adopted as a promising therapeutic approach for preventing future relapses in individuals diagnosed with AN.

Disclosures: E. Kim: None. Y. Dong: None. J. Wang: None. C.J. Aoki: None.

Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.06/GG12

Topic: H.05. Mood Disorders

Title: Naringenin, but not curcumin, produces dose-dependent anti-anxiety like effects in the chick social-separation stress test

Authors: *B. HURT¹, A. MCCABE², S. W. WHITE³;

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³Psychology, Appalachian State Univ., Boone, NC

Abstract: Phytochemicals are biologically active compounds found in plants and possess numerous health benefits such as anti-cancer, anti-ageing, antioxidant, and anti-inflammatory, and neuroprotective properties as well as promoting healthy immune system functioning. Preclinical and clinical data suggest several phytochemicals, such as curcumin and naringenin, produce anxiolytic and/or antidepressant effects. However, these findings have not been evaluated in a model of treatment-resistant depression (TRD). Using the TRD Black Australorp genetic line, the current study sought to evaluate the potential anxiolytic and/or antidepressant effects of curcumin and naringenin in the chick social-separation stress test, a dual drug screening assay. Separate dose-response studies were conducted for both phytochemicals at doses of 25, 50, & 100 mg/kg. Starting on day-aged 2, male Black Australorps were orally administered either vehicle or one of the three doses of curcumin or naringenin twice a day for 8-9 days. On day aged 9 & 10, animals were exposed to a 90 min isolation stressor which elicits distress vocalizations (DVocs). Sample sizes for all groups were n = 12 - 18. In both studies, vehicle-treated animals displayed high DVoc rates during the first five min of isolation indicative of a panic-like state (i.e., anxiety-like phase) which gradually decline and remain low but stable for the last hour of isolation (31 - 90 min) indicative of behavioral despair (i.e., depression-like phase). Curcumin had no effect on DVoc rates during either phase of the test. However, naringenin caused a dose-dependent reduction in DVoc rates during both phases of isolation. A one-way ANOVA for the anxiety-like phase approached significance [$F(3,62) = 2.300, p = .086$]. Post-hoc test using Fisher's LSD revealed naringenin 100 mg/kg significantly reduced DVoc rates compared to vehicle ($p = .05$) representing an attenuation of panic. A one-way ANOVA for the depression-like phase revealed a significant effect for treatment [$F(3,62) = 2.820, p = .046$]. Post-hoc tests using Fisher's LSD revealed naringenin 100 mg/kg significantly reduced DVoc rates during the depression-like phase. Preclinical and clinical studies report curcumin displays anxiolytic and antidepressant effects, yet this study failed to detect such effects, although further testing is needed. The results also suggest naringenin may be useful in reducing symptoms of panic. However, the reduction in DVoc rates during the depression-like phase resemble sedative effects produced by anxiolytics in the model. Additional studies are needed to replicate these findings and determine naringenin's anxiolytic mechanism.

Disclosures: B. Hurt: None. S.W. White: None.

Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.07/GG13

Topic: H.05. Mood Disorders

Title: Betanin, the primary phytochemical from the beet root plant, produces anxiolytic-like and diuretic-like effects in the chick social-separation stress test

Authors: *S. W. WHITE¹, V. A. VILLARREAL²;

¹Appalachian State Univ., Boone, NC; ²Psychological & Brain Sci., Texas A & M, College Station, TX

Abstract: Clinical and preclinical reports suggest betanin, the primary betacyanin phytochemical from the beetroot plant, can reduce symptoms and biomarkers associated with anxiety and/or depression. However, these findings have not been evaluated in a model of treatment-resistant depression (TRD). Utilizing the Black Australorp TRD genetic line, the purpose of this study was to evaluate betanin's potential anxiolytic and/or antidepressant effects in the chick social-separation stress test. Beginning on day-aged 2, male Black Australorp chicks received either saline or betanin 50 mg/kg twice daily orally for 8-9 days. Separate groups of chicks received either clonidine 0.15 mg/kg or maprotiline 2.5 mg/kg i.m 30 min prior to isolation for anxiolytic and antidepressant comparisons, respectively. On day-aged 9-10, chicks were exposed to a 90-min isolation stressor which elicits distress vocalizations (DVocs) as the primary behavioral endpoint. Sample sizes for all treatment groups were $n = 16-18$ and treatments were randomly assigned. An additional goal of the study was to assess weight gain across the saline and betanin groups. For the anxiety-like phase (0-5 min), a one-way ANOVA revealed a significant effect of treatment [$F(3,64) = 52.517, p < .001, \eta_p^2 = .711, 1-\beta = 1.000$]. Post-hoc via Fisher's LSD revealed clonidine 0.15 mg/kg ($p < .001$) and betanin 50 mg/kg ($p = .009$) significantly lowered DVoc rates compared to saline, indicative of attenuation of panic (i.e., anxiolytic-like effects). For the depression-like phase (31-90 min), the one-way ANOVA revealed a significant effect of treatment [$F(3,64) = 7.162, p < .001, \eta_p^2 = .251, 1-\beta = .976$]. Fisher's LSD post-hoc tests revealed maprotiline 2.5 mg/kg significantly elevated DVoc rates ($p < .001$; i.e., antidepressant-like effect) compared to saline. Research suggests betanin also possesses diuretic effects and an independent samples t-test revealed betanin-treated animals change in weight (per day) was significantly less than saline-treated animals [$t(40) = 2.289, p = .027, d = .706$]. These findings suggest betanin may be useful in the treatment of anxiety disorders but may possess diuretic effects. Future studies should attempt replicate betanin's anxiolytic-like effects and weight as well as measure betanin's effect on biomarkers associated with anxiety such as corticosterone and gut microbiota.

Disclosures: S.W. White: None. V.A. Villarreal: None.

Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PST294.08/Web Only

Topic: H.05. Mood Disorders

Support: DOST-PCHRD Grant DOST2020-04-A1-265

Title: *Lactobacillus rhamnosus* GG prevents increases in BDNF in the blood plasma of female rats exposed to chronic unpredictable mild stress

Authors: K. P. MATIAS¹, V. Q. CARIAGA¹, S. A. CRUZ¹, *G. J. QUIRK², B. I. BULATAO^{1,3}, R. M. MANALO-CABALINAN^{1,4};

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Abstract: Depression is a widespread mental health disorder affecting 3.3 million adults in the Philippines, with a higher prevalence among women compared to men. The high cost of SSRIs deters people with depression from seeking treatment. Thus, we explored the antidepressant effects of the readily available probiotic *Lactobacillus rhamnosus* GG (LGG). Forty 8-week old Sprague-Dawley rats (male, n=22; female, n=18) were subjected to chronic unpredictable mild stress (CUMS) for 6 weeks to induce depression-like behavior. The rats underwent an additional 4 weeks of CUMS, with daily administration of 10¹⁰ CFU/mL LGG or saline via oral gavage. A non-stressed control group (male, n=7; female, n=8) was housed in a controlled environment with *ad libitum* food and water. The Sucrose Preference Test (SPT) and Forced Swim Test (FST) were conducted post-LGG administration. Following behavioral testing, blood plasma was collected for the quantification of corticosterone and brain-derived neurotrophic factor (BDNF) levels. While there was no significant difference in the SPT in either sex, there was a trend toward increased immobility time in CUMS female rats during the FST (control: 75.36 sec, CUMS: 102.7 sec, $t=1.788$ (17,7), $p = 0.086$). Treatment with LGG in CUMS females did not significantly decrease immobility time (LGG: 95.45 sec, ANOVA main effect of group: female, $p=0.121$). Interestingly, biological markers revealed sex-specific effects. We observed a trend toward increased corticosterone in CUMS females (control vs CUMS-sal: 1.868 (6,5), $p=0.089$) but not in males (control vs CUMS-sal: $t=1.268$ (4,9), $p=0.227$). However, BDNF showed a highly significant increase in CUMS females (control: 237.5 pg/ml, vs CUMS-sal: 1,184 pg/ml, $t=3.991$ (6,6), $p=0.002$) but not in males (control vs CUMS-sal: $t=0.942$ (5,9), $p=0.362$). Furthermore, the administration of LGG in CUMS female rats significantly decreased plasma BDNF to control levels (CUM-sal: 1,184 pg/ml, vs CUMS-LGG: 305.3 pg/ml, $t=3.349$ (6,9), $p=0.004$). These findings suggest that the CUMS model produced more pronounced depression-like symptoms in female rats than in male rats, replicating earlier findings (Dalla et al., 2005). Treatment with LGG can reverse the effect of stress on plasma BDNF levels, suggesting a possible beneficial effect of probiotics in stressed females.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR294.09/GG14

Topic: H.05. Mood Disorders

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MH138104-01, N.W.M.
24-4860, A.F., J.R.-R
HEU-101109833-PNOCMD, A.F.
1018134.0, N.C.P.
31213, J.R.-R

Title: Subcutaneous minipumps containing a GnRH-R antagonist reversibly suppress the estrous cycle in female mice

Authors: *I. CASTRO RIVERA^{1,2}, A. FLORIDO^{6,7}, H. POTTHOFF², N. W. MILLER^{8,3}, J. RODRIGUEZ-ROMAGUERA^{8,4,5};

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Abstract: Psychiatric disorders are not reflected equally across sexes. Females are more than twice as likely to suffer from stress-related disorders. These differences are thought to be related to the effects of reproductive hormones on the brain circuits that regulate anxiety- and mood-related behaviors. Current methods that suppress reproductive hormones include (1) gonadectomies that are invasive and non-reversible or (2) daily injections of a gonadotropin releasing hormone receptor (GnRH-R) agonist which is reversible but initially generates a flare in estrogen levels. In this study, we evaluate if implanting osmotic minipumps containing a GnRH-R antagonist called cetrorelix acetate will reversibly suppress the estrous cycle in female mice without inducing a flare. To test the efficacy of this approach, we subcutaneously implanted osmotic minipumps containing either cetrorelix acetate or saline in either adult or prepubertal female mice. We collected daily vaginal smears and determined the stage of the estrous cycle. A postmortem tissue analysis of the ovaries was also performed to assess the maturation of ovarian follicles. The onset of puberty in prepubescent female mice was assessed daily by monitoring vaginal opening. Our preliminary results show that cetrorelix acetate arrests the estrous cycle in adult female mice (n=5) and prevents the onset of puberty (n=4), as confirmed by unvarying cytologies in adult mice and lack of vaginal opening in adolescent mice. Additionally, once

cetrorelix acetate is no longer present, mice revert to natural cycling patterns. In summary, we are observing the efficacy and reversibility of minipumps containing cetrorelix acetate to suppress the estrous cycle. Future studies will increase the sample size and determine if minipumps containing cetrorelix acetate prevent the initial flare by measuring estrogen levels using mass spectrometry within the first 24 hours of implantation.

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Poster

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Topic: H.05. Mood Disorders

Support: Work on "Non-Invasive Spectroscopic Quantification of Depression Biomarkers" is supported by Wellcome Leap as part of the Multi-Channel Psych Program

Title: Pharmacokinetic Evaluation of the Effect of Sapropterin Dihydrochloride on Tetrahydrobiopterin Concentrations in Cerebrospinal Fluid of Sheep

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Abstract: Fifteen percent of the twenty-one million Americans with clinical depression are non-responsive to treatments thus classifying their depression as treatment refractory. Tetrahydrobiopterin (BH4) is a cofactor required for synthesis of dopamine, serotonin, norepinephrine, melatonin, and pain modulators. A BH4 deficiency has been identified in the cerebrospinal fluid (CSF) of a subset of patients diagnosed with treatment-refractory depression. Sapropterin dihydrochloride (SAP), a synthetic form of BH4, is prescribed to patients with phenylketonuria. Recently, an off-label prescription of SAP was found to improve symptoms of treatment refractory depression in patients with low CSF BH4. However, there is a lack of pharmacokinetic data on CSF concentrations of BH4 following oral administration of SAP. In this study, SAP was administered to adult female sheep (n = 4; 1500 mg daily via gastrostomy tube) for 5 days. CSF samples were collected from an intracisternal magna catheter for acute and steady-state pharmacokinetic analysis. We hypothesized that there would be an increase in CSF concentrations of BH4 following SAP administration via gastrostomy tube in sheep. A non-compartmental analysis using a two accessible pool model, an exogenous input pool (stomach) and a sampling pool (CSF), was applied to determine pharmacokinetic parameters in this study. The Day 5 (assumed steady-state) BH4 normalized concentration was fit to a constant plus mono-exponential function, with the constant indicating the baseline, endogenous BH4

concentration and the mono-exponential function indicating the decrease in BH4 concentration over time, arising from either the exogenously delivered BH4 (SAP) crossing the blood brain barrier or stimulating the production of additional endogenous BH4. The normalized mean residence time of Kuvan in the CSF, after the exogenous bolus, was ~2.5 hrs. and the normalized relative clearance from the CSF was 0.00064 mol/hr. In this study, we confirmed that BH4 administered into the stomach results in an increase in BH4 concentration in the brain, either from SAP crossing the blood brain barrier and entering the brain or SAP inducing increased endogenous production of BH4. The mean residence time and clearance rate determined in this study can be beneficial in determining appropriate dosing rates for using SAP in patients with treatment-refractory depression.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

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Program #/Poster #: PSTR294.11/HH1

Topic: H.05. Mood Disorders

Support: NIH Grant R16GM154678

Title: Impact of escitalopram on postpartum sleep/wake activity and maternal behaviors

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Abstract: The postpartum period is characterized by profound physiological and psychological changes, including disruptions in sleep and activity patterns, which may contribute to the onset of postpartum depression. If left untreated, postpartum depression can result in long-term consequences, including cognitive impairments, emotional dysregulation, impaired maternal-infant attachment, and the onset of sleep-related disorders. Selective serotonin reuptake inhibitor (SSRI), escitalopram, is prescribed for the treatment of postpartum depression due to its efficacy, favorable safety profile, and relatively low cost. While escitalopram is considered an effective treatment for depression and anxiety, far less is known about its impact on maternal behaviors during the postpartum period. This study aims to (1) characterize sleep/wake patterns and affective-like behaviors across the perinatal period, and (2) investigate the impact of escitalopram on maternal behaviors when administered during the postpartum period. To this end, nulliparous Long-Evans female rats were administered escitalopram 10 mg/kg or control (IP) for 10 consecutive days during the postpartum phase (P2-11). An array of behaviors were

assessed before pregnancy (baseline), at gestation days 7 and 14, and postpartum days 2, 11, and 16. Activity levels were monitored over a 24-hour period using non-invasive activity tracking devices. Sucrose preference test, open field, and maternal-pup interactions were assessed. Preliminary results suggest that rest/wake patterns and affective-like behaviors changed across gestation and postpartum periods. In assessing preconception compared to pregnancy, activity during the dark period became more fragmented. Time spent in the center of the open field decreased from preconception to the postpartum period. Escitalopram treated dams demonstrate a significant reduction in the number of rest episodes, more anxiety-like behavior on the open field, and an increase in pup interaction early in the postpartum period compared to controls.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

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Program #/Poster #: PSTR294.12/HH2

Topic: H.05. Mood Disorders

Title: Electroconvulsive seizure regulates the transcription of PAR family of bZIP genes via *Egr1* in the rat frontal cortex

Authors: *Y. KIM¹, S. KIM², H. PARK³;

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Abstract: The PAR family of bZIP genes defined by the presence of a proline and acidic amino acid-rich protein (PAR) domain is consist of three polypeptides, D-box binding protein (Dbp), thyrotroph embryonic factor (Tef), and hepatic leukaemia factor (Hlf). Dbp controls the brain excitability, and was suggested as the candidate gene of mood disorder. Tef affects transcription of prolactin genes and thyroid-stimulating hormone-beta gene. Hlf plays important role in cell survival and proliferation. Dbp, Tef, and Hlf display similar circadian and tissue-specific expression but have different target promoter preferences. In this study, we investigated the effect of a single and repeated treatment of electroconvulsive seizure (ECS), an animal model of ECT, on the transcription of PAR family of bZIP genes in the rat frontal cortex. Single treatment of ECS (E1X) induced time-dependent changes in the mRNA expression level of *Dbp*, *Tef*, and *Hlf* in rat frontal cortex. mRNA levels of three genes were significantly decreased at 60 min after ECS. The repeated ECS treatments for 10 days (E10X) significantly up-regulated mRNA levels of *Dbp*, *Tef*, and *Hlf* compared to those in sham control group at 24 h after E10X in rat frontal cortex. The expression of three genes showed same pattern of changes in response to ECS treatment, indicating possible common regulatory mechanisms. We examined the transcription factor binding sites within the promoter regions (encompassing 1,500 bp from transcription start site) of *Dbp*, *Tef*, and *Hlf* genes. The binding sites for early growth response-1 (*Egr1*), Rev-Erba,

and RAR-related orphan receptor alpha (*Rora*) were commonly found in the promoter regions. mRNA expression of *Egr1*, *Rev-Erba*, *Rora* after ECS treatments were investigated. As results, mRNA level of *Egr1* was increased at 60 min after E1X, and decreased at 24 h after E10X as previously reported [3, 4], which suggest that *Egr1* could negatively regulate the transcription of *Dbp*, *Tef*, and *Hlf* genes. Being analyzed using chromatin immunoprecipitation assay (ChIP), the binding of *Egr1* to promoter of *Dbp*, *Tef*, and *Hlf* were was increased after ECS treatments. In addition, the transcripts of *Dbp*, *Tef*, and *Hlf* were significantly up-regulated in the frontal cortex of *Egr1*^{-/-} mice brain compared to those in wild type mice. In summary, ECS treatments affects the transcription of *Tef*, *Hlf*, and *Dbp*, via negative transcriptional regulation of *Egr1* in the rat frontal cortex. The common effects of ECS on the different members of PAR family of bZIP genes could provide understanding on the various effects of ECT on various cognitive, mood, and physical domains, which will require further functional studies.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

Location: SDCC Halls B-H

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Program #/Poster #: PSTR294.13/HH3

Topic: H.05. Mood Disorders

Title: Antidepressant-like effects of repetitive transcranial brain stimulation in mice

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Abstract: Despite the availability of antidepressants, nearly 50% of patients with depression fail to achieve remission, highlighting the need for alternative treatments. Repetitive transcranial magnetic stimulation (rTMS) targeting the dorsolateral prefrontal cortex (DLPFC) is an effective neuromodulation technique for treating depression in humans. However, its underlying mechanisms remain poorly understood due to limited preclinical research stemming from challenges in delivering focal stimulation in animals. Here, we developed a translational model of rTMS by applying repetitive transcranial brain stimulation (rTBS) to the prelimbic cortex—homologous to the human DLPFC—in female BALB/cJ mice. We predicted that using a chronic rTBS protocol equivalent to effective rTMS protocols used in humans, chronic rTBS would reverse the depression-like behavioral effects of chronic mild stress (CMS) in mice. We surgically implanted an electrode targeting the left prelimbic cortex of mice, and used stimulation parameters scaled from effective clinical protocols. Mice were subjected to either CMS or standard facility housing for six weeks, with either daily rTBS or sham stimulation administered during the final three weeks. At the end of the six-week period, behavioral

assessments, including the social interaction test and open field test, were conducted to evaluate antidepressant-like effects. We demonstrated that chronic rTBS significantly increased sociability in the social interaction test only in mice receiving CMS without altering locomotion in the open field test. To further investigate how rTBS affects neural activity, we conducted c-Fos immunohistochemistry in a separate cohort of animals after a single rTBS session. Preliminary results revealed increased c-Fos expression in the PrL, the site of stimulation. Analyses of additional brain regions are ongoing. Together, we demonstrate that rTBS initially increases neuronal activity in the prelimbic cortex, and chronic rTBS reduces stress-induced alterations in behavior in mice. This model provides a translational framework for investigating rTMS mechanisms in humans, and circumvents some of the technological limitations that have hindered its application in preclinical research.

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Poster

PSTR294: Mood Disorders: Animal Models of Therapeutics

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Topic: H.05. Mood Disorders

Support: Italian Ministry of Health (RF-2021-12373484; CUP: C53C22001570001) "Ricerca Corrente 2025" from Fondazione Policlinico Universitario "A. Gemelli" IRCCS

Title: Transcranial direct current stimulation ameliorates cognitive and emotional profiles in a mouse model of ischemic stroke of the primary motor cortex

Authors: *C. D'AMELIO^{1,2}, C. FEROLETO^{1,2}, I. PAOLETTI², C. CALIGIURI¹, M. MOROTTI², C. CODAZZI¹, F. D'ALELIO², L. LEONE^{1,2}, C. GRASSI^{1,2}, M. V. PODDA^{1,2}; ¹Univ. Cattolica del Sacro Cuore, Rome, Italy; ²Fondazione Policlinico Universitario A. Gemelli IRCCS, Rome, Italy

Abstract: Post-stroke depression (PSD) is the most prevalent psychiatric disorder following cerebrovascular events, impacting nearly one-third of stroke survivors. Clinically, PSD is characterized by symptoms like anxiety, hopelessness, anhedonia, social withdrawal, and cognitive impairments, which reduce quality of life and impede the rehabilitation process post-stroke. Therefore, there is an urgent need to identify effective therapeutic strategies to alleviate the multifaceted impacts of stroke-related psychiatric and cognitive complications. Transcranial Direct Current Stimulation (tDCS) has recently emerged as a promising non-invasive neuromodulatory technique for neurological and psychiatric disorders, including depression and cerebral ischemia. In line with this evidence, our research group previously demonstrated that bihemispheric tDCS facilitates motor recovery in a mouse model of photothrombotic ischemic

stroke of the primary motor cortex (M1), enhancing cortical connectivity and structural plasticity (PMID:35291824). Since M1 region is densely interconnected with the prefrontal cortex and limbic circuits, we employed the same animal model to elucidate the specific contribution of focal cortical damage to the genesis of neuropsychiatric symptoms and to investigate tDCS potential in mitigating long-term post-stroke cognitive and emotional alterations. Stroke mice received either tDCS or sham treatment daily for three consecutive days, starting from 72 hours post-stroke, and were tested for behavioral outcomes at four weeks after stroke. Cognitive profile was assessed using novel object recognition, object place recognition and Y-maze tests. Forced swim, elevated plus maze and open field tests were employed to evaluate the emotional profile while three chambers and social interaction tests were used to assess social behaviors. To explore the underlying physiological mechanisms of tDCS, synaptic plasticity was assessed by ex vivo electrophysiological recordings to measure long-term potentiation (LTP) at CA3-CA1 synapses in the hippocampus. Our results indicated that sham-stroke mice exhibited anxiety, depressive behavior, social deficits, and cognitive impairment compared to the control group, which were significantly ameliorated by tDCS. Furthermore, electrophysiological recordings showed a significantly increased LTP value at CA3-CA1 synapses of the ipsilesional hippocampus in mice subjected to tDCS compared to the sham group. These findings emphasize tDCS potential as a multifaceted therapeutic intervention not only for motor rehabilitation, but also to address stroke-related psychiatric symptoms.

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Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

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Title: Drug addiction vulnerability linked to high novelty-seeking: The role of G Protein-Coupled Receptor Kinases

Authors: *R. I. GATICA¹, B. I. RIVERA¹, S. DAVILA¹, C. PINO¹, C. STAROCELSKY¹, F. GONZÁLEZ-REINOSO¹, R. MORENO MARTÍNEZ², P. AGUILERA MATURANA¹, B. MANRIQUEZ¹, M. GARCIA¹, F. CODOCEDO¹, C. GUZMÁN MUÑOZ¹, M. SCHENKE¹, A.

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Abstract: Prolonged consumption of drugs of abuse can lead to addiction, although only a proportion of individuals develop compulsive use. High novelty-seeking is an endophenotype linked to increased drug addiction vulnerability, characterized by a hyperdopaminergic profile and lower dopamine D2 receptor (D2R) expression in the striatum. However, the mechanisms underlying this association remain unknown. G protein-coupled receptor kinases (GRKs) are key regulators of receptors like D2R and modulate their desensitization, internalization, and recycling. Previous studies showed that GRKs regulate the dopaminergic system and the locomotor effects of drugs of abuse, but their role in modulating novelty-seeking behavior has not been explored. Here, we studied the relevance of GRKs in D2R regulation and novelty-seeking behavior in rodents. First, we analyzed the presynaptic and postsynaptic localization of GRK2, GRK3, GRK5, and GRK6 in mesencephalic dopaminergic circuits, using immunofluorescence in synaptosomes and fixed brain tissue of adult male Sprague Dawley rats. We found that GRK2 and GRK5 are expressed in medium spiny neurons (MSNs) and dopaminergic neurons, while GRK3 and GRK6 showed more restricted expression in these neuron populations. We also found that GRK2 is expressed in MSNs and dopaminergic neurons in adult male C57BL/6J mice. We then studied the effect of an acute injection of the D2R agonist quinpirole (QNP) 0.5 mg/kg ip in striatal GRK levels, using western blot. In rats, acute QNP injection induced a transient increase in GRK2 levels in the nucleus accumbens, whereas GRK6 showed increased levels 24 hours after injection. Notably, these effects were not found after acute injection of QNP 1.0 mg/kg ip in mice. Next, GRK levels in striatal synaptosomes from rats classified as high responders (HR) and low responders (LR) to novelty were quantified using western blot. HR rats exhibited lower dorsal striatum levels of GRK2 and higher levels of GRK5 compared to LR rats. Finally, pharmacological inhibition of GRK2/3 using Cmpdp101 1.0 mg/kg ip reduced locomotion in a novel context only in HR rats. Our findings indicate that D2R stimulation modulates GRKs in the striatum, and that GRK activity influences the novelty-seeking, suggesting a role in drug addiction vulnerability.

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Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

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Program #/Poster #: PSTR295.02/HH6

Topic: H.08. Drugs of Abuse and Addiction

Support: BBRF NARSAD Young Investigator Award

Title: Neurodevelopmental Trajectories of Dopamine and Decision-Making Across Adolescence

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Abstract: Adolescence represents a precarious period of vulnerability wherein a variety of biological processes coalesce, during which behavioral and neurobiological traits are predictive of later neuropsychiatric diagnoses across a wide range of disease states. Among these changes, the dopaminergic pathways undergo rapid and extensive changes in both structure and function. Given dopamine's well-known role in reward and motivational behaviors, consistently demonstrated as differentially expressed within this period (e.g. elevated risk taking or resistance to extinction), mapping dopamine dynamics and differential plasticity across the lifespan, and as well as their relationship to decision-making processes within this window is imperative for understanding both normal and disrupted development. Yet, few studies exist delineating how dopamine's relationship to behavior in adolescence may differ from adulthood and how these changes precipitate vulnerability to maladaptive decision-making during this period. Thus, to treat neuropsychiatric disease effectively across the lifespan, it is imperative that we expand the present scope of study to mechanisms of dopaminergic encoding and plasticity in adolescence and map their relationship to enduring vulnerability to dysfunction in adulthood. Using cutting-edge combinations of *in vivo* and *ex vivo* electrochemical neurotransmitter detection and optical imaging methods with complex, longitudinal behavioral phenotyping, our newly established lab seeks to test the hypothesis that age-related changes in dopaminergic plasticity and encoding may represent a link between phenotypic differences in adult versus adolescent reward learning and motivation. Preliminary findings, shown here, examine the differences between adults and adolescents in their ability to obtain action-outcome contingencies for natural reinforcers and explore biological markers within the dopaminergic microcircuit that covary with both adolescent reward learning propensity and adult interactions with drugs of abuse. By identifying developmental shifts in dopamine function that shape learning and motivation, this work aims to uncover mechanistic links between early-life behavioral phenotypes and long-term susceptibility to neuropsychiatric illness, informing age-tailored prevention and intervention strategies.

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Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

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Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant DA 044331
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Title: Rostromedial tegmental stimulation induces behaviorally-specific avoidance for natural and drug rewards

Authors: *J. R. WATSON¹, E. CARLSON², P. J. VENTO³;

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³Psychology, Univ. of South Carolina, Columbia, SC

Abstract: Decades of research have revealed the complex ways in which reward seeking and associative learning are shaped by activation in mesocorticolimbic dopamine (DA) pathways, yet it is increasingly recognized that inhibitory signals to DA neurons likely play a similarly important role in avoidance learning and suppression of reward-seeking behaviors. While DA neurons receive inhibitory input from a variety of both local and distributed sources, the rostromedial tegmental nucleus (RMTg), a GABAergic midbrain region, densely innervates DA neurons in the ventral tegmental area (VTA), and inactivation of the RMTg-VTA pathway impairs punishment-induced suppression of reward seeking. To investigate how RMTg-VTA stimulation promotes avoidance behaviors, we tested rats in an operant food-seeking task where lever pressing for food pellets triggered contingent optogenetic RMTg-VTA stimulation. In this Single Reward Task, we observed robust real-time suppression of lever pressing, which intensified across repeated sessions, while responding quickly returned to baseline upon removal of stimulation, suggesting a strong, yet transient, effect on behavioral inhibition. To test the effects of RMTg-VTA stimulation under conditions of expanded behavioral choice, we next developed a Dual Reward Choice Task (DRCT) in which lever pressing for one of two distinct food rewards was repeatedly paired with RMTg-VTA stimulation. In contrast to the Single Reward Task, rats in the DRCT developed a robust and persistent avoidance specifically to the stimulation-paired reward option, with this aversion lasting weeks beyond stimulation. Notably, the reward-specific avoidance did not induce changes in motivational effort for either of the two reward options, as measured by progressive ratio, nor did it cause generalized changes in reward processing as measured by sucrose preference. To evaluate the effects of RMTg-VTA stimulation in modulating drug use and craving, we used these same optogenetic stimulation parameters contingently paired with cocaine self-administration (SA) in rats. Unlike the inhibitory influence on lever pressing for food, we found minimal effects of RMTg-VTA stimulation on ongoing cocaine SA. Yet, prior pairing of stimulation with cocaine use profoundly blocked cue-induced reinstatement of cocaine seeking when tested nearly two weeks later. Together, these findings highlight a remarkable role for the RMTg-VTA circuit in enduring forms of conditioned avoidance behavior. Ongoing experiments aim to reveal how RMTg-VTA stimulation alters midbrain DA and downstream mesocorticolimbic activity to mediate long-lasting changes in aversive learning.

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Poster

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Topic: H.08. Drugs of Abuse and Addiction

Title: Nucleus accumbens and basolateral amygdala neuronal ensembles mediate cocaine-seeking after abstinence

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Abstract: Despite the prevalence of relapse and overdose-related deaths in the human population, existing clinical interventions are lacking, making it essential to characterize the neurobiological mechanisms underlying cue-induced craving that drives drug seeking. Growing evidence suggests that sparse populations of cells, referred to as neuronal ensembles, play a critical role in encoding learned associations between drugs of abuse and drug-related cues. Here we explore the synergistic contribution of neuronal ensembles in the nucleus accumbens (NAc) and basolateral amygdala (BLA), key regions in mediating positive valence and reward, in encoding of cocaine-related memories. We assess the impact of memory perturbations such as extinction learning on the recruitment of NAc and BLA ensembles and characterize these cells' molecular properties with the ultimate goal of elucidating neuronal substrates that mediate drug responses, and how this is disrupted after extinction. We utilize *Arc*-CreER^{T2} mice and cocaine intravenous self-administration to tag *Arc*-ensemble neurons recruited during an initial drug-seeking test (test 1). To uncover the impact of extinction training on the recall of a drug-associated memory, mice underwent a second drug-seeking test one week later after extinction training. We demonstrate that extinction learning decreased the reactivation of *Arc*-ensembles in both the NAc and BLA that was not observed in homecage abstinent controls. These results uncover a mechanism by which extinction of drug-context associative memories attenuates drug responses via the modulation of ensemble reactivation. Furthermore, we posit that our findings contribute to understanding how extinction destabilizes drug-related memories to improve drug-seeking outcomes after forced abstinence. RNA- and ATAC-sequencing are underway to examine the molecular profile of the ensemble cells that encode cocaine-seeking and uncover how extinction changes their molecular properties to impact reactivation.

Disclosures: A.R. LaBanca: None. R. Futamura: None. Y. Yim: None. V. Kondev: None. T. Markovic: None. L.M. Holt: None. B.T. Kipp: None. M. Salery: None. E.J. Nestler: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.05/HH9

Topic: H.08. Drugs of Abuse and Addiction

Support: NSFC Grant 82071498

Title: Engrams and circuits crucial for forgetting of drug memory

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Abstract: Drug-associated memories are a principal driver of relapse in substance-use disorders. Conditioned-stimulus (CS) retrieval followed by extinction training—conducted within the reconsolidation window—robustly attenuates cue-evoked craving, yet the neural mechanisms that support this “retrieval-extinction” intervention remain obscure. Here we demonstrate that the infralimbic cortex (IL), but not the prelimbic cortex (PL), is essential for the protective effect of CS retrieval-extinction against methamphetamine (METH)-primed reinstatement. Chemogenetic silencing of IL neurons before retrieval-extinction abolished the reduction in drug seeking, whereas PL inhibition had no effect. Using activity-dependent viral tagging, we selectively labelled IL neurons active during retrieval-extinction (the “forgetting ensemble”) or during standard extinction (“extinction ensemble”). Optogenetic or chemogenetic activation of the forgetting ensemble—but not the extinction ensemble—mimicked the behavioral benefit of retrieval-extinction, indicating that this discrete neuronal population mediates memory suppression. E-SARE-driven labeling combined with c-Fos immunostaining, as well as one-photon miniscope calcium imaging, revealed that forgetting and extinction ensembles are non-overlapping and encode distinct behavioral states. Retrograde tracing showed that forgetting-ensemble neurons project preferentially to the midline thalamus (MT) rather than the basolateral amygdala. Consistent with this anatomy, in vivo electrophysiology and ex vivo recordings revealed enhanced excitability in IL→MT projections during retrieval-extinction. Critically, chemogenetic activation of the IL→MT pathway suppressed METH-primed reinstatement, recapitulating the effect of retrieval-extinction training. Together, these findings identify a previously unrecognized IL-to-midline-thalamus circuit that supports CS retrieval-extinction-induced “forgetting” of drug memories, providing mechanistic insight and a potential therapeutic target for preventing relapse.

Disclosures: Y. Xue: None. W. zheng: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.06/HH10

Topic: H.08. Drugs of Abuse and Addiction

Support: Rosetrees Trust MB2022\100002

Title: The differential reliance on multiple memory systems determines the habitual but not the compulsive nature of a coping response in male and female rats

Authors: *J. S. INNES, M. PUAUD, D. J. BELIN;
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Abstract: Convergent evidence supports a multiple systems organisation of mammalian memory. Individual differences in the tendency to rely on distinct, dissociable, neural systems subserving different types of learning/memory have been revealed using a spatial discrimination task, which can be solved using either procedural-like response or declarative-like place strategies. The individual tendency to rely on response learning has been associated with an increased proclivity to form habits and an increased vulnerability to develop compulsions. This hypothesis has not been tested in the context of instrumental coping behaviours, which can easily turn, in vulnerable individuals, into compulsive habits. In a longitudinal study, we investigated in 45 Sprague-Dawley rats (23 females) whether the individual tendency to rely on different memory systems predicted the habitual and/or compulsive nature of adjunctive drinking under a schedule-induced polydipsia procedure. Rats were first trained in a T-maze discrimination task, solvable using a hippocampus-dependent, allocentric place or a dorsal striatum-dependent, egocentric response strategy, which result in opposite turns at the choice point when the maze is rotated 180°. Rats were bilaterally implanted with cannulae above the anterior dorsolateral striatum (aDLS) and underwent 20 sessions of fixed time-scheduled food delivery, conditions that produce adjunctive polydipsic drinking in most individuals. However, some lose control over this behaviour and develop hyperdipsia, a compulsive coping response, which is predicated on the engagement of the aDLS dopamine-dependent habit system. Compulsive coping was assessed by the level of water consumption after 20 days and its resistance to quinine adulteration. The devolvement of control of the coping response, adaptive or compulsive, to the aDLS dopamine-dependent habit system was assessed by measuring the sensitivity of polydipsic/hyperdipsic drinking to intra-aDLS infusion of the dopamine receptor antagonist α -flupentixol. Rats identified as place and response learners did not differ with respect to the level nor the quinine-resistant nature of adjunctive drinking after 20 days of intermittent food delivery. However, adjunctive drinking was sensitive to aDLS dopamine receptor blockade, and thus reliant on the neural locus of instrumental habits, in response but not in place learners. These findings suggest that individuals characterised by the tendency to utilise procedural-like memory are more likely to develop habitual control of instrumental coping responses, but this does not necessarily increase the vulnerability to develop compulsions.

Disclosures: J.S. Innes: None. M. Puaud: None. D.J. Belin: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

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Program #/Poster #: PSTR295.07/HH11

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant GR040337
NIDA Grant P30 DA048736

Title: Medial prefrontal calcium dynamics during rat model of opiate withdrawal and subsequent psilocybin treatment

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Abstract: Opioid addiction continues to drive mortality and morbidity in the U.S (Kolodny et al., 2015), while currently available medication-assisted treatments for opioid use disorder are limited by their addictiveness and high relapse rates (Volkow et al., 2018). Psilocybin, a serotonergic agonist, has shown efficacy in treating depression, alcoholism, and nicotine addiction with a rapid-acting and non-addictive profile (Bogenschultz et al., 2015, Johnson et al., 2014, Carhart-Harris et al., 2017). The medial prefrontal cortex (mPFC), a key region in reward processing and drug-induced sensitization, undergoes long-lasting modifications as a result of opiate exposure (Jiang, et al., 2019; Qu et al., 2020). Notably, psilocybin alters mPFC morphology and activity, enhancing flexible behavior (Shao, et al., 20021; Morgan, et al., 2023). In this study, we used a rat model of opiate withdrawal, induced by administering twice-daily injections of morphine for 10 consecutive days, followed by abrupt cessation of dosing to induce spontaneous withdrawal. We combined this model with the analysis of basic behaviors (i.e. rearing, wet-dog shakes, head-twitch; DeepLabCut; SimBA), a novel object recognition task, and one-photon calcium imaging in the mPFC (Inscopix). We aimed to investigate whether psilocybin treatment restores the behavioral and neural alterations following opiate withdrawal to baseline conditions. Our preliminary findings show that opiate withdrawal results in hypofunction of the mPFC during task-relevant events which mirror the altered behavioral phenotypes and novel object discrimination impairments. Single-dose psilocybin treatment, compared to saline treatment, appears to restore calcium dynamics in the mPFC, along with improvements in novel object recognition. Interestingly, higher mPFC activation when interacting with the novel object, as opposed to the familiar object, correlated with a higher discrimination index across conditions. These results support the claim that mPFC activity reflects novelty-driven memory processes, which become impaired during opiate withdrawal.

Disclosures: V. Hones: None. S.J. Mizumori: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

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Program #/Poster #: PSTR295.08/HH12

Topic: H.08. Drugs of Abuse and Addiction

Support: Center for Neuroscience and Behavior, American University

Title: Navigating reward and aversion: the role of interoceptive signals in disambiguating outcomes to regulate consumption

Authors: *S. HUANG¹, A. L. RILEY²;
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Abstract: The ability to reduce drug use when faced with adverse consequences distinguishes voluntary (controlled) drug use from involuntary (compulsive) drug use, a hallmark of substance use disorder (SUD). Interoception, the processing of internal bodily signals, has been implicated in the emergence of SUD, yet its role in regulating drug use remains unclear. In a model of saccharin self-administration, we determined whether rats could use interoceptive cues to disambiguate when further intake would produce aversive effects and reduce consumption accordingly. Male and female rats consumed a saccharin solution for 25 sessions. “Master” rats received illness-inducing lithium chloride (LiCl) injections if consumption exceeded a threshold (50% of each rat’s initial baseline saccharin consumption). “Yoked” controls received noncontingent LiCl injections each time the paired master was injected. After 10 sessions, master rats learned to titrate saccharin intake. Between sessions 11-25, saccharin consumption fell within a 20% range (+/-) from the threshold in 64% of sessions for males and 54% of sessions for females. Of these sessions, consumption was titrated below the threshold in 65% and 61% of sessions for males and females, respectively, suggesting that rats were titrating to avoid adverse effects. In contrast, yoked rats showed no evidence of titration near the threshold, confirming that titration depended on interoceptive signals predictive of response-contingent outcomes rather than the frequency or schedule of LiCl injection. These findings indicate that interoceptive signals can come to predict post-ingestion aversive effects, thereby suppressing consumption. Importantly, this regulation occurred despite long delays between consumption and LiCl-induced aversive effects. This demonstration with saccharin reward provides a basis for interoceptive regulation of drug use. By disambiguating when reward will turn aversive, interoceptive drug signals could be key regulators in preventing excessive drug use.

Disclosures: S. Huang: None. A.L. Riley: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.09/HH13

Topic: H.08. Drugs of Abuse and Addiction

Title: Evaluating the abuse potential of gabapentin in combination with buprenorphine using conditioned place preference in mice

Authors: *L. A. MEDINA;
Psychological Sci., Northern Michigan Univ., Marquette, MI

Abstract: Opiate use disorder (OUD) has become a major public health issue, resulting in widespread, devastating effects on individuals, families, and society. Current opiate addiction treatment often includes medication-assisted treatment to prevent relapse. These medications include methadone or suboxone (buprenorphine). Recently, gabapentin (GBP) has been prescribed along with buprenorphine to prevent drug cravings and subsequent relapse. Although GBP has been presumed to have no addictive properties, recent literature reports a higher potential for individuals with OUD to abuse GBP. This research aims to determine the abuse potential of GBP using two doses (100 mg/kg and 300 mg/kg; i.p.) alone and in combination with buprenorphine (BUP) to assess the potential rewarding effects in CD-1 male mice. Additionally, this research aims to assess the effects of naltrexone, an opiate antagonist, on GBP alone and gabapentin combined with BUP. Morphine (7.5 mg/kg; i.p.) was used as a positive control in this study, while saline served as a negative control. Mice were randomly assigned to one of 13 groups consisting of 8-14 mice, including; saline/saline, morphine/saline, GBP (100 or 300 mg/kg)/saline, BUP/saline, buprenorphine+GBP 100 or 300 mg/kg/saline, naltrexone/saline, naltrexone+GBP 100 or 300/saline, naltrexone/BUP and lastly, naltrexone+GBP 100 or 300+BUP/saline. Drug reward was assessed using an unbiased, three-compartment conditioned place preference (CPP) paradigm. Each compartment contained visual and textural differences prominent enough for mice to determine a preferred compartment. After a baseline preference test, an 8-day conditioning period began; the compartment least preferred was paired with the drug, while the compartment most preferred was paired with saline. After the conditioning period, mice were again tested for their preferred context to assess if treatment affected preference. Data analysis revealed that both 100 mg/kg and 300 mg/kg of GBP are rewarding, indicating a significant risk for abuse. Naltrexone was effective in inhibiting rewarding effects in GBP 100 mg/kg but not in the 300 mg/kg. Lastly, naltrexone was not successful in reducing rewarding effects when combined with either dose of GBP and BUP. The results of this research indicate that GBP may have significant abuse potential alone and when combined with buprenorphine. The results also indicate that the rewarding effects GBP may be mediated by activity on opiate receptors.

Disclosures: L.A. Medina: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.10/HH14

Topic: H.08. Drugs of Abuse and Addiction

Support: UConn InCHIP
UConn IBACS

Title: Modeling Addictive Learning in Humans: Virtual Reality Conditioned Place Preferences to Nicotine and Alcohol Cues in Humans

Authors: *R. S. ASTUR¹, F. L. ZAKAS¹, S. M. SKLENARIK²;

¹Univ. of Connecticut, Storrs, CT; ²Psychological Sci., Central Connecticut State Univ., New Britain, CT

Abstract: Associative learning between substances and environmental cues plays a critical role in the maintenance and relapse of addictive behaviors. In two virtual reality (VR)-based conditioned place preference (CPP) studies, we investigated whether humans could develop a CPP for visually distinct VR rooms previously paired with either nicotine or alcohol cues. In Study 1 ($N = 44$), undergraduates with nicotine dependence received a 4 mg nicotine or placebo lozenge during conditioning sessions in VR rooms, followed by a reward-free test session. Participants showed significant explicit CPP (greater enjoyment ratings and preference for the nicotine-paired room, $p < .05$), but no significant implicit CPP (time spent in the nicotine room). However, implicit CPP was positively correlated with e-cigarette dependence ($r = .45$, $p = .002$), and sex differences emerged: females displayed implicit CPP, while males showed explicit CPP.

In Study 2 ($N = 95$), participants with varying levels of alcohol use were exposed to VR rooms with or without alcohol-related cues. In a cue-free test session, participants spent significantly more time in the alcohol-paired room ($p = .016$), demonstrating implicit CPP. Explicit CPP (enjoyment ratings) was not significant, but CPP was positively correlated with alcohol cravings ($p < .05$).

Together, these findings indicate that VR contexts previously associated with substances—either through primary (nicotine) or secondary (alcohol-related cues) reinforcement—can elicit conditioned approach behaviors in humans. Importantly, individual differences such as substance dependence severity and sex modulate the expression of CPP. This work highlights the translational potential of VR-based CPP paradigms for studying substance-related learning and offers a novel framework for future addiction research and intervention design.

Disclosures: R.S. Astur: None. F.L. Zakas: None. S.M. Sklenarik: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

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Topic: H.08. Drugs of Abuse and Addiction

Support: National Institute on Alcohol Abuse and Alcoholism (ZIAAA000550)
National Institute on Alcohol Abuse and Alcoholism (ZIAAA000130)

Title: Neurobiological craving signature expression in opioid use disorder

Authors: *F. N. VEENKER¹, P. MANZA^{1,3}, S. B. DEMIRAL¹, M. L. SCHWANDT¹, V. A. RAMCHANDANI¹, N. DIAZGRANADOS¹, G.-J. WANG¹, N. D. VOLKOW^{2,1};

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Abstract: Craving is a hallmark symptom of opioid use disorder (OUD) that can persist even with long-term treatment, complicating recovery and motivating return to drug use. Though craving is subjective and notoriously challenging to measure, brain imaging studies have revealed a ‘neurobiological craving signature’ (NCS) that offers the promise of objective quantification of craving and a possible target for therapeutic intervention. However, to our knowledge no one has tested if this marker is modifiable with OUD treatment and sensitive to drugs that modify addiction-related circuitry. We enrolled a cohort of OUD patients (n=48) at different stages of recovery (mean treatment duration = 3.64 +/- 4.64 years) who underwent functional magnetic resonance imaging (fMRI) during exposure to drug and food cues. Participants had fMRI scans on two separate days, once after a placebo and once after a dose of the dopamine-boosting stimulant medication methylphenidate (MP, 60 mg p.o.), since craving and anticipation of drug reward is associated with dopaminergic signaling. In the placebo condition, OUD patients exhibited higher NCS expression than healthy controls (t=2.94, p=0.004), which did not significantly correlate with treatment duration (r=0.21, p=0.19), indicating that elevated drug craving-related neural activity persists throughout various stages of treatment and recovery. Oral MP did not significantly modify NCS expression in OUD participants, but MP increased NCS expression in control participants, which might suggest that MP enhanced the salience of drug cues in these individuals. These findings provide insight into the enduring neurobiological markers of craving and provide potential treatment targets for OUD.

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Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.12/II1

Topic: H.08. Drugs of Abuse and Addiction

Title: Network Mapping Reveals Common Functional Network Alterations Across Substance and Behavioral Addictions Despite Structural Heterogeneity

Authors: *H. ZHENG;

Natl. Inst. on Drug Abuse, Baltimore, MD

Abstract: Background: Addiction involves dysfunction of multiple brain networks, including reward, habit, salience, executive, and memory networks. While unified theories suggest excessive reward network activity and impaired executive control contribute to addiction, it remains unclear whether these processes share common neural mechanisms across different

addiction types. Current meta-analyses are limited to group-level differences and fail to capture individual variability, making it challenging to develop reliable neuroimaging biomarkers for tracking disease severity and treatment response. This study aims to explore commonality and specificity in altered brain functional networks across addiction types, investigate how structural heterogeneity maps onto functional network alterations, and examine clinical symptoms. **Methods:** We analyzed neuroimaging data from substance use disorders (cocaine n=106, alcohol n=128, nicotine n=99, heroin n=32) and behavioral addictions (internet gaming disorder n=64), compared with healthy controls (n=957). The study employed normative modeling for structural analysis and activity network mapping (ANM) for functional connectivity to examine common and distinct patterns across addiction types. Self-network mapping (SNM) was used to identify patient-specific alterations and evaluate predictive capabilities for clinical outcomes. **Results:** Structural analysis using voxel-based morphometry revealed heterogeneous patterns of gray matter atrophy (10-20% overlap). Network mapping through ANM demonstrated that over 60% of structural alterations were functionally connected to common regions in the medial prefrontal cortex, and middle/posterior cingulate cortex. SNM revealed unique patterns in reward-related regions, particularly the orbitofrontal cortex and basal ganglia. Critically, SNM demonstrated superior predictive performance for addiction severity measures across both substance and behavioral addictions ($r=.161-.182$, $p<.05$) compared to ANM approaches. Furthermore, SNM showed enhanced predictive capability for TMS treatment response ($r=.379$, $p<.001$). **Conclusion:** Despite heterogeneous structural changes, functional network alterations converge on common circuits across addiction types, suggesting shared neural mechanisms. The superior predictive performance of SNM for both addiction severity and treatment response highlights the importance of individual-specific network patterns in clinical applications. These findings support developing both transdiagnostic therapeutic targets and personalized treatment approaches based on individual network profiles.

Disclosures: H. Zheng: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.13/II2

Topic: I.04. Executive Functions

Title: Cognitive Impairment and Mentalization in Polysubstance Dependents Undergoing Residential Rehabilitation

Authors: *A. C. DÍAZ¹, B. TELLEZ-ALANIS²;

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Abstract: Introduction. Polysubstance dependence—the use of multiple psychoactive substances—is a growing public health issue affecting both physical and mental health. This phenomenon is on the rise in several countries, including Mexico. Polysubstance dependence is

defined as the use of at least three different groups of substances without any single substance predominating over a 12-month period (Acosta et al., 2011). Cognitive impairment, the deterioration of mental functions, is commonly assessed using the MoCA test. Mentalization—the capacity to infer mental states from others' gaze—can be evaluated using the Eyes Test. Previous studies have reported that substance dependence is associated with cognitive impairment (Dean et al., 2013; Carolien et al., 2019) and reduced performance on the Eyes Test (Uhlmann et al., 2018). **Objective.** To evaluate alterations in cognitive functions—specifically attention, language, memory, abstraction, and mentalizing—in individuals with polysubstance dependence undergoing residential rehabilitation. **Method.** Forty-four individuals with polysubstance dependence (38 men, 6 women; aged 16-49) currently residing in a rehabilitation facility participated in the study. The instruments used were a structured questionnaire on substance use history, the MoCA test, adapted for the Mexican population (Aguilar-Navarro et al., 2017) and the Mexican version of the Eyes Test (30 items) (Téllez-Alanis et al., 2022). Participants and a family member provided informed consent. **Results.** Only seven participants had higher education. Half of the sample was of high socioeconomic status, and all had been in residential care for at least one week. Participants reported consuming at least six substances in the past year and experienced at least two hospitalizations. MoCA results showed 3 participants (6.8%) without cognitive impairment (>25), 34 (77.3%) with mild impairment (scores 19-25) and 7 (15.9%) with moderate impairment (≤ 18). The most affected cognitive domains were attention ($M = 3.4$), memory ($M = 2.0$) and abstraction (mean = 0.8). The mean accuracy on the Eyes Test was 63.3% ($SD = 11$), which is lower than that typically observed in university students. No significant correlations were found between MoCA and Eyes Test scores, nor with variables such as duration of substance use or length of stay in the facility. **Conclusions.** Polysubstance dependence is associated with cognitive impairment, particularly in attention, memory, abstraction, and mentalization. These findings have important implications for neuropsychology, particularly in designing interventions for the treatment and prevention of substance abuse.

Disclosures: A.C. Díaz: None. B. Tellez-Alanis: None.

Poster

PSTR295: Addiction: Mechanisms, Learning, and Memory

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR295.14/II3

Topic: H.08. Drugs of Abuse and Addiction

Title: Involvement of KCC2-dependent chloride extrusion in the use and effects of drugs of abuse: a scoping review

Authors: A. B. AMENDOLARA, S. A. SALAZAR, W. C. THOMPSON, H. A. WRIGHT, *A. J. PAYNE;

Biomed. Sci., Noorda Col. of Osteo. Med., Provo, UT

Abstract: The K⁺-Cl⁻ cotransporter KCC2 is a membrane protein specific to central nervous system neurons. Its primary function is to export Cl⁻ from the intracellular space and maintain a normal Cl⁻ gradient. Because this Cl⁻ gradient is essential for a vast majority of inhibitory neurotransmission, disruption of KCC2 expression or function results in varied and severe physiological consequences, up to and including death. Previous work has shown a connection between KCC2 and several neurological disorders, notably including epilepsy. Recent evidence also suggests a role of KCC2 in the function or effects of drugs of abuse. Here we sought to systematically gather and assess all available evidence published in the peer-reviewed literature relating to a physiological connection between KCC2 and any drug that may be considered abusable. Databases searched included Medline, SciFinder, Cochrane Library, Embase, Scopus, and PsycInfo. A total of 20,315 records were collected in the search. These results were deduplicated using Deduplick and then screened using ASReview and manual full text review, resulting in 112 relevant original research articles. We found that expression patterns of KCC2 were significantly impacted by various potentially abusable drugs, including morphine, remifentanyl, ethanol, diazepam, zolpidem, memantine, phenobarbital, pentylentetrazol, nicotine, cannabinoids, methamphetamine, prochlorperazine, TCB2, propofol, isoflurane, and sevoflurane, though some reports varied on the directionality of that expression change with specific drugs. Related KCC2-relevant changes in GABAergic signaling, eIPSC characteristics, and chloride transport caused by abusable drugs were observed, among other cellular effects. Additionally, alterations in KCC2 expression and activity impact alcohol and diazepam-induced changes in locomotor activity, morphine-induced conditioned place preference, and ethanol self-administration. Lastly, modulation of KCC2 activity was seen to improve opioid-induced hyperalgesia (OIH), and nicotine withdrawal-induced hyperalgesia may also be KCC2 dependent. These results suggest a strong connection between KCC2 and drugs of abuse. This work advances KCC2 as a potential drug target for treatment of substance use disorders and highlights areas of interest for future work.

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Poster

PSTR296: New Vistas in Preclinical Alcohol Research

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Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 1R01AA029129
NIH Grant P50AA10761

Title: Repeated ethanol disrupts the stability of sleep-wake states, state-specific in vivo activity of the hypothalamic GABA neurons and enhances their intrinsic excitability

Authors: L. A. LÓPEZ MUCIÑO¹, S. N. ROBERTS², J. J. WOODWARD³, *C. A. BLANCO-CENTURION⁴;

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Abstract: Repeated ethanol (EtOH) intoxication severely disturbs sleep. Yet, it is unknown if sleep disruptions correlate with changes in the activity of key sleep and/or arousal-promoting neurons like lateral hypothalamic (LH) GABA neurons. We hypothesized that EtOH disturbs sleep by changing the in vivo activity profiles of these neurons and their intrinsic excitability. We tested the hypothesis in 10 adult vGAT-Cre mice (5 ♂) who had repeated IP injections of three moderate doses of EtOH (1.2, 1.6, 2 g/kg [counterbalanced order], 15% v/v in PBS) for 15 days, two hours after light onset. Sleep-wake states and in vivo single-neuron Ca⁺⁺ imaging of LH GABAergic activity were recorded for five hours before (BL), during repeated intoxications (1-15 EtOH), and for one month into the withdrawal (WD). Additional recordings were made during nighttime (ZT21-23). After two weeks of repeated intoxications, sleep-wake states became significantly fragmented (BL=7.4±0.4 vs. 11-15 EtOH=9±0.4 transitions/10 min; P<0.05), worsening between WD days 6-10 (>11±0.5; P<0.001). Remarkably, instability persisted during nighttime (BL=5.9±0.5 vs. 6-10 WD=9.3±0.87; P<0.05), normalizing after week three of the WD, and happening earlier at nighttime. Also, we tracked the in vivo activity of 102 identifiable LH GABA neurons across BL, early WD, and late WD. At BL, most neurons exhibited a bimodal (AW/R=48) or unimodal R-max activity profile (n=30), while fewer were AW-max (n=9) or No-change (n=9). No-change neurons dramatically increased (n=35) in early WD, recovering their BL profiles by late WD (n=10). During early WD, the average Ca⁺⁺ peak area of the AW/R and R-max neurons significantly decreased in R (AW/R-max: BL=7.4±1.1 vs. early WD=3.2±0.39 AUP. P<0.001; R-max: BL=12±2.2 vs. early WD=6.6±1.4 AUP, P<0.001) or AW (AW/R-max: BL=7.9±1.3 vs. early WD=3.3±0.8 AUP. P<0.001), accounting for the loss of their activity profile. During late WD, average Ca⁺⁺ peaks reverted to BL levels. In a different set of vGAT-Cre mice (233 neurons, 26 mice, 13 ♂), slice electrophysiology studies showed that repeated EtOH intoxications increased the number of neurons exhibiting depolarization block in response to depolarizing currents (PBS= 81% vs. EtOH=96%). During early WD, the LH GABA neurons showed enhanced spiking (PBS= 76.7±5.7 vs. EtOH=106.3±7.06 total spikes, P<0.01) during current injections, accompanied by a lower rheobase (P<0.01). Altogether, our preliminary findings indicate repeated EtOH intoxications, even at moderate doses, chronically disrupt sleep-wake stability, cause loss of specificity in the activity profile of most LH GABA neurons, and heighten cell excitability.

Disclosures: L.A. López Muciño: None. S.N. Roberts: None. J.J. Woodward: None. C.A. Blanco-Centurion: None.

Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.02/II5

Topic: H.08. Drugs of Abuse and Addiction

Support: Emmanuel College Faculty Startup Funding

Title: Examining environmental enrichment effects on voluntary alcohol and caffeinated alcohol intake via the two-bottle choice paradigm in female mice

Authors: S. ALVAREZ, T. R. HARRISON, E. S. VIENNEAU, *E. J. CROFTON;
Emmanuel Col., Boston, MA

Abstract: Alcohol use disorder (AUD) is characterized by uncontrolled and impulsive use and dependency upon alcohol despite harmful social, physical, and mental effects. Although many humans drink alcohol, not everyone develops alcohol use disorder suggesting that individual differences may influence dependence on alcohol. Commonly alcoholic beverages include caffeine, another substance with risk of dependence, which may increase alcohol consumption. Globally, millions suffer from AUD yet current understanding of factors impacting differential susceptibility and the interaction between alcohol and caffeine is limited. The present study used female C57BL/6J mice that were housed in isolation or environmental enrichment for 30 days following weaning to investigate individual differences in alcohol intake and caffeinated alcohol intake. Enriched females were group-housed with access to plastic toys and isolated mice were single-housed. The elevated plus maze and open field test were conducted to evaluate anxiety-like behaviors and locomotor activity prior to alcohol and caffeine exposure. Open source software DeepLabCut developed by the Mathis laboratory was used to track behavior in the elevated plus maze and open field test. We found that environmental enrichment decreases locomotor activity in the open field test but there were no differences in anxiety-like behavior in the elevated plus maze. Mice were then exposed to the voluntary two-bottle choice paradigm with ethanol (20% v/v) alone and ethanol plus various doses of caffeine (0.01%, 0.02%, and 0.03%) following a M-W-F schedule. Two-bottles were held by and drinking data during each 24 hour drinking day were tracked using the Lick Instance Quantifier Home Cage Devices (LIQ-HD) developed by the Winder laboratory. We found that enriched females drank more alcohol compared to isolated females with no changes in water intake or preference for alcohol. One cohort showed a further increase in alcohol intake with 0.01% caffeine only in enriched females, however a second cohort showed no further alterations with any dose of caffeine. Molecular analyses including immunohistochemistry to investigate the underlying molecular mechanisms and the involvement of retinoic acid signaling in the individual differences in alcohol and caffeinated alcohol intake are ongoing. Preliminary analyses show potential alterations in retinoic acid signaling pathway proteins via immunohistochemistry. Overall, environmental enrichment in female mice results in individual differences in alcohol intake that may be altered by co-consumption of caffeine.

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Poster

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Program #/Poster #: PSTR296.03/II6

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH: R01AA025337 (JB, PI)

Title: Acute alcohol effects on cerebral-cerebellar network topology

Authors: *S. J. REWEY^{1,2}, L. A. BIESSENBERGER (DAVIS)², B. STENNETT-BLACKMON³, J. BOISSONEAULT²;

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Abstract: Acute alcohol intake has profound neurobehavioral effects, including cognitive and psychomotor impairments. The cerebellum, which is highly interconnected with numerous cortical and subcortical regions, plays a key role in these domains. Furthermore, its function is significantly disrupted by alcohol use. However, the impact of alcohol on the integration of the cerebellum with the rest of the brain is not well understood. Graph theory provides a means of characterizing alcohol effects on brain function by constructing a brain network or ‘graph’ composed of regions of interest (ROIs; nodes) and their connections to one another (edges). In this study, we examined how alcohol impacts the integration of the cerebellum with the rest of the brain. Healthy individuals (n=48, 21-45 yrs, 79% women) participated in two double-blinded sessions where they consumed a placebo drink (sugar-free lemon lime soda) or alcohol (0.08 g/dL target breath alcohol concentration). Approximately 30 minutes after beverage administration, resting state functional magnetic resonance imaging (rsfMRI) was obtained using a Siemens Prisma 3T scanner. Using the CONN toolbox, we used general linear models (GLM) to identify alcohol effects (vs. placebo) on common graph theory metrics in a network of 106 cerebral and 26 cerebellar and vermis regions of interest (ROI; $p_{FDR} < .05$). An ROI-ROI cost threshold (k) $> .25$ was used to define significant edges. Results showed that alcohol significantly reduced the degree (i.e., number of significant edges) in right cerebellar lobules 4 and 5 ($b = -9.54$, $t_{46} = -3.50$, $p_{FDR} = .023$). Alcohol also decreased eigenvector centrality, which reflects the influence of a node on the broader network, in right ($b = -.53$, $t_{46} = -4.46$, $p_{FDR} = .003$) and left ($b = -.40$, $t_{46} = -3.56$, $p_{FDR} = .014$) lobules 4 and 5, as well as right ($b = -.45$, $t_{46} = -3.27$, $p_{FDR} = .016$) and left ($b = -.39$, $t_{46} = -2.19$, $p_{FDR} = .04$) lobule 6. Interestingly, alcohol-induced change in degree and eigenvector centrality for left lobules 4 and 5 were correlated with participant ratings of subjective intoxication ($r = .32$, $p = .03$ and $r = .34$, $p = .02$, respectively). Together, findings suggest that alcohol significantly reduces the importance of aspects of the cerebellum to the broader brain network. Furthermore, this disruption appears to at least partially drive individual perceptions of intoxication.

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Poster

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Program #/Poster #: PSTR296.04/II7

Topic: H.08. Drugs of Abuse and Addiction

Support: NIAAA R01AA025380

Title: Behavioral and Neuronal Evidence of Hangover-Like States in Laboratory Rats

Authors: V. ZHYROV¹, M. J. SPINETTA², *J. LEASURE¹;

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Abstract: Binge drinking is highly prevalent and associated with a variety of adverse health outcomes, including hangover (HO). Symptoms of HO include malaise, gastrointestinal disturbance, fatigue, cognitive impairment, and changes in affect, among others. Importantly, HO is a meaningful predictor of the subsequent development of alcohol use disorder, making it an impactful marker for investigation. Yet despite its pervasiveness and the recent increase in scientific interest, HO remains poorly understood, as evidence is limited and relies primarily on human self-report. No pre-clinical model exists to study HO, therefore, the goal of this study is to develop a behavioral and neuronal profile of a HO-like state in laboratory animals. There is also no established treatment for HO, but emerging evidence suggests that physical exercise may lessen the severity of HO-related symptoms. In this study, we implemented a 5-week rat model of binge exposure (5 g/kg of ethanol via intragastric gavage once per week). Male and female Long-Evans rats were randomly assigned to Binge/Control and Exercise/Sedentary conditions. Exercised animals had access to running wheels for 3 hours 4d/w. Five-minute home cage videos were recorded the morning after each binge. Behavioral markers of the HO-like state (time spent immobile, time spent grooming, number of rears, and number of jumps on the food box) were quantified using an ethogram we developed. Our preliminary behavioral data show that the binged rats spent more time immobile and displayed fewer exploratory behaviors (rearing and jumping) relative to controls, consistent with an HO-like state. Ongoing analyses will determine whether HO-like behaviors diminish across 5 weeks, and whether the symptoms are lessened in exercised animals. The day after the final binge session, brains were collected and processed for detection of Δ FosB, a marker of sustained neuronal activity. We will quantify neuronal activity in the posterior insular cortex (pIC), anterior cingulate cortex (ACC), central amygdala (CA), and bed nucleus of the stria terminalis (BNST), regions previously implicated in interoceptive malaise, in order to determine if these areas are differentially active in rats that demonstrate HO-like symptoms. Developing a behavioral and neural profile of HO-like behaviors in rats will further rigorous investigation and enhance prevention and treatment efforts for alcohol use disorder.

Disclosures: V. Zhyrov: None. M.J. Spinetta: None. J. Leasure: None.

Poster

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Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 5R00AA028265-04

Title: Effects on forced swim stress on home cage ethanol consumption patterns

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Abstract: Repeated stress exposure is associated with the development of alcohol dependence. Previous rodent studies have demonstrated that repeated forced swim stress (FSS) facilitates an increase in ethanol consumption in dependent animals. However, the effect of repeated stress models, such as FSS, on the patterning of ethanol intake has yet to be determined. To investigate differences in drinking pattern resulting from repeated stress exposure, adult mice were subjected to a 10-day stress and drinking paradigm, in which half of the mice underwent daily FSS. One hour later, all mice were given time-limited access to 20% ethanol via their home cages equipped with capacitive lickometers. Here, it was hypothesized that FSS would be associated with a frontloading pattern of ethanol consumption assessed via the area under curve of the cumulative distributions. The ethanol dose consumed across the drinking period was also analyzed. No effect of stress, sex, or time (first two days versus last two days) on cumulative lick distribution area under the curve was observed. Ethanol dose consumed was significantly greater on the final two days of drinking when compared to the first two days of drinking, regardless of stress condition. However, when compared to non-stressed controls, stressed animals consumed less ethanol overall across the drinking period, contrasting the body of literature on this topic. Future analyses will investigate if behavioral data related to stress response (e.g., time spent floating) is related to drinking pattern or overall consumption. These results provide novel information about stress-related ethanol consumption patterns, a relevant behavioral measure for deepening the understanding of alcohol use.

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Poster

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Program #/Poster #: PSTR296.06/II9

Topic: H.08. Drugs of Abuse and Addiction

Support: Emmanuel College

Title: Ankyrin 3 reduction in male and female mice does not alter voluntary alcohol intake in two-bottle choice paradigm but may increase anxiety during alcohol withdrawal

Authors: *A. L. DRILLEN¹, S. ALVAREZ¹, E. J. CROFTON², M. P. LEUSSIS³;
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Fac. for Undergraduate Neurosci. (FUN), Boston, MA

Abstract: Ankyrin-G (*Ank3*) is a scaffolding protein crucial for neuronal excitability and implicated in stress susceptibility. Psychiatric comorbidities suggest *Ank3* may be a contributor to Alcohol Use Disorder (AUD) risk. However, its role in voluntary alcohol intake and withdrawal-related anxiety remains unclear. To investigate the behavioral impact of *Ank3* disruption, male and female *Ank3* heterozygous (HET) knockout and wild-type (WT) mice were assessed in a two-bottle choice voluntary alcohol consumption paradigm. The two-bottle choice paradigm is a 28-day long experiment where the mice are exposed to 20% volume/volume alcohol on alternating days, resulting in a voluntary intermittent pattern. Following exposure, mice were evaluated for anxiety-like behavior during withdrawal using the elevated plus maze (EPM). We found *Ank3* HET mice did not differ significantly from WT mice in alcohol intake or preference, indicating that *Ank3* disruption does not directly alter voluntary alcohol consumption. However, a significant main effect of sex was observed, with females consuming more alcohol than males across days. During alcohol withdrawal, no significant genotype differences were found in EPM metrics; however, *Ank3* HET mice did not display their previously reported risk-taking phenotype, suggesting that withdrawal may increase anxiety-like behaviors in the *Ank3* HET mice. Sex differences were also found in EPM, showing females exhibited more closed arm entries and fewer rearing behaviors, particularly among HET mice. While *Ank3* disruption did not increase alcohol consumption, it may modulate anxiety-like or exploratory behaviors during alcohol withdrawal. These findings highlight the importance of incorporating sex as a biological variable and suggest a potential role for *Ank3* in mediating behavioral adaptation to internal distress states such as withdrawal. Future studies should explore the neurobiological mechanisms underlying these effects, including stress circuitry and hormonal regulation.

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Poster

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Topic: H.08. Drugs of Abuse and Addiction

Support: R01AA023288
F31AA031433
T32AA007474

Title: Functional and molecular adaptations in the thalamic nucleus reuniens in alcohol dependent male and female mice

Authors: *K. L. LINDQUIST¹, S. BERTO², J. A. RINKER³, P. J. MULHOLLAND⁴;
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Abstract: Chronic alcohol exposure can cause cognitive deficits that interfere with alcohol abstinence and increase the odds of relapse. The nucleus reuniens (RE) is a ventral midline thalamic nucleus vital for cognitive function that remains understudied within the alcohol use disorder field. I used the chronic intermittent ethanol (CIE) model in adult male and female C57BL/6J mice to test for spatial working memory deficits using a T-maze-based spontaneous delayed alternation task (n=10-13/group). CIE exposed males and females performed significantly worse on the task in comparison to controls. To measure physiological changes that could potentially be driving the cognitive impairment, I used slice electrophysiology (n=5-7/group) to measure evoked action potential firing and synaptic transmission in the RE. CIE produced divergent effects on intrinsic excitability. RE neurons from CIE-exposed male mice displayed reduced AP firing, many of which entered into depolarization block. In contrast, RE neurons from females displayed increased evoked firing. Because control male action potential firing was significantly higher than control female action potential firing, we interpreted entrance into depolarization block as a potential marker of increased excitability. Although there was no change in spontaneous glutamatergic postsynaptic currents, CIE exposure increased the amplitude of spontaneous inhibitory postsynaptic currents in both sexes. Based on increased intrinsic excitability, I wanted to determine if inhibition of the RE could reverse CIE induced working memory deficits. Male mice (n=6-8) were surgically infused with a Gi-DREADD into the RE prior to CIE exposure. Mice underwent baseline T-maze testing before CIE and were retested following completion of CIE exposure. In a counterbalanced fashion, mice were administered 0.1 mg/kg deschloroclozapine or vehicle administration 20 minutes before T-maze testing. Preliminary data suggest that inhibiting the RE improved working memory performance in CIE exposed male mice. To determine molecular alterations that could drive CIE-induced behavioral and functional adaptations, we performed bulk RNA-seq on the RE (n=6-7 mice/group). Weighted Gene Correlated Network Analysis (WGCNA) was employed to determine networks of genes impacted by CIE. WGCNA identified 17 distinct gene modules with differential expression based on either sex or CIE treatment. In summary, this study demonstrated molecular and functional adaptations in the RE in alcohol dependent mice, indicating that these changes contribute to chronic alcohol-induced cognitive deficits.

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Poster

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Schimmel Family Chair

Title: Early life voluntary alcohol consumption impairs cognitive and affective functions in Marchigian Sardinian alcohol preferring and Wistar rats

Authors: *M. PALMISANO¹, A. E. GONZALEZ¹, C. ST. ONGE¹, H. MRIZAK^{1,2}, I. MUNOZ GIL¹, O. SHARLAI¹, J. BENEDICT¹, B. CRUZ¹, R. CICCOCIOPPO², M. ROBERTO¹;

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Abstract: Alcohol use disorder (AUD) is a chronic, relapsing neuropsychiatric disease characterized by compulsive alcohol use despite harmful consequences. Alarming, alcohol use is increasing among young individuals, a developmental period marked by ongoing brain development and heightened neuroplasticity. Accumulating evidence links young adult drinking to long-term impairments in cognitive function, emotional regulation, and increased vulnerability to neurodegeneration later in life. Understanding the cognitive and affective consequences of chronic alcohol exposure in young individuals, particularly in genetically vulnerable populations, is critical for developing targeted interventions. Thus, our study employed a preclinical AUD model: the Marchigian Sardinian alcohol preferring (msP) rats, genetically prone to high alcohol intake, relapse, and an anxious-depressive-like phenotype. We aimed to compare effects of prolonged alcohol consumption on behavior in msP and non-alcohol preferring Wistar rats. To model voluntary alcohol intake, we used the two-bottle choice paradigm over a five-week period, where animals had 24 hr access to two bottles (one containing water, and another containing alcohol 20%). After alcohol exposure, we evaluated memory, anxiety, and compulsive-like behaviors using a battery of behavioral tests, including the novel object recognition (NOR) (24hr), Y-maze, light and dark box, and marble burying test. Consistent with previous works, msP rats reliably consumed more alcohol than Wistars, confirming their heightened genetic vulnerability to alcohol. Chronic alcohol exposure selectively impaired long-term NOR memory in msPs, but not in Wistars, compared to their naïve counterparts, suggesting that the msP strain is particularly vulnerable to alcohol-induced memory impairment. Interestingly, Wistars showed deficits in short-term memory compared to msPs in the Y-maze task. While there was no effect of strain or sex, both msPs and Wistars exhibited compulsive-like behavior in the marble burying task, irrespective of alcohol vulnerability. Moreover, in the light and dark box task, msP rats spent less time and made fewer transitions into the light compartment compared to Wistars, indicating a heightened anxiety phenotype. Taken together our data suggest that chronic

voluntary alcohol consumption during early life can impair cognitive function in a strain-specific manner. These results highlight how genetic predisposition and early alcohol exposure interact to shape cognitive and emotional outcomes, providing valuable insights into the neurobehavioral mechanisms underlying AUD.

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Poster

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Program #/Poster #: PSTR296.09/II12

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 5T32GM144891-02

Title: Ethanol Consumption Increases Delay Discounting in Male Wistar Rats

Authors: ***A. REMES**, C. C. LAPISH;
Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Purpose. Impairments in decision-making have been shown to be a risk factor for and consequence of heavy ethanol consumption. Prior work has established that rodent lines with heritable risk for excessive drinking tend to exhibit high impulsivity, therefore establishing it as a phenotypic trait. However, demonstrating that increases in impulsive states can be elicited in rodent models following voluntary ethanol consumption is still forthcoming. The goal of the current work is to determine if voluntary ethanol consumption leads to an increase in impulsivity in outbred animals. **Methods.** Male (n=29) and female (n=25) Wistar rats, were put through 4 weeks of an intermittent access protocol (IAP). Next, they received 2 weeks of a restricted access protocol (RAP) where they were transferred to a new environment and ethanol was available for 1-hour. Finally, they were trained in a delay discounting task where impulsivity was measured by calculating preference for a larger delayed reward vs a smaller immediate reward. The delayed reward times were increased after each week and impulsivity was quantified by fitting a hyperbolic function to the discounting curve of each animal. **Results.** Ethanol drinking behavior was heterogeneous in this group of rats. Animals were sorted into one of two groups based on their average ethanol consumption. The “drinkers” consumed greater than 1.5 g/kg 15% EtOH in IAP and 0.4 g/kg 10% EtOH in RAP. The rats that failed to reach these criteria were classified as “nondrinkers.” The two groups exhibited phenotypic stability across both drinking tasks with the drinkers consistently consuming more than the nondrinkers. In the delay discounting task, increases in the selection of the immediate reward was observed in all animals by systematically increasing delays. Male, but not female, rats exhibited an increased discounting delay compared to the male water-consuming control rats. **Conclusion.** Ethanol consumption may induce

changes in impulsive states in a sex-dependent manner. A potential caveat of this study is that impulsivity was not assessed prior to alcohol consumption, thereby making it difficult to disambiguate state versus trait impulsivity. Notwithstanding, this data deepens our understanding of how impulsive states may be generated by alcohol consumption and could facilitate novel strategies for long-term abstinence and recovery.

Disclosures: A. Remes: None. C.C. Lapish: None.

Poster

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Program #/Poster #: PSTR296.10/II13

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant R01AA006059

Title: Orexin receptor 1 antagonist reduces motivated drinking and improves sleep/wake patterns after adolescent ethanol consumption

Authors: C. CUETO¹, K. GUERRERO LEON², A. MORA⁴, L. GONZALEZ⁵, M. R. GONZALES⁶, S. GOMEZ³, *L. R. AMODEO⁷, C. L. EHLERS⁸;

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Abstract: Disturbances in sleep and activity rhythms are significant health risks associated with alcohol use during adolescence. Many investigators support the theory of a reciprocal relationship between disrupted circadian rhythms, sleep patterns, and alcohol usage. However, in human studies, it is difficult to disentangle other factors when determining what is causal in the relationship between substance use and sleep/activity disruptions. The orexin system is involved in the homeostasis of sleep/wake cycles, feeding, drinking, and motivation. Alcohol consumption has been shown to increase orexin activity, leading to higher motivational and drug-seeking behavior for alcohol use and negatively impacting sleep. Ideally, inhibiting the orexin system could be a potential target for reducing consumption and motivation for alcohol use in adulthood, as well as improving alcohol-related sleep issues. Previous work has shown that SB-33487, an orexin-1 receptor antagonist, reduces ethanol-related behavior, but there is a lack of research regarding sleep. For this study, we investigated the effects of SB-33487 on adult drinking behaviors and concurrent rest-wake activity after adolescent alcohol consumption. To this end, adolescent female and male Wistar rats will receive 24-hour access to a 2-bottle choice paradigm of 20% ethanol and water for 5 weeks during adolescence. Rest/wake activity measures will be assessed at the end of adolescence, young adulthood, and adulthood. At the start of adulthood,

rats were assessed for motivated drinking behavior using fixed-ratio and progressive ratio tasks for 20% ethanol access. SB-33487 (30 or 60 mg/kg) or control was administered before progressive ratio testing and rest-wake activity monitoring. High ethanol-consuming adolescent rats continue to consume more in adulthood and display more motivated drinking. Orexin 1 antagonist, SB-33487 dose-dependently reduced motivated drinking and improve rest-wake activity patterns.

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Poster

PSTR296: New Vistas in Preclinical Alcohol Reserch

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Program #/Poster #: PSTR296.11/II14

Topic: H.08. Drugs of Abuse and Addiction

Support: Butler University Holcomb Awards
Indiana Academy of Sciences

Title: Male mice exposed to intermittent caffeine-mixed alcohol during adolescence consume higher quantities of alcohol as adults

Authors: L. FIELDS, L. ROZEBOOM, *J. N. BERRY;
Psychology; Neurosci., Butler Univ., Indianapolis, IN

Abstract: In recent years, the notion that caffeine reduces the depressant effects of alcohol has spread, leading to a drastic increase in alcohol and caffeine co-consumption as well as increased negative consequences, including higher risk for alcohol dependence. Furthermore, young people are often the target audience for the marketing of highly caffeinated beverages, which often translates into this population being especially susceptible to the negative outcomes of such co-consumption. For this study, C57BL/6J adolescent mice (n=56, 28 males and 28 females) were acquired at 4 weeks and then provided intermittent access (24 hours on followed by 24 hours off; IA) to the experimental bottle for a duration of 28 days. Experimental bottles contained one of four liquids- either caffeine (0.015% w/v), alcohol (10% v/v EtOH), water, or caffeine+alcohol at the previous concentrations. Withdrawal symptoms were assessed approximately 6 hours after removal of the experimental bottles on day 28 using an open field test (OFT). Following the OFT during acute withdrawal, mice were returned to their cages and allowed to age for 10 additional days, then given a 24 hour challenge period where all mice received access to 10% alcohol. Male, but not female, adolescent mice consumed significantly more alcohol when combined with caffeine compared to male adolescent mice that had access to alcohol alone. The OFT revealed that mice exposed to IA caffeine+alcohol traveled significantly less compared to their control (water exposed) counterparts. During the 24 hour challenge period in early adulthood, male mice exposed to caffeine+alcohol during adolescence consumed significantly more 10% alcohol

compared to male mice given water during adolescence. Findings from this study were congruent with previous data from past experiments, showing that exposure to combined alcohol and caffeine resulted in significantly increased consumption in adolescent male, but not female, mice. The consumption of caffeine was significantly increased in both male and female adolescent mice when combined with alcohol as opposed to the either drug alone. Similarly, locomotion was significantly reduced in the acute withdrawal period from combined alcohol and caffeine, but not individually. These results suggest that intermittent caffeine-mixed alcohol during adolescence may alter later alcohol intake in male mice.

Disclosures: L. Fields: None. L. rozeboom: None. J.N. Berry: None.

Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.12/JJ1

Topic: H.08. Drugs of Abuse and Addiction

Support: R01AA016959
T32AA007471

Title: The impact of binge alcohol exposure on learning and memory in adult female sprague-dawley rats

Authors: *S. A. COLLINS¹, N. N. NAWARAWONG², K. NIXON³;
²Pharmacol. and Toxicology, ³Pharmacol. & Toxicology, ¹The Univ. of Texas at Austin, Austin, TX

Abstract: Excessive alcohol consumption, a defining feature of an alcohol use disorder, can result in neurodegeneration and behavioral impairments, which may recover with abstinence. The dentate gyrus region of the hippocampus, an area that plays an important role in learning and memory, is highly susceptible to the damaging effects of alcohol. This region also contains neural stem cells that contribute to alcohol-induced damage and recovery. Previously, we observed impaired acquisition of the Morris Water Maze (MWM), a hippocampal-dependent learning and memory task, in binge ethanol-treated male rats after 1 week of abstinence. However, recovery and overshoot of better performance were observed by 7 weeks post-ethanol, an effect paralleled by a reactive increase in adult neurogenesis. While these effects were demonstrated in males, it is not known whether similar effects occur in females. Thus, same-aged (PND76-86), female Sprague-Dawley rats were subjected to a binge ethanol paradigm, where rats were gavaged with an ethanol (25% w/v) or isocaloric control diet in vanilla Ensure Plus® every 8 hours for 4 days. The dose of ethanol was determined by their intoxication behavior, resulting in a mean dose of 10.5±0.2 g/kg/day and blood ethanol concentrations of 390.3±11.4 mg/dL. Beginning 1 week after the last dose, one group of rats underwent training in the MWM for 4 days (T7-T10), followed by a probe test on the 5th day (T11). A second group

completed the same training (T49-T52) and probe test (T53), beginning 7 weeks after the last dose. All rats were sacrificed via transcardial perfusion 90 minutes after the probe test. While latency to find the platform improved over the 4-day training period for both groups ($p < 0.05$), binge alcohol-exposed females took significantly longer to find the platform compared with their control counterparts after 1 week ($p < 0.05$), but not 7 weeks. A similar improvement was observed during the training period for both groups when it came to distance to the platform, however, females exposed to alcohol traveled significantly longer distances at 1 week ($p < 0.05$), but not 7 weeks. Despite these differences in training, there were no observed differences in recall of the MWM task during the probe trial for either time point. These findings suggest that binge-like ethanol exposure impairs learning performance in female rats 1 week into abstinence, but appears to recover, but not overshoot, by 7 weeks. These data align with less robust reactive adult neurogenesis in female ventral dentate gyrus versus males. Ongoing studies are investigating the neuronal ensembles underlying these behaviors and how they may differ between groups.

Disclosures: S.A. Collins: None. N.N. Nawarawong: None. K. Nixon: None.

Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.13/JJ2

Topic: G.03. Stress and the Brain

Support: NIH Grant AA030256
NIH Grant MH104603

Title: Low Activity Monoamine Oxidase A and Early Life Stress Alter Binge-Like Alcohol Consumption and Aggression in a Sex Dependent Manner

Authors: *N. M. RUSSELL¹, D. CARRIZALES SAUCEDA¹, C. BABCOCK¹, Y. VO², P. KAHANAK¹, C. MOORE², T. LIME³, M. BORTOLATO⁴, R. A. MANGIERI¹;

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Abstract: In humans, low activity of the enzyme monoamine oxidase A (MAOA), caused by a gene polymorphism, is associated with violent offences, substance use, and antisocial behavior. These psychiatric effects are often catalyzed by adverse early life environments. We translated this relationship to mice with a hypomorphic MAOA mouse line (MAOA-L) and performed an early-life stress (ELS) paradigm (3 hours maternal separation and flank needle poke during postnatal day [PND] 3-7). We expected mice predisposed to higher rates of aggression (ELS-MAOA-L) would consume more ethanol. MAOA-L and wildtype (WT) mice underwent ELS or a no stress (NS) period before screening for binge-like alcohol consumption or aggression. To

explore binge-like alcohol consumption, the drinking-in-the-dark (DID) paradigm was performed in early adult mice (PND 56-73). For the DID paradigm, mice were given four-hours access to a drinking solution, beginning three hours into their dark cycle, four days/week, for three weeks (one week each of 20% ethanol, water, and saccharin). Our findings revealed sex differences in the DID paradigm. Male mice showed no effect of genotype or stress on any measure of alcohol, water, or saccharin consumption. Male ELS-MAOA-L mice did, however, have higher BEC's compared to NS-MAOA-L mice ($p=0.039$). In female mice, average alcohol consumption per session showed a trend towards a main effect of genotype ($p = 0.051$), that was driven by ELS-MAOA-L mice, which drank more alcohol overall compared to ELS-WT ($p=0.053$), as well as NS-MAOA-L ($p=0.054$). There was a main effect of ELS ($p=0.0041$), and a simple effect of ELS in MAOA-L mice ($p=0.0064$), to increase alcohol drinking in the first half of the session. By the final ethanol session, ELS-MAOA-L females drank more than NS and WT controls ($p's<0.03$), but BECs did not significantly differ. During the resident intruder aggression screening, male ELS-MAOA-L mice showed a shorter attack latency compared with all groups (NS-WT [$p=0.003$], ELS-WT [$p=0.019$], and NS-MAOA-L [$p=0.011$]). A small pilot study using two weeks of ELS did not find any group differences in aggressive behavior in female mice during the resident intruder test. In summary, male ELS-MAOA-L mice displayed no differences in alcohol consumption but had higher BEC's, suggesting that they achieve greater level of intoxication with the same amount of consumption. Male ELS-MAOA-L mice displayed signs of increased aggression. Female ELS-MAOA-L exhibited greater consumption and frontloading of alcohol, but no signs of increased aggression. Thus, low-activity MAOA combined with ELS produced different phenotypes in each sex relating to aggression and alcohol misuse.

Disclosures: N.M. Russell: None. D. Carrizales Saucedo: None. C. Babcock: None. Y. Vo: None. P. Kahanek: None. C. Moore: None. T. Lime: None. M. Bortolato: None. R.A. Mangieri: None.

Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.14/JJ3

Topic: G.03. Stress and the Brain

Support: NIH Grant AA030256

Title: Investigating Long-Access Ethanol Drinking in a Mouse Model of Low Activity Monoamine Oxidase A and Early Life Stress

Authors: *C. BABCOCK¹, N. RUSSELL¹, D. CARRIZALES SAUCEDA¹, Y. VO², M. BORTOLATO⁴, R. A. MANGIERI³;

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Abstract: Genetic polymorphism leading to low expression of monoamine oxidase A (MAOA) is present in individuals with a history of alcohol or substance use disorders at higher rates than the general population. When individuals with low activity MAOA gene variants experience childhood maltreatment, the relationship with substance use disorders becomes more robust, suggesting a gene-by-environment interaction. To study this interaction, we used a genetically modified line of mice containing a low-activity MAOA mutation (MAOA-L). Male and female MAOA-L and wild-type (WT) mice were assigned to either a non-stress (NS) or early-life stress (ELS) condition. At post-natal day (PND) 1, ELS mice were subjected to 30 minutes of maternal separation and received a toe tattoo for identification. At PND 2, they were subjected to 1 hour of maternal separation. At PND 3-7, they were subjected to a total of 3 hours of daily maternal separation and a daily flank needle poke to model parental neglect and physical trauma. Later, at approximately PND 56, mice began ethanol intermittent access sessions in which they were given 24-hour access to a choice between water and an ethanol solution (3, 6, or 10% for week 1, 20% ethanol for weeks 2-4), 3 sessions per week, separated by 24 or 48 hours. Dose of ethanol consumed (g/kg) and preference for the ethanol solution (ethanol solution intake/(ethanol + water intake)) were analyzed using 3-way repeated measures ANOVA for the first 4 sessions (when ethanol concentrations increased between each session). The average intake and preference over all 20% sessions were analyzed by 2-way ANOVA. Male and female data were analyzed separately. For males, there were no significant genotype or stress main effects on intake or preference across the first 4 sessions, but there was a genotype-by-stress interaction ($F_{1,29} = 4.4$, $p=0.046$), in which ELS increased intake by WT and not MAOA-L male mice. There were no main or interaction effects on average intake or preference during 20% drinking sessions. For females, intake did not differ for any period of analysis. Preference during the first 4 sessions was significantly lower in MAOA-L mice overall (main effect of genotype, $F_{1,34} = 4.3$, $p=0.046$), but there were no effects on average preference during 20% sessions. These results indicate that the chronic 2-bottle choice drinking model used in this study does not reproduce the potentiation of alcohol intake by early-life stress reported in the human literature and our other work using a model of binge-like ethanol drinking (see companion abstract by Russell et al.). We are now conducting further investigation of ethanol drinking behavior in an operant self-administration paradigm.

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Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.15/JJ4

Topic: H.08. Drugs of Abuse and Addiction

Support: R01 AA029386
T32 AA007471

Title: Consuming alcohol during withdrawal reduces withdrawal symptom severity but does not influence relapse risk in rats

Authors: *K. BANKSTON¹, M. OLVERA¹, M. H. MONFILS^{1,2}, H. J. LEE¹;

¹Dept. of Psychology, ²Inst. for Neurosci., The Univ. of Texas at Austin, Austin, TX

Abstract: Many individuals with Alcohol Use Disorder (AUD) become physically dependent on alcohol, which entails experiencing withdrawal symptoms in its absence. It is thought that physical dependence contributes to the maintenance of AUD, as individuals will drink alcohol to avoid aversive symptoms of withdrawal— a type of learning known as negative reinforcement. Most rodent models of AUD induce physical dependence via chronic intermittent ethanol (CIE) vapor exposure, but do not account for the negative reinforcement learning that is present in human cases. As such, it is poorly understood whether this negative reinforcement learning affects relapse following cue exposure (extinction) therapy. The aim of this experiment was to determine if negative reinforcement learning influences relapse risk in rats. Long-Evans male rats were first trained to drink 15% unsweetened alcohol in their home cages via an intermittent access 2-bottle choice (IA2BC) paradigm. Then, they underwent Pavlovian conditioning in which a light cue was paired with a sipper containing 15% alcohol. To induce physical dependence, rats were exposed to CIE vapor (14 hrs on : 10 hrs off daily) 5 days a week for 4 weeks. To model negative reinforcement, rats were given access to a bottle containing 15% alcohol for 2 hrs during withdrawal from CIE vapor (i.e., 6-8 hrs post-vapor cessation). As a control, half of the subjects were exposed to air instead of vapor and half of each vapor/air group were not given alcohol during the withdrawal period. Relapse was measured as alcohol consumption in the home cage and cue-reactivity after extinction training in which a light cue was presented with an empty sipper. We found that drinking alcohol during the withdrawal period did not alter future alcohol consumption or cue reactivity. Interestingly, rats exposed to vapor consumed less alcohol during the withdrawal period compared to rats exposed to air. Nevertheless, drinking alcohol during withdrawal decreased withdrawal symptom severity in rats exposed to vapor. These findings indicate that although consuming alcohol reduced withdrawal symptoms, it did not influence future alcohol consumption or cue reactivity. Our results suggest that withdrawal severity during the cycle of physical dependence to alcohol does not influence relapse-like behavior induced by alcohol cues. We plan to further examine this effect in female rats.

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Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.16/JJ5

Topic: H.08. Drugs of Abuse and Addiction

Support: R01 R01AA029386
T32 AA007471

Title: Dose-dependent effects of Levonorgestrel on Alcohol Consumption and Relapse Behavior in Female Rats

Authors: *P. ANTONACCI^{1,2}, A. VASQUEZ², P. MENDOZA³, R. GONZALES⁴, M. H. MONFILS⁵, H. J. LEE²;

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Abstract: Women are more sensitive to alcohol and at greater risk for alcohol use disorder (AUD), with ovarian hormones such as estrogen and progesterone linked to increased alcohol intake. In the U.S., about 65% of women use contraceptives, most commonly oral hormonal contraceptives (the pill); yet, little is known about their interaction with alcohol. This study investigated the dose-dependent effects of Levonorgestrel (LNG) on alcohol consumption and relapse-related behaviors. LNG is a progestogen-based contraceptive that functions to prevent ovulation and reduce circulating estrogen and progesterone levels. In the present study, naturally cycling female Long-Evans rats were trained to consume 15% unsweetened alcohol using a home-cage drinking procedure. Rats were then assigned to one of three treatment groups receiving daily oral administration of either 2mg LNG, 500ug LNG, or No LNG (control) distributed in a vanilla cookie paste for the remainder of the experiment. Following exposure to alcohol, rats underwent Pavlovian conditioning in which a 20-second light cue was paired with access to 15% alcohol for 10 seconds via a sipper. After 12 days of conditioning sessions, rats underwent extinction training in which the light cue was presented without alcohol (i.e., empty sipper). Subsequently, they were tested for long-term extinction memory and reinstatement of alcohol-seeking behavior. Finally, rats were re-conditioned and blood samples were collected to measure blood alcohol concentrations (BAC). Our results showed that rats in the high-dose LNG group (2mg LNG) consumed more alcohol during conditioning compared to the low-dose (500 ug) and control groups. However, this group also showed the least relapse-like behavior in which their contact with the empty sipper remained low during the long-term extinction memory and reinstatement testing. These findings highlight a complex, dose-dependent relationship between hormonal contraceptive exposure and alcohol-seeking behaviors. Our work has important implications for understanding how contraceptives modulate learning and relapse mechanisms relevant to AUD and may inform prevention and treatment strategies for women.

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Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.17/JJ6

Topic: H.08. Drugs of Abuse and Addiction

Support: 7/8 NADIA U01
Developmental Exposure Alcohol Research Center

Title: Targeting p75 Neurotrophin Receptor with LM11A-31 Mitigates Alzheimer's-Like Deficits Caused by Adolescent Alcohol Exposure in Rats

Authors: *E. L. KERR¹, L. M. SAVAGE²;

¹Psychology, Binghamton Univ., Vestal, NY; ²Psychology-Behavioral Neurosci., SUNY - Binghamton Univ. Behavioral Neurosci., Binghamton, NY

Abstract: Excessive alcohol use during adolescence produces long lasting changes on brain structure and function that appear to increase risk of pathological aging and Alzheimer's Disease (AD). While it has been shown that adolescent intermittent ethanol (AIE) exposure may exacerbate AD-related pathology, potential therapeutics remain underexplored. This study investigates the effect of LM11A-31, a small molecule modulator of the p75 neurotrophin receptor, treatment on mitigating AD-related pathology in TgF344-AD rats following adolescent intermittent ethanol (AIE). Male and female TgF344-AD and Wild-type (WT) rats underwent AIE treatment (5 g/kg) via intragastric gavage from postnatal day (P) 28 to 59. From 5 to 8 months of age rats received daily oral treatment of either LM11A-31 or vehicle solution. Following the end of LM11A-31 treatment, behavioral assessments of anxiety-like behavior, recognition memory, and spatial learning and memory were conducted. Rats were euthanized following completion of behavioral tests via perfusion and brain were collected for immunohistochemical analysis of neurogenesis, amyloid-beta plaques, and phosphorylated tau. Preliminary results indicate AIE exacerbated cognitive and behavioral impairments, which were rescued by LM11A-31. We will further determine whether LM11A-31 treatment rescues the negative effects of AIE on AD-related pathology (reduced neurogenesis, increased amyloid beta deposits and phosphorylated tau) in TgF344-AD rats. These findings suggest that AIE accelerates AD progression and targeting p75 neurotrophin receptor signaling via LM11A-31 has therapeutic potential.

Disclosures: E.L. Kerr: None. L.M. Savage: None.

Poster

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Program #/Poster #: PSTR296.18/JJ7

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 5U01AA028710-05
NIH Grant 2P50AA017823-16

Title: Adolescent intermittent ethanol exposure in rats alters cholinergic but not GABAergic proteins in the mPFC and hippocampus

Authors: *M. FECIK^{1,2}, L. M. SAVAGE²;

¹Psychology, Binghamton Univ., Kirkwood, NY; ²Psychology-Behavioral Neurosci., SUNY - Binghamton Univ. Behavioral Neurosci., Binghamton, NY

Abstract: Adolescent intermittent ethanol (AIE) exposure causes lasting deficits to the cholinergic phenotype in the basal forebrain. However, some cholinergic neurons also co-transmit GABA, potentially modulating the function of the hippocampus (HPC) and medial prefrontal cortex (mPFC). Presently, it remains unclear if AIE affects the GABAergic phenotype in these cells. Therefore, the goal of the current study was to determine if AIE differentially affects proteins involved in cholinergic and GABAergic signaling in the mPFC and HPC. Long Evans rats of both sexes received AIE or water on a two-day-on two-day-off schedule from postnatal day (PD) 25-54 and were euthanized in young adulthood at PD 105. Western blot was used to quantify cholinergic proteins (VACHT, AChE, N-type Ca²⁺ channel, $\alpha 5$ nAChR, and $\alpha 7$ nAChR) and GABAergic proteins (GAD65, GAD67, P/Q-type Ca²⁺ channel) in the mPFC and HPC. Contrary to our hypothesis, AIE increased $\alpha 5$ nAChR in the mPFC and AChE in the HPC of females and increased the N-type Ca²⁺ channel in males. These changes may be due to compensatory upregulation following AIE. Notably, VACHT was unchanged in the mPFC, contrasting previous data and suggesting that this protein may recover following a period of abstinence. No significant changes to GABAergic proteins were observed in either region. These results indicated that AIE produces region and sex-specific alterations to downstream targets of the cholinergic basal forebrain that could potentially lead to some of the long-term learning and memory deficits seen in this model. These results will eventually be accompanied by data taken 24 hours after AIE treatment to examine time-dependent changes.

Disclosures: M. Fecik: None. L.M. Savage: None.

Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.19/JJ8

Topic: H.08. Drugs of Abuse and Addiction

Title: Galantamine reverses habitual responding induced by adolescent binge alcohol exposure in rats

Authors: *A. B. ROY;

Psychology, Binghamton Univ., Binghamton, NY

Abstract: Adolescent heavy binge-type alcohol exposure is known to induce structural brain alterations, particularly affecting late-developing regions such as the orbitofrontal cortex (OFC). In rodent models, adolescent intermittent ethanol (AIE) exposure leads to persistent behavioral

and neurobiological changes. The OFC plays a crucial role in reward evaluation and decision-making and is particularly vulnerable to ethanol's neurotoxic effects. One measure of behavioral flexibility is reward devaluation, which assesses an individual's ability to adapt to changes in reward value. Impairments in this domain, often observed following AIE, are linked to the development of habitual behavior.

Previous research has demonstrated that AIE suppresses expression of choline acetyltransferase (ChAT) in the nucleus basalis of Meynert (NbM) and reduces vesicular acetylcholine transporter (VACHT) expression in the OFC, suggesting deficits in cholinergic signaling. Enhancing cholinergic tone may reverse these AIE-induced deficits. Galantamine, a cholinesterase inhibitor used clinically to treat Alzheimer's Disease and Related Dementias, has shown promise in restoring cholinergic markers following AIE.

Our findings indicate that AIE impairs OFC function, operant reward behavior, and reward devaluation behavior, consistent with increased behavioral inflexibility and habitual responding. Importantly, galantamine treatment restores goal-directed behavior in AIE-exposed animals: Rats were trained to lever press on a Fixed Ratio schedule for 3 days, then a Random Interval-30 for 2 days, and followed by a Random Interval-60 schedule for 4 days. Rats received either banana or chocolate flavored sucrose pellets during these training sessions, then the reward was devalued by a sensory-specific satiation procedure in which rats received access to either the same training flavor for devaluation or a novel flavor ad libitum before an extinction session.

Behavior during the extinction session revealed that saline-treated AIE rats did not decrease responding to the devalued flavor, whereas galantamine treated AIE rats and saline-treated control rats showed the traditional suppression of lever pressing for the devalued flavor. These results suggest that galantamine may rescue AIE-induced impairments in behavioral flexibility by enhancing cholinergic signaling in the OFC.

Disclosures: A.B. Roy: None.

Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

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Topic: H.08. Drugs of Abuse and Addiction

Support: San Antonio Medical Foundation
South Texas Undergraduate Research Opportunities Program, Summer
Physiology Undergraduate Researcher (SPUR) Program (R25NS115552,
PI: Manzoor Bhat)

Title: Assessing Blood-Brain Barrier Transport of Systemic Oxytocin Injections in Two Mouse Strains

Authors: *P. B. STEWART¹, J. JARAMILLO³, K. RIEDER⁴, A. B. SHAKOCIUS⁵, M. RUBAL³, K.-C. LEONG⁶, G. G. GOULD²;

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Abstract: Alcohol Use Disorder (AUD) is a persistent and complex neurological and psychological concern in the United States that has become taxing on American healthcare infrastructure. Stress and anxiety are driving factors in AUD progression and incidence of relapse. BTBR T+ Itpr3tf/J mice exhibit behavioral parallels of autism and elevated corticosterone comparable to elevated peripheral cortisol in people with autism, of whom 1 in 3 are suspected to have alcohol or substance use disorders. C57BL/6J mice were included as a control for the model. Oxytocin, when administered systemically and directly into the central nervous system, has anxiolytic effects and reduce stress-induced alcohol relapse in rodents. Previous data indicate that systemic administration of oxytocin does little to provide therapeutic effects without administering large doses. To increase the incidence of oxytocin transport across the blood-brain barrier and binding its receptors at lower treatment concentrations, oxytocin molecules were encapsulated in a unique liposome formulation. We hypothesized BTBR mice would have strain-specific blood-brain barrier transport and/or oxytocin receptor density differences inhibiting central oxytocin binding from a peripheral injection relative to C57BL/6J mice. In this project, BTBR and C57BL/6J males received intraperitoneal injections of 1 or 10 mg/kg oxytocin and were euthanized after 3, 30, or 60 minutes with the aim of measuring oxytocin binding to brain receptors. It was observed that blood oxytocin increased 30 and 60 min after oxytocin injection relative to control treatment. Serum was used to measure oxytocin levels by enzyme linked immunosorbent assay. Brains were coronally sectioned to 20 um thickness in a cryostat and mounted onto subbed microscope slides. Quantitative autoradiography was performed using ¹²⁵I ornithine vasotocin analog (OVTA) due to its high affinity for oxytocin receptors. After thawing, sections on slides were incubated in ¹²⁵I OVTA, rinsed, dried, and opposed to film for 48 hours. The film was developed and images of the anterior olfactory nucleus, lateral septal nucleus, hippocampal cornu ammonis 3 (CA3), and the ventral tegmental area were calibrated and quantified using ImageJ software. In BTBR males, acute injection of 1 mg/kg oxytocin did not reduce ¹²⁵I OVTA binding in any brain region measured. In C57BL/6J males, ¹²⁵I binding was significantly lower ($p < 0.05$), determined by an analysis of variance, in the hippocampus CA3 60 minutes after an acute 10 mg/kg injection.

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Poster

PSTR296: New Vistas in Preclinical Alcohol Research

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR296.21/JJ10

Topic: H.08. Drugs of Abuse and Addiction

Support: San Antonio Medical Foundation Collaborative Grant 2024

Title: Enhanced oxytocin diminishes yohimbine-induced alcohol responding under a progressive ratio schedule in female Long-Evans rats

Authors: *N. E. CORNEJO¹, J. JARAMILLO³, K. N. WOLFHOPE¹, A. J. GUPTA¹, S. PATEL¹, G. G. GOULD⁴, M. RUBAL³, K.-C. LEONG²;

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Abstract: Alcohol use disorder (AUD) is a debilitating condition of recurrent alcohol use and is complicated by the experience of stress, which is known to exacerbate alcohol use, craving, and relapse. Therefore, viable therapeutics for AUD should be assessed for potential to minimize the influence of stress on alcohol-seeking behavior and use. Oxytocin (OXT) is a neuropeptide with an established anxiolytic profile that has been shown to attenuate alcohol intake and relapse in rodents and may be a relevant treatment for AUD, though the therapeutic value of OXT is limited by various factors. Advanced delivery mechanisms such as liposome encapsulation of the compound can provide direct transportation of drugs to the brain, increasing bioavailability with minimal enzymatic degradation and thus may optimize the beneficial effect of OXT in a rodent model of AUD. In order to elucidate the potential therapeutic effect of OXT on stress-induced alcohol responding, we trained female Long-Evans rats (n = 30) to self-administer alcohol (20% v/v) on a FR1 schedule of reinforcement, followed by a FR3 schedule of reinforcement. After establishing stable responding, animals underwent progressive ratio (PR) testing to evaluate motivated responding for a single ethanol reward. Administration of the pharmacological stressor yohimbine (2 mg/kg, i.p.) resulted in significantly increased ethanol responding. When systemic OXT (1 mg/kg, i.p.) was co-administered with yohimbine, this attenuated yohimbine-evoked alcohol responding on a PR schedule of reinforcement as shown through a one-way ANOVA. Importantly, early evidence indicates concurrent administration of liposome-encapsulated OXT (1 mg/kg, i.p.) and yohimbine produced an enhanced attenuation of active lever presses and breakpoint during a PR test. These results demonstrate that systemic OXT and liposome-encapsulated OXT can attenuate stress-enhanced alcohol responding, with the enhanced formulation of liposome-OXT providing a greater therapeutic effect. Further investigation of enhanced OXT delivery in rodent models of AUD may yield optimized treatment for AUD-afflicted individuals.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

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Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant R25DA057786
NIH Grant R01DA052618

Title: Investigating the structure of perineuronal nets during fentanyl withdrawal

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Abstract: Perineuronal nets are specialized net-like structures of the extracellular matrix that are involved with adult brain plasticity. They surround specific neurons in the brain, and the reticular structure acts as a physical barrier to new incoming synaptic inputs. However, perineuronal nets can be remodeled due to experience and environmental factors. While there has been some research on the involvement of perineuronal nets in opioid responses, much remains to be done on what could be an important plasticity marker. The existing literature on morphine self-administration found that during acute withdrawal, there was an increase in perineuronal nets in the infralimbic area (ILA). Given this, we hypothesize that there will be an increase in the density and intensity of perineuronal nets in the infralimbic cortex during withdrawal. To test this hypothesis, we used two distinct mouse models of exposure to fentanyl with C57BL/6 mice ($n_{\text{total}}=48$, males= 24, females = 24, sex differences were not assessed). The first was 1 round of escalating doses of fentanyl or saline (acute) and the second was 5 cycles of 5-day escalating doses of fentanyl or saline with a 4-day withdrawal period between each escalation (prolonged). At different time points after the last intraperitoneal injection (16 hrs, 24 hrs, and 7 days), brains were extracted and sliced according to our regions of interest. Using lectin histochemistry to directly examine the perineuronal nets in the ILA, the results currently indicate that there are no significant differences between the total intensity of PNNs in the ILA of those exposed to fentanyl, compared to the vehicle controls ($\text{Prob} > |t| = 0.0913$). However, these results only include half of our experimental animals; we plan to further analyze the other group of animals in addition to analyzing individual PNN intensities within one sample of the ILA section to compare between experimental groups. Time permitted, we may also investigate different regions of interest, including a sample of the cortex and the central amygdala. If fentanyl withdrawal does affect perineuronal nets, there could be future research into perineuronal net interventions that may have positive implications on patients who are addicted to fentanyl.

Disclosures: M.T. Rice: None. A. Asarch: None. A.Y. Coyne: None. D.S. Hurlimann: None. K.R. Coffey: None. J.F. Neumaier: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.02/JJ12

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 1ZIADA000566-12

Title: The peripherally restricted opioid antagonist naloxone-methiodide reverses fentanyl-induced brain hypoxia via central mechanisms

Authors: *E. A. KIYATKIN¹, F. LIMAC², M. NOYA³, Y. SHAHAM⁴;
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Abstract: Brain hypoxia is a critical consequence of opioid overdose, traditionally attributed to the activation of central opioid receptors. However, peripheral receptors have also been implicated, as studies using naloxone-methiodide, a peripherally restricted opioid antagonist, show that it can reverse fentanyl-induced respiratory depression. This interpretation assumes naloxone-methiodide does not enter the brain, yet our previous mass spectrometry data revealed that it crosses the blood-brain barrier at a ~1:50 ratio compared to naloxone-HCl. In this study, we used amperometric oxygen sensors in freely moving rats to assess the role of central versus peripheral opioid receptors in fentanyl-induced (30 µg/kg IV) brain hypoxia. We compared the effects of naloxone-methiodide (2.0 mg/kg IV) and naloxone-HCl and found that while naloxone-methiodide significantly reduced hypoxia. However, naloxone-methiodide was less potent and its effects were shorter than a ten-fold lower dose of naloxone-HCl. Furthermore, naloxone-HCl at 50- and 250-times lower doses (50 and 8 µg/kg), but not at 1000-fold lower dose (2 µg/kg), mimicked the effects of naloxone-methiodide. These results suggest that the ability of naloxone-methiodide to block fentanyl-induced brain hypoxia is at least partially mediated by central opioid receptor antagonism, challenging its designation as a strictly peripherally acting compound and emphasizing the need to reevaluate its use in mechanistic studies.

Disclosures: E.A. Kiyatkin: None. F. Limiac: None. M. Noya: None. Y. Shaham: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Program #/Poster #: PSTR297.03/JJ13

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 1ZIADA000566-12

Title: Comparison of fentanyl-induced brain oxygen responses following intravenous and intraperitoneal injections in rats

Authors: *F. LIMAC¹, M. NOYA¹, E. A. KIYATKIN²;
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Abstract: Fentanyl, a potent synthetic opioid, is known to cause rapid respiratory depression and brain hypoxia. To investigate its physiological impact beyond respiration, oxygen amperometry was used to monitor changes in brain (nucleus accumbens) and peripheral (subcutaneous) oxygen levels following intravenous (IV) and intraperitoneal (IP) fentanyl administration in rats. To minimize stress, IP injections were delivered via chronically implanted catheters. Subcutaneous oxygen levels showed a dose-dependent monophasic decrease. In contrast, brain oxygen responses were dose- and route-dependent: low doses ($<3 \mu\text{g/kg IV}$, $<10 \mu\text{g/kg IP}$) induced modest increases, while higher doses ($\geq 10 \mu\text{g/kg IV}$, $\geq 270 \mu\text{g/kg IP}$) produced biphasic responses (decrease followed by rebound). IV fentanyl elicited faster, more potent effects, with over 10-fold greater efficacy in inducing brain hypoxia than IP fentanyl. Traditional needle-based IP injections triggered rapid brain oxygen increases and SC oxygen decreases, consistent with stress-related effects. Interestingly, stress-free low-dose IP fentanyl also produced transient brain oxygen increases and behavioral activation, independent of opioid receptor action. This effect was attributed to the low pH (4.8) of fentanyl citrate and activation of sensory nerves in the abdominal cavity. These findings highlight how administration route, injection stress, and drug formulation contribute to fentanyl's oxygen and behavioral effects.

Disclosures: F. Limiac: None. M. Noya: None. E.A. Kiyatkin: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

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Program #/Poster #: PSTR297.04/JJ14

Topic: H.08. Drugs of Abuse and Addiction

Support: National Science and Technology Council, R.O.C.

Title: Modulation of environmental enrichment to morphine-induced conditioned place preference in the animal model

Authors: *J. PAN¹, A. C.-W. HUANG²;

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Abstract: Concerning the issue that how to ameliorate morphine addiction via the intervention of environmental enrichment (EE) remains unclear. This study investigated this issue by using the morphine-induced conditioned place preference (CPP) and a 21-day conventional EE procedure. Following this, half of the rats received intraperitoneal injections of 10 mg/kg morphine, while the control group received normal saline. During the CPP paradigm, animals were confined to one compartment of a CPP box for 30 minutes after morphine injections on odd days. On even days, all rats received normal saline injections and were placed in the alternate compartment for 30 minutes. Finally, all rats underwent a 15-minute free-choice test in the CPP box, allowing for measurement of time spent in each compartment. Our results showed that

morphine effectively induces CPP. Furthermore, and contrary to some expectations, our findings suggest that EE manipulations may facilitate, rather than attenuate the morphine-induced CPP. The immunohistochemical analysis was performed to examine the expression of brain-derived neurotrophic factor (BDNF) and phosphorylated tropomyosin receptor kinase B (p-TrkB) in the medial prefrontal cortex, amygdala, and hippocampus following the CPP tests. These findings are crucial for elucidating the underlying brain mechanisms by which environmental enrichment influences morphine addiction.

Disclosures: J. Pan: None. A.C. Huang: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.05/JJ15

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH U54 MD007579

Title: Fibroblast growth factor 2 expression is associated with sex-dependent fentanyl seeking in adolescence following maternal separation

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Abstract: Early childhood neglect (ECN) accounts for up to 75.3% of child maltreatment cases in the U.S. annually and is associated with increased susceptibility to opioid use disorder (OUD). Adolescent abusers are 17.5 times more likely to develop OUD throughout their lifetime. Reductions in fibroblast growth factor-2 (FGF-2), a key neurodevelopmental protein, have been linked to heightened stress responses and impaired synaptic plasticity in brain regions implicated in addiction. Furthermore, fentanyl enhances corticotropin-releasing hormone (CRH) release, driving hypothalamic-pituitary-adrenal (HPA) axis activation. Here, we assess the effects of maternal separation (MS), a rodent model of ECN, on fentanyl-seeking behavior, corticosterone levels, and FGF-2 expression in the prefrontal cortex (PFC) and hippocampus. Sprague Dawley rat pups (n=70; male=34, female=36) were cross-fostered at birth and subjected to MS (3h/day; P2-P18) or full maternal care (controls). Rats underwent conditioned place preference (CPP) at adolescence (P32) or adulthood ($\geq$ P50) under one of four conditions: (1) control-saline, (2) control-fentanyl (5 μ g/kg, s.c.), (3) MS-saline, or (4) MS-fentanyl. To examine the role of sex hormones, females received testosterone propionate (1 mg/kg, s.c.) on P1. Plasma corticosterone levels were measured via ELISA at 30 minutes and 2 hours after fentanyl injection, while FGF-2 expression in the PFC and hippocampus was assessed using Western blot. We found that MS enhanced the magnitude and persistence of fentanyl seeking in adolescent and adult males as compared to control non-MS rats (p<0.05). In contrast, MS impaired the formation of fentanyl

seeking in adolescent and adult female rats as compared to control non-MS rats ($p < 0.05$). Masculinized females displayed fentanyl-seeking behavior similar to MS-exposed males. MS potentiated the HPA axis response to fentanyl in males but blunted it in females. MS also reduced FGF-2 levels in the PFC and hippocampus of both sexes, but fentanyl exposure restored FGF-2 expression in MS-exposed males, potentially contributing to relapse vulnerability. Our findings demonstrate that early-life neglect produces sex-dependent alterations in fentanyl reward-seeking behavior, HPA axis function, and neurodevelopmental signaling. These results underscore the importance of considering sex as a biological variable in addiction research and highlight the role of early-life stress in opioid vulnerability.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Title: Cross-ancestral gene-target prioritization for opioid use disorder

Authors: *S. DE ALMEIDA¹, P. ROUSSOS², G. VOLOUDAKIS³;
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Abstract: Recent genome-wide association studies (GWAS) have identified genetic loci linked with Opioid Use Disorder (OUD), revealing promising therapeutic targets. However, variability in disease definitions and population structures among cohorts often dilute genetic association signals, a challenge known as effect size dilution. Additionally, existing OUD GWASs lack cell-type specificity and comprehensive cross-ancestry analyses. To address effect size dilution and enhance discovery power, our study employs an innovative strategy to mitigate phenotypic heterogeneity. Furthermore, by combining cell-type-specific gene expression data and performing comprehensive cross-ancestry analyses, we aim to pinpoint causal genetic variants associated with OUD. This integrative approach advances the understanding of gene dysregulation mechanisms underlying OUD in diverse populations.

To correct for effect size dilution, we employed PheMED, a cutting-edge statistical methodology designed to enhance the power of GWAS discoveries. Preliminary analyses were conducted on the largest reported OUD GWAS data from the Million Veterans Program for African and European genetic ancestries. We further incorporated state-of-the-art expression quantitative trait locus (eQTL) reference panels to map gene activity patterns to specific brain cell types. Mendelian Randomization, a causal inference statistical framework, was utilized to integrate cell-type-specific genetic variants that have direct targets of clinically available compounds. Our results demonstrate that correcting for effect size dilution using PheMED significantly enhances gene discovery power across ancestry. Leveraging eQTL data within a Mendelian Randomization framework revealed several novel and previously reported genes as potential treatment targets for OUD in multiple ancestral groups. For African genetic ancestry, *RTN4* and *EPHX2* were identified in astrocytes involved in lipid metabolism and neural signaling. For European genetic ancestry, *NCAM1* was found in inhibitory neurons and *GABRA2* in excitatory neurons previously linked to heroin dependence. Correcting for phenotypic heterogeneity and utilizing cell-type-specific gene expression data reveals novel mechanistic insights into OUD and identifies actionable gene targets for therapeutic development. This research has the potential to expand personalized medicine approaches for OUD treatment and improve reproducibility in genetic studies involving diverse populations.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.07/JJ17

Topic: H.08. Drugs of Abuse and Addiction

Support: DA059601 (MEW)

Title: Incubation of oxycodone craving: pathway-specific upregulation of calcium-permeable AMPAR receptors in the rat nucleus accumbens core and shell

Authors: *H. M. KUHN^{1,2}, E.-K. HWANG², K. A. ENGELN², M. M. BEUTLER², S. J. WEBER^{3,2}, G. S. DRISCOLL², J. G. WESTLAKE², A. M. WUNSCH², M. E. WOLF²; ²Behavioral Neurosci., ¹Oregon Hlth. & Sci. Univ., Portland, OR; ³Neurobio., Univ. of Maryland, Baltimore, Baltimore, MD

Abstract: One diagnostic criterion for opioid use disorder is intense craving for opioids even after protracted abstinence. We study the basis of this persistent craving with a focus on oxycodone (Oxy), which remains a highly prescribed opioid despite high abuse liability, using the incubation of craving model. Adult male and female Long Evans rats self-administer Oxy (6 h/d x 10 d, 0.1 mg/kg/infusion; controls self-administer saline) followed by forced abstinence

during which cue-induced Oxy seeking progressively increases (incubates) and then remains high for at least a month. We previously published behavioral data indicating that expression of Oxy incubation is blocked on forced abstinence day 15 (AD15) and AD30 by intra-nucleus accumbens (NAc) core or shell injection of Nasp^m, a selective antagonist of Ca²⁺-permeable AMPARs (CP-AMPARs), suggesting CP-AMPARs upregulate during Oxy incubation. We are further investigating this using D1-Cre and A2a-Cre (A2a colocalizes with D2) transgenic rats, crossed with a reporter line for slice recordings (sample size ≥8-9 cells/5-6 rats). All studies use both sexes, but we are not powered to detect sex differences because both sexes show Oxy incubation and CP-AMPAR upregulation. We found elevated CP-AMPARs in core and shell subregions on both D1 and A2a medium spiny neurons (MSN; p<0.05 vs saline for all MSN subtypes), contrasting with CP-AMPAR upregulation only on D1 MSN after psychostimulant incubation. Thus, Oxy incubation entails unique plasticity, making it important to identify glutamate inputs that are involved. We are focusing on the paraventricular nucleus of the thalamus (PVT)-NAc pathway (implicated in both rewarding and aversive behaviors) and the basolateral amygdala (BLA)-NAc pathway (which encodes cue-driven behaviors). Preliminary optogenetic studies (AAV-ChR2 in posterior PVT or BLA) show CP-AMPAR upregulation following incubation in the PVT-NAc shell pathway (p<0.001 vs saline), but not PVT-NAc core, and in both BLA-NAc core and BLA-NAc shell pathways (p<0.001 vs saline). Complementary chemogenetic studies for both pathways are underway in which wild-type rats receive AAV-hM4Di (inhibitory DREADD; or mCherry control virus) infusion in posterior PVT or BLA prior to Oxy self-administration and CNO (or vehicle) is delivered into NAc core or shell prior to a cue-induced seeking test on AD21. Results to date indicate augmentation of seeking after inhibition of the PVT-NAc core pathway (p<0.05, CNO vs vehicle) and no effect in PVT-NAc shell. Taken together, these results do not support a role for CP-AMPAR plasticity in the PVT-NAc inputs in driving Oxy incubation, while the BLA-NAc inputs remain a candidate.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.08/JJ18

Topic: H.08. Drugs of Abuse and Addiction

Support: ZIA-DA000644

Title: Distinct Amygdala Neuronal Populations Control Opioid Use and Withdrawal in Mice

Authors: *L. SILVA TORTORELLI¹, H. OO¹, S. HAHN¹, Y. ALVAREZ¹, Y. CARRASQUILLO², L. F. VENDRUSCOLO³;

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Abstract: Opioid use disorder (OUD) is a chronic and recurring psychiatric disorder that is associated with high morbidity and mortality. The central nucleus of the amygdala (CeA) undergoes neuroadaptations in both humans with OUD and opioid-dependent rodents. As part of a heterogeneous microcircuits, the CeA integrates internal and external sensory inputs that drive innate and adaptive behaviors. Key CeA neuronal populations, including protein kinase C- δ (PKC- δ), corticotropin-releasing factor (CRF), and somatostatin (SST) neurons, regulate behaviors that are disrupted in addiction, such as pain, stress, reward function, and anxiety/arousal. We hypothesized that these CeA neuronal populations differentially regulate opioid-related behaviors. Using *in situ* hybridization, we identified a decrease in μ -opioid receptor (MOR; *Oprm1*) mRNA expression in the CeA in opioid-dependent mice that were undergoing withdrawal compared with nondependent mice. In contrast, the expression of PKC- δ (*Prkcd*), CRF (*Crh*), and SST (*Sst*) mRNA levels remained unchanged. To assess the functional role of these CeA neuronal populations, we used behavioral and molecular approaches in opioid-dependent mice. The chemogenetic inhibition of CeA^{PKC- δ} neurons decreased fentanyl vapor self-administration and alleviated fentanyl withdrawal-induced hyperalgesia. The inhibition of CeA^{CRF} neurons reduced irritability and somatic withdrawal signs. The activation of CeA^{SST} neurons reduced somatic withdrawal signs. These findings suggest that distinct CeA neuronal populations uniquely regulate different aspects of opioid use and withdrawal, highlighting cell-type specific targets for potential therapeutic interventions.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

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Topic: H.08. Drugs of Abuse and Addiction

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NIDA Grant T32DA007288-30
NIDA Grant K99DA057390

Title: Ventral pallidum cell and circuit opposing regulation of heroin seeking versus refraining behavior

Authors: *B. N. KUHN¹, J. L. HOPKINS¹, A. BERCKMUELLER², P. W. KALIVAS³;
¹Neurosci., Med. Univ. of South Carolina, Charleston, SC; ²Baylor Univ., Williamsburg, VA;
³Neurosci. Res., Med. Univ. S Carolina, Charleston, SC

Abstract: Substance use disorder (SUD) is a chronic neuropsychiatric disorder whereby compulsive drug seeking and relapse occurs regardless of attempts to refrain from drug use. Understanding the neurobiological mechanisms underlying drug seeking versus refraining (i.e., withholding from seeking) are necessary to better treat those afflicted with SUD. The ventral pallidum (VP) exhibits opposing regulation of appetitive and aversive motivated behaviors, including cell-specific regulation over cocaine seeking behavior. The goal of the current study is to assess VP cell and cell-specific circuit regulation of heroin seeking behavior. Male and female transgenic mice (n=7-10/group) expressing Cre recombinase selectively in either GABA, enkephalin (co-expressed in a subset of GABA cells) or glutamate cells underwent heroin self-administration and extinction training, followed by tests for cued reinstatement. During reinstatement, cell-specific chemogenetic inhibition or stimulation occurred to assess cellular contributions to heroin-seeking behavior. Selective chemogenetic stimulation of VP GABA or enkephalin cells enhanced cue-induced reinstatement of heroin-seeking behavior, whereas VP glutamate neuron stimulation promoted refraining behavior. Selective inhibition had opposing effects for all cell types. Next, an intersectional chemogenetic approach was used to assess the contribution of cell-specific VP efferent pathways in mediating heroin seeking. Activation of VP glutamate projections to the lateral hypothalamus, a critical component of the motive circuitry, attenuated heroin seeking whereas stimulation of GABAergic efferents had no effect. Contrary to expectations, activation of VP GABAergic, but not enkephalin, efferent projections to the subthalamic nucleus, a recently focused upon node involved in reward seeking, also reduced heroin-seeking behaviors. This effect was not due to a general locomotor deficit. The VP GABA pathway driving seeking behavior is currently under investigation. Extending upon these findings, using a rat model capturing individual variation to SUD and RNAscope technology, VP cell-specific neuronal activation in vulnerable versus resilient rats following cued reinstatement is currently under investigation. The objective of this experiment is to elucidate how VP cell types naturally engage in promoting seeking behavior in different addiction phenotypes. Overall, these results emphasize the complex and VP cellular and circuitry contributions to heroin seeking and refraining behaviors.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Topic: H.08. Drugs of Abuse and Addiction

Support: NIH/NIDA R37 DA054370

Title: Fentanyl distorts phasic dopamine release dynamics in the ventral striatum encoding natural reward and aversive processing.

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Abstract: Temporally specific dopamine (DA) release in the ventral striatum (VS) plays key roles in shaping reinforcement learning in animals by facilitating obtaining natural rewards and avoidance of aversive stimuli. The potent mu opioid agonist fentanyl increases the tonic activity of DA neurons, promoting opioid reward and consequentially opioid addiction, but fentanyl's effects on phasic DA release patterns is not well understood. Previous studies have also focused on the rewarding effects of fentanyl itself, however, its effects on phasic DA responses to natural rewards, aversive stimuli, and their predictive cues are poorly understood. In this study, we determined the effects of acute doses of fentanyl on phasic DA dynamics encoding conditioned and unconditioned responses to liquid sucrose rewards and aversive footshock stimuli (0.35 mA, 200ms) using fiber photometric recordings of GRAB-DA signals in the ventral striatum of freely behaving male C57BL/6J mice. Behaviorally, we observed that both 0.2 mg/kg and 0.1 mg/kg subcutaneous injections of fentanyl significantly reduced the anticipatory reward port entries and overall liquid sucrose consumption, as well as freezing responses to shock predictive cues. 0.1 mg/kg dose of fentanyl produced paradoxical increases in motoric responses to footshocks. These behavioral responses were correlated with the phasic DA release patterns measured in the VS. Both 0.2 mg/kg and 0.1 mg/kg injections of fentanyl significantly reduced DA phasic release in response to sucrose-predictive auditory cues as compared to saline control injections, whereas, 0.1 mg/kg fentanyl injections caused significant positive shifts in DA phasic responses to aversive stimuli and their predictive cues. Overall, our findings indicate that acute doses of fentanyl significantly distort DA encoding of natural reward and aversion-related stimuli, in a manner that parallels altered behavioral responses to these same stimuli.

Disclosures: O. Culver: None. T.C. Jhou: None. N. Schaffer: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Program #/Poster #: PSTR297.11/JJ21

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH DA031900
NSF GRFP No. 2041772

Title: Opioids modulate dopamine uptake in the nucleus accumbens through local circuit mechanisms

Authors: *A. R. BASHFORD, K. R. SAMSON, Z. D. BRODNIK, R. A. ESPAÑA;
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Abstract: Opioid use disorder remains a pressing public health crisis, yet the mechanisms by which opioids alter dopamine transmission remain incompletely understood. It is traditionally thought that opioids increase dopamine levels in the nucleus accumbens (NAc) through mu opioid receptor (MOR) activation on midbrain GABA neurons that results in disinhibition of dopamine neurons in the ventral tegmental area. However, recent work from our laboratory demonstrates that the opioid oxycodone reduces dopamine uptake in a NAc brain slice preparation that does not include midbrain dopamine neurons, revealing a local circuit mechanism independent of the ventral tegmental area. To further investigate this phenomenon, we used fast-scan cyclic voltammetry to examine the effects of several MOR agonists (oxycodone, fentanyl, morphine, and DAMGO) on dopamine uptake in the NAc. We found that all tested opioids significantly reduced dopamine uptake, suggesting a common mechanism across MOR agonists. Given that dopamine terminals lack MORs, we next probed the role of local interneuron populations that contain MORs as potential intermediaries. Pharmacological inhibition of cholinergic signaling with the $\beta 2$ -containing nicotinic receptor antagonist dihydro- β -erythroidine (DH β E) reduced dopamine uptake, mimicking opioid effects. Likewise, inhibition of GABAergic signaling with the GABAB receptor antagonist Saclofen also reduced dopamine uptake. Notably, our preliminary data shows that pretreatment with DH β E occludes the effect of subsequent oxycodone application, suggesting a shared mechanism. These results implicate cholinergic and GABAergic interneurons as potential mediators of opioid-induced modulation of dopamine uptake in the NAc. Ongoing studies using RNAscope will confirm the specific populations of NAc interneurons that contain MORs. Together, these findings provide novel insight into local circuit mechanisms by which opioids modulate dopamine signaling and suggest new avenues for understanding the reinforcing properties of opioids beyond canonical midbrain pathways.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Topic: H.08. Drugs of Abuse and Addiction

Support: VASDHS I01BX005782

Title: Circuit dissection of mu-opioid-receptor expressing neurons in ventral pallidum

Authors: *Y.-C. CHEN¹, A. J. FLORES², L. FAGET³, T. S. HNASKO⁴;

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Abstract: The ventral pallidum (VP) is a key brain region involved in motivation and reward-seeking behavior. Electrophysiological and pharmacological studies have implicated VP activity in substance abuse. Notably, the VP contains both pre- and post-synaptic mu-opioid receptors (MOR) that may contribute to opioid reward and dependence. However, the mechanisms by which opioids modulate VP activity remain poorly understood. To address this knowledge gap, we employed a multidisciplinary approach. Viral tracing showed that VP receives MOR+ afferents from the nucleus accumbens and sends MOR+ efferents to the lateral habenula, the ventral tegmental area, and other regions. *Ex vivo* slice electrophysiology showed that bath application of DAMGO inhibits MOR-expressing VP neurons, while *in vivo* fiber photometry revealed a multiphasic calcium response of MOR-expressing VP neurons following acute morphine injection. Finally, RNAscope-based in situ hybridization identified differential cell-type-specific expression of MOR in VP neurons. Together, these findings provide mechanistic insight into how opioids alter VP activity. Building on this foundation, we aim to investigate how prolonged opioid exposure reshapes this circuit to examine the role of the VP in opioid dependence.

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Program #/Poster #: PSTR297.13/KK1

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 5R37DA054370-04

Title: Anatomic Resolution of Mu-Opioid Receptor Expression Across GABAergic Projections to the Ventral Tegmental Area

Authors: *M. HOHMEISTER¹, T. C. JHOU²;

¹Neurobio., ²Neurosci., Univ. of Maryland Sch. of Med. Program In Neurosci., Baltimore, MD

Abstract: **Anatomic Resolution of Mu-Opioid Receptor Expression Across GABAergic Projections to the Ventral Tegmental Area.** **Authors** Maddie Hohmeister, Paul Culver, Thomas Jhou; Neurobiology, University of Maryland, Baltimore, MD **Disclosures** Maddie Hohmeister: None, Paul Culver: None, Thomas Jhou: None **Abstract** The addictive properties of opioids are widely considered to be driven by the drug's ability to increase dopamine neural activity via activation of mu opioid receptors (MORs) on inhibitory inputs to the ventral tegmental area (VTA) (Johnson and North, 1992). However, the VTA receives distinct GABAergic inputs that have a diverse impact on dopamine neural activity and subsequent behavioral phenotypes (Soden et al., 2020), and it is still unclear which of these inputs are disrupted by opioids. Multiple studies have identified certain VTA-projecting inhibitory pathways that may be more sensitive to the effects of opioids (Matsui et al., 2014), and thus more

therapeutically relevant. However, a comprehensive assessment of opioid sensitivity across all major input pathways is a necessary step for identifying therapeutic targets of opioid addiction. Our study utilized retrograde tracing, combined with RNA scope in situ hybridization to map *oprm1* (the MOR gene) distribution among VTA-projecting GABA neurons across multiple brain regions in male mice (n=4). Using this method, we identified that VTA-projecting neurons exhibiting expression of both *oprm1* and *gad2* (a marker of GABA neurons) were widespread across multiple brain regions. Interestingly VTA projecting GABA neurons originating from the bed nucleus of the stria terminalis (BNST), medial and lateral preoptic area (MPO/LPO), nucleus accumbens (NAc), and rostromedial tegmental nucleus (RMTg) exhibited particularly high concentrations of *oprm1* puncta compared to GABA neurons originating from other prominent projection pathways. These findings support previous evidence and provide further resolution about how certain inhibitory inputs to the VTA are more affected by the actions of opioids, thus illuminating more specific targets for therapeutic interventions.

Disclosures: M. Hohmeister: None. T.C. Jhou: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.14/KK2

Topic: H.08. Drugs of Abuse and Addiction

Support: Shirley and Stefan Hatos Foundation

Title: An Endogenous Opioid System in the Pacific Hagfish, a Vertebrate-Invertebrate Transition Species, Analogous to Mammalian Opioid Systems

Authors: *G. VENUMBAKA¹, R. SETIA², A. ROMERO², S. CHEN², P. BHANDARY⁴, P. J. FRED A⁵, A. ALAYOUBI⁶, C. J. EVANS³;

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Abstract: In mammals, the opioid system modulates a wide array of physiological processes, ranging from pain perception, reward, gut motility, and respiration. The mammalian endogenous opioid system comprises a family of four, highly homologous opioid G-protein coupled receptor genes (MOR, DOR, KOR, and ORL1) and four peptide precursors (proenkephalin (PENK), proopioidmelanocortin (POMC), prodynorphin (PDYN), pronociceptinophanin FQ (PNOC)). Here we describe the opioid system in the genome and transcriptome of the Pacific Hagfish (*Eptatretus stoutii*) which diverged from mammalian lineages over 400 million years ago. We identified four different opioid-like receptors that retain the same intron/exon boundaries as well as many other conserved features found in the mammalian opioid receptor family. The sequences of *E. stoutii* were compared with other hagfish species found in the Pacific Ocean. Conserved

features included N-terminal N-linked glycosylation sites, consensus regulatory kinase sites at the C-terminus, consensus palmitoylation sites, and highly conserved membrane-spanning domains and intracellular loops. In addition to the four hagfish opioid receptors, four opioid peptide precursors were identified in both the hagfish genome and tissue transcripts. Two of the precursors were highly homologous to mammalian PENK. The two other opioid precursors identified in the Hagfish predicted several novel opioid peptides that are unobserved in mammalian opioid precursors but retain the conserved YGGF-X opioid peptide motif preceded by pair basic residues (consensus sites for prohormone convertase cleavages). The precursors additionally retain conserved features at the N-terminus of the precursor proteins. No opioid-like precursors that were clearly homologous to PNOC, or POMC were identified in the hagfish genome or transcripts. Extensive analysis of invertebrate genomes found no homologous sequences to the hagfish or mammalian opioid peptide precursors. Although homologous GPCRs to opioid receptors have been annotated as “opioid receptors” in invertebrate genomes they lack key conserved features that are common to all members of the vertebrate opioid receptor family. This analysis confirms the expression of a heterogeneous opioid system that diverged from mammalian lineages over 400 million years ago. The study raises questions as to the evolution of the opioid system from invertebrate lineages.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.15/KK3

Topic: H.08. Drugs of Abuse and Addiction

Title: Ventral tegmental area projection neurons differentially encode aspects of fentanyl reward

Authors: *A. MONTEMARANO, H. SOHAIL, S. PANDEY, L. D. FOX, F. A. ALKHALEEL, A. E. OSTMAN, L. B. MURDAUGH, M. E. FOX;
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Abstract: Neurons in the ventral tegmental area (VTA) undergo molecular and physiological changes in response to opioid use that are heavily implicated in drug relapse. Previous work indicates morphine exposure induces structural and functional adaptations in VTA dopamine neurons depending on their downstream projection target, however no work has examined how this translates to different circuit activity during behavior, or if this extends to the synthetic opioid fentanyl. Here, we test the hypothesis that VTA neurons projecting to the nucleus accumbens (VTA-NAc) or medial prefrontal cortex (VTA-PFC) differentially encode aspects of fentanyl reward. First, we used GCaMP fiber photometry to monitor calcium activity of specific VTA projections during fentanyl conditioned place preference. We show VTA-NAc and VTA-PFC exhibit distinct patterns of calcium activity in response to fentanyl injection, where fentanyl

increases calcium transient amplitude and frequency in VTA-NAc neurons, but only increases calcium transient amplitude in VTA-PFC neurons with no effect on the frequency. These projections also exhibit different patterns of activity associated with the fentanyl-paired context after conditioning. We found VTA-PFC, but not VTA-NAc, shows increased calcium activity immediately preceding entry to the fentanyl-paired context. Next, we used dLight fiber photometry to monitor dopamine activity in the PFC and found increased dopamine in the PFC immediately preceding entry to the fentanyl-paired context. Lastly, we used local pharmacology to administer the dopamine D1 receptor antagonist SCH23390 in PFC or NAc. We found that blocking D1 receptor activity in PFC, but not in NAc, reduces preference for the fentanyl-paired side after conditioning. Together, these findings support the unique significance of the mesocortical pathway in the expression of fentanyl-context associations. Ongoing work is expanding these studies to a self-administration paradigm to investigate the role of these circuits in fentanyl seeking.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.16/KK4

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant 1R21NS136962-01A1
NIH Grant T32-NS105604

Title: Opioid modulation of the cerebellum: Exploring the cerebellar role in morphine-induced signaling and drug-associated behavior

Authors: *C. CLARK¹, P. G. MERMELSTEIN², M. CVETANOVIC¹;
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Abstract: As a leading cause of overdose death worldwide, opioids present a major health crisis that costs many lives and financially burdens health care systems. At the crux of this crisis is Opioid Use Disorder (OUD), a chronic drug disorder that is associated with high rates of drug relapse, presenting a challenge to treatment and recovery. Opioids have garnered significant research interest, but most studies investigate traditional reward-associated pathways and circuitry. Notably, deep cerebellar nuclei (DCN) have glutamatergic monosynaptic connections with the ventral tegmental area (VTA), a midbrain structure closely tied to reward processing. Recent work has shown DCN-VTA activity to modulate reinforcement learning, indicating this circuit could modulate opioid-associated behaviors. Considering the cerebellar role in opioid-associated behaviors is not well characterized, we sought to further explore cerebellar signaling during morphine administration. Using TRAP mice which permanently express tdTomato in

recently active neurons in the presence of Tamoxifen, we identified robust and widespread cerebellar neuronal activation in morphine versus saline treated animals. To explore how DCN signaling was influenced by repeated morphine exposure, we analyzed neuronal activation using the immediate early gene, cFos, after animals underwent 5 days of morphine or saline treatment. We found that repeated morphine administration induced a cFos response in the DCN which was not observed under saline or movement control conditions. I'm currently using an inhibitory DREADD expressed within the DCN of mice undergoing morphine conditioned place preference (CPP) to determine the effect on morphine-associated behavior. I hypothesize that cerebellar signaling will modulate morphine-associated preference and behavior. Collectively, these results suggest that the cerebellum likely plays a powerful role in opioid-associated reinforcement learning and subsequent drug-seeking behavior.

Disclosures: C. Clark: None. P.G. Mermelstein: None. M. Cvetanovic: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

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Program #/Poster #: PSTR297.17/KK5

Topic: H.08. Drugs of Abuse and Addiction

Support: 5R01DA058755-03

Title: Oxycodone self-administration increases ventral pallidal glutamatergic activity

Authors: *C. XU¹, J. TOOLEY², L. FANG³, M. CREED⁴;

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Abstract: Prescription opioids are powerful analgesics with high abuse liability, making them a major driver of opioid use disorder. The initial reinforcing effects of opioids are driven by increase in mesolimbic dopamine release; and chronic opioid intake leads to a constellation of withdrawal symptoms. Following this chronic intake, opioid use is driven by the alleviation of negative affect, rather than by the acute reinforcing effects. The neuroadaptations mediating negative affective state and negative reinforcement remain largely unknown. Ventral pallidum glutamatergic (VP^{Glu}) neurons regulate dopamine release and express mu opioid receptors (Mu-OR). In this study, we examined how oxycodone (OXY) self-administration induces adaptations in VP^{Glu} neurons and Mu-OR signaling pathways within VP circuits. We hypothesized VP^{Glu} neurons would exhibit increased excitability and synaptic output during protracted withdrawal due to sustained inhibition experienced during repeated opioid use. We collected VP tissues from VGluT2-cre; Ai14 mice for whole-cell patch-clamp recording and found VP^{Glu} neurons from abstinent mice fired more frequently in response to current injection compared to control neurons. Application of DAMGO, a selective Mu-OR agonist, reduced excitability in both naïve

and abstinent group. Nevertheless, neurons in the abstinent group maintained higher excitability relative to the control neurons in the presence of DAMGO. To assess synaptic output, we injected ChR2 into the VP of VGluT2-cre mice and recorded light-evoked postsynaptic currents from lateral habenula (LHb) neurons, the main target of VP^{Glu} neurons. We found that VP^{Glu} neurons release more glutamate onto LHb during abstinent without changes in release probability. DAMGO suppressed glutamate release at VP^{Glu}:LHb synapses in naïve but not in abstinent group. To determine whether opioid use induces pathway-specific adaptations, we recorded light-evoked postsynaptic responses from neurons in the ventral tegmental area (VTA) and rostromedial tegmentum. We found DAMGO inhibited glutamate release from VP projections to the midbrain in both naïve and abstinent group. In conclusion, VP^{Glu} neurons become more excitable during protracted withdrawal, and repeated opioid use blunts the ability of opioids to inhibit glutamate release onto the LHb but not onto the midbrain. Increased VP^{Glu} neuron outputs would lead to heightened LHb activity and reduced VTA dopamine activity, which are thought to contribute to the negative affective state. Our work suggests that inhibiting VP^{Glu} neurons may reduce opioid dependence by alleviating negative affect during protracted withdrawal.

Disclosures: C. Xu: None. J. Tooley: None. L. Fang: None. M. Creed: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Program #/Poster #: PSTR297.18/KK6

Topic: H.08. Drugs of Abuse and Addiction

Support: RO1HL169679-02

Title: Respiratory Muscle Rigidity Induced by Fentanyl is Independent from Systemic Oxygenation and Reversible with Naloxone

Authors: *J. SOTO¹, A. J. GARCIA, III², S. WEE¹;

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Abstract: The rise in ultrapotent synthetic opioid use, particularly fentanyl, poses a significant public health threat and is the leading cause of opioid-related deaths. The lethality of fentanyl is primarily attributed to respiratory depression resulting in severe hypoxia and metabolic distress. A growing body of evidence suggests that Fentanyl, unlike other opioids, increases propensity for overdose compared to control conditions. Fentanyl also induces chest wall rigidity, a distinct motor phenotype that may further compromise ventilation and increase the susceptibility to later stages of overdose, believed to persist upon reversal of respiratory depression. However, factors that influence fentanyl-induced chest wall rigidity remain poorly understood. Here, we test how oxygenation and reversal of respiratory depression impact this fentanyl-induced respiratory

motor phenomenon in a mouse model of fentanyl overdose. While fentanyl (0.7mg/kg i.p.) caused phasic inspiratory activity to become tonically active, neither hypoxia nor hyperoxia affected the fentanyl-induced motor phenotype. However, naloxone reversed both respiratory depression and tonic motor activity. Together, these observations show that fentanyl induces a discoordinated respiratory motor phenotype that is oxygenation-independent yet can be targeted by mu opioid receptor antagonism. The findings from this ongoing exploratory study provide new insight into the mechanisms of fentanyl-induced respiratory dysfunction and highlight naloxone's potential to reverse both ventilatory and motor components of overdose.

Disclosures: J. Soto: None. A.J. Garcia: None. S. Wee: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.19/KK7

Topic: H.08. Drugs of Abuse and Addiction

Support: 5R37DA054370-04

Title: Acute fentanyl administration suppresses VTA glutamate afferent signaling in instrumental behavior for sucrose rewards

Authors: *N. D. SCHAFFER¹, O. P. CULVER², T. C. JHOU¹;

¹Neurobio., Univ. of Maryland Sch. of Med., Baltimore, MD; ²Neurosciences, MUSC, Charleston, SC

Abstract: Glutamatergic transmission onto dopamine (DA) neurons in the ventral tegmental area (VTA) play a central role in evoking DA neuronal phasic activations, thus driving motivated behaviors and shaping instrumental learning. Previous research in our lab shows phasic DA responses to natural reward cues are diminished during acute fentanyl administration, however, which circuits drive this reduction is unknown. Thus, we hypothesized that acute fentanyl administration would diminish glutamatergic signaling onto VTA DA neurons during an instrumental task. To test this hypothesis, we injected male and female transgenic Dat-IRES-Cre mice with a Cre-dependent glutamate sensor (iGluSnFr) to examine glutamate signals onto VTA DA neurons (iGluSnFr-DA). Water-restricted mice were trained in an instrumental behavioral task to press a lever to receive a liquid sucrose reward. After training, we then photometrically measured iGluSnFr-DA signals during additional instrumental sessions in which mice were administered acute subcutaneous fentanyl (0.1-0.2 mg/kg) immediately prior to session start. As compared to saline control sessions, we found that acute fentanyl injections increased overall locomotor activity but eliminated reward-directed behavior. Moreover, iGluSnFr-DA activations to reward cues were markedly reduced during fentanyl sessions, with these activations returning 24 hours later. We further found that satiating animals with *ab libitum* water mimicked fentanyl's inhibitory effect on iGluSnFr-DA excitations and animal behavior. We hypothesize

that this effect constitutes a mechanism by which opioids transiently devalue natural rewards, thereby reducing previously conditioned instrumental responses to natural rewards and their associative cues.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Program #/Poster #: PSTR297.20/KK8

Topic: H.08. Drugs of Abuse and Addiction

Support: P30 DA051355-01

Title: Disruption of sleep architecture and sleep-state transition dynamics during opioid dependence

Authors: *S. PATEL¹, R. J. DOUGHERTY¹, T. B. C. JOHNSTONE², C. QU², R. C. VIZIN², H. ITO⁴, E. NAVRATILOVA², S. L. COWEN^{1,3}, F. PORRECA²;

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Abstract: Opioid use is frequently accompanied by persistent sleep disturbances and negative affective states, which can promote drug-taking behavior and relapse of opioid use disorder (OUD). While mu-opioid receptor (MOR) activation is associated with positive affective states, kappa opioid receptor (KOR) signaling plays a critical role in mediating negative affective states associated with opioid withdrawal. The contribution of KOR activity to morphine-induced alterations in sleep architecture is poorly understood. In this study, we aim to investigate whether KOR activation contributes to sleep fragmentation and dysphoric states during morphine dependence in mice and how morphine alone alters sleep architecture. We also investigated the effects of morphine and KOR inhibition on the likelihood of transitioning between waking (WAKE), slow-wave non-rapid-eye-movement (NREM), and rapid-eye movement (REM) sleep.

Approach: Male C57BL/6J mice (n=19) were implanted with morphine pellets and subjected to naloxone-precipitated withdrawal to assess physical and affective signs of dependence, including somatic behaviors and conditioned place avoidance. EEG/EMG electrodes were surgically implanted to monitor sleep with recordings collected continuously over 7 days. **Results:** Continuous morphine caused significant and lasting reductions in both NREM and REM sleep along with increased WAKE time. Pre-treatment with nor-binaltorphimine (nor-BNI), a long-acting KOR antagonist, 24 hours before morphine or placebo pellet implantation, restored NREM sleep to levels comparable to placebo controls but did not normalize REM sleep. To investigate how morphine alters the probability of transitioning between different states (NREM, REM, WAKE), we calculated state transition matrices from the 24-hour recordings. Examination of the transition matrices indicated that morphine disrupts normal sleep architecture by

increasing the frequency of WAKE-to-REM and NREM-to-REM transitions while reducing NREM-to-WAKE transitions. Co-administration of nor-BNI partially rescued these effects, restoring more stable and physiologically typical transitions. Together, these data indicate that morphine profoundly alters sleep architecture and the transition probabilities between WAKE, NREM, and REM, and that KOR inhibition can partly rescue these effects.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.21/KK9

Topic: H.08. Drugs of Abuse and Addiction

Support: R01HL163965-02S1

Title: Repeated Opioid Use Modulates Amyloid Beta Clearance and Microglia Responses in a Mouse Model of Alzheimer's Disease

Authors: ***S. WEE**¹, A. J. GARCIA, III², J. V. GOGOLA¹;

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Abstract: Alzheimer's disease (AD) is a complex neurodegenerative disease characterized by the accumulation of amyloid- β (A β) plaques, progressive cognitive decline, and behavioral changes. While it is well-recognized that neuroimmune status influences AD progression, a need exists for understanding how modifiable risks, such as chronic opioid use, may affect AD progression, as chronic opioid use can suppress immune signaling. Indeed, in a previous report, postmortem analyses of brains from human opioid users found accelerated changes in A β , other AD-related biomarkers, and immune response. However, the correlational nature of such retrospective epidemiological studies leaves open questions of the basis and interactions between repeat opioid use (ROU) and AD progression. This ongoing exploratory study aims to examine how ROU impacts amyloidogenesis using the APP/PS1 mouse model of AD exposed to repeated fentanyl administration (0.35mg/kg; 20 days). We focused on cortex and hippocampus because this model has a different seeding time course between the areas. We hypothesized that during early AD progression (P130-150), ROU would enhance A β plaque burden by suppressing the ability for microglia to metabolize A β . However, we found decreased soluble A β content following ROU (n=4) in cortex ($1.83 \text{ ng/mg} \pm \text{SE}0.075$) and hippocampus (0.22 ± 0.039) relative to saline control (n=4; cortex= 2.52 ± 0.47 , $p=0.01$; hippocampus= 0.69 ± 0.20 , $p=0.029$). The ROU induced decrease in A β content was consistent for insoluble A β (cortex: ROU= 1.92 ± 0.15 , SAL= 2.17 ± 0.03 , $p=0.008$; hippocampus: ROU= 1.83 ± 0.17 , SAL= 2.17 ± 0.09 , $p=0.06$)

Furthermore, histological labeling revealed a reduction in A β plaque burden following ROU in the hippocampus (n=4; ROU=67.69 $\mu\text{m}^2 \pm 5.7$, SAL=103.11 ± 11.3 , $p=0.029$) and cortex (ROU=124.51 ± 3.9 , SAL=143.13 ± 5.8 , $p=0.008$). This was accompanied by increased localization of disease-associated microglia (DAM) at A β plaques in hippocampus (ROU=57.7% ± 0.56 , SAL=18.80 ± 0.31 , $p=0.002$) and cortex (ROU=64.53 ± 1.4 , SAL=28.42% ± 0.34 , $p=0.007$). Preliminary data suggests an increased incidence of internalized A β among plaque-associated microglia. Together, these results demonstrate that ROU increases A β clearance during early amyloidogenesis. While contrary to our working hypothesis, these findings suggest that ROU decreases A β burden, likely by increasing microglia phagocytosis. The ongoing work aims to characterize the microglial-specific transcriptional profiles and determine the mechanisms by which ROU mediates these phenomena.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Program #/Poster #: PSTR297.22/KK10

Topic: H.08. Drugs of Abuse and Addiction

Support: CAPPI Pilot Grant

Title: Acute morphine and glucocorticoid signaling regulate FKBP5 expression in astrocytes

Authors: *A. CLEERE¹, R. A. PHILLIPS, III³, G. TWA⁴, N. ROBINSON⁵, L. IANOV⁶, R. E. SORGE⁷, J. J. DAY⁵, J. J. TUSCHER²;

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⁷Psychology, Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: Acute morphine and pain have been shown to induce transcriptional changes in the ventral tegmental area (VTA), a brain structure important in drug-seeking behavior, both *in vivo* and *in vitro*. We recently used single-nucleus RNA sequencing (snRNA-seq) in the VTA of adult rats to identify cell-type specific changes in gene expression after acute morphine experience in a chronic pain model. Surprisingly, we found that glial cell populations, which are non-neuronal cells that help support neuron function, had more changes in gene expression than neuronal cell populations. Astrocytes, a type of glial cell, were particularly transcriptionally responsive to acute morphine and showed strong induction of *Fkbp5*, regardless of pain state. Previous literature has implicated glucocorticoid receptors (GR) in pain responses and transcriptional changes of *FKBP5*. Here, we employed a human-derived astrocyte cell culture system to investigate the mechanisms behind opioid and glucocorticoid-induced changes in gene expression. We differentiated neural progenitor cells into astrocytes and delivered

pharmacological treatments. Astrocyte treatment with DAMGO, a mu opioid receptor antagonist, did not alter *FKBP5* expression. In contrast, treatment with either the endogenous glucocorticoid cortisol, or the GR agonist dexamethasone, both increased *FKBP5* expression in astrocytes. Pretreatment with the GR antagonist mifepristone blocked GR agonist-induced transcriptional changes. To improve both cell-type and receptor specificity, we next used an astrocyte-selective CRISPR interference (CRISPRi) tool for targeted silencing of *NR3C1*, the glucocorticoid receptor. Selective repression of *NR3C1* in astrocytes blocked induction of *FKBP5* following cortisol treatment. Collectively, these findings suggest GR, not mu opioid receptor, activation is both sufficient and necessary to induce *FKBP5* expression in astrocytes. We are currently working on validation of a CRISPRi tool for selective repression of *Fkbp5* in astrocytes for use in a rodent IVSA model to investigate its role in opioid seeking behavior.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Program #/Poster #: PSTR297.23/KK11

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH Grant R01DA053070

Title: Contingent and Non-contingent Fentanyl Regulates Different Potassium Channels Families in the Prelimbic Cortex

Authors: *F. WU¹, J. SHAYKIN², M. T. BARDO⁴, P. I. ORTINSKI³;

¹Univ. of Kentucky, Lexington, KY; ²Univ. of Kentucky, Lexington, KY; ³Neurosci., Univ. of Kentucky, LEXINGTON, KY; ⁴Dept Psychol, Univ. Kentucky, Lexington, KY

Abstract: Fentanyl abuse presents a devastating public health crisis in the US. Cellular responses to opioids in laboratory animals and humans involve potassium (K⁺) channels. However, which of the K⁺ channel groups may underlie cellular and behavioral plasticity associated with fentanyl self-administration remains poorly understood. Our study aimed to investigate how escalated fentanyl self-administration remodels subtype-specific potassium (K⁺) channel activities to impact neuronal excitability within the infralimbic cortex (IL) - nucleus accumbens shell (NAc) pathway, a circuit heavily involved in escalation of drug use. Young Adult Sprague-Dawley rats underwent a triadic model to study drug use behavior in three groups: contingent fentanyl, yoked fentanyl, and yoked saline. Following 3 weeks of fentanyl or saline administration in long-access (6 hrs) daily sessions, animals were sacrificed for brain extraction and brain slice preparation. *Ex vivo* whole-cell patch-clamp techniques were used to isolate activities of six distinct groups of K⁺ channels in the IL and the NAc neurons by applying channel-specific protocols. We observed a significant contingency-dependent increase of IL G-

protein coupled inwardly rectifying K⁺ (GIRK) channel activity. Contingent Fentanyl rats exhibited enhanced baclofen-induced GIRK currents compared to Yoked Fentanyl and Yoked Saline groups. Our results also indicate that rats exposed to fentanyl display increased amplitude of calcium-activated, BK, channel-mediated current in the IL, compared to yoked saline controls. Conversely, NAc showed no alteration of K⁺ channel activities. Calcium-activated SK channels, delayed rectifier K⁺ channels, A-type K⁺ channels, and two-pore domain K⁺ channels were unaffected in either region. Together, these results reveal a type-specific adaptation of K⁺ channels to fentanyl use behavior, which proposes a potential target for future treatment development.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

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Topic: H.08. Drugs of Abuse and Addiction

Support: F31DA054759
R01DA035217

Title: Burst firing by low voltage-activated T-type channels in the paraventricular thalamus influences somatic signs of opioid withdrawal

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Abstract: High rates of opioid abuse and overdose deaths represent a substantial public health issue in the United States. Avoidance of withdrawal symptoms is a primary driver of continued opioid use and overdose. Understanding the neuroadaptations induced by chronic opioid exposure holds great promise for the identification of novel and efficacious therapeutics. The paraventricular thalamic nucleus (PVT), located in the dorsal midline thalamus, encodes behavioral states relevant to drug addiction, including arousal, hunger, reward-seeking, and aversion. Recent studies have pointed to a critical role of the PVT and its projections in regulating many behavioral effects of opioids. The Mu-opioid receptor (MOR) is expressed in many brain regions, which collectively mediate many cellular and behavioral effects of opioids, but the extent to which the MOR in the PVT contributes to morphine withdrawal-induced somatic signs remains unknown. In addition, several important questions remain poorly understood. How does morphine exposure and subsequent withdrawal affect PVT action potential (AP) firing? To what extent do the changes in neuronal activity contribute to morphine withdrawal-induced somatic signs? Opioid withdrawal induces a wide array of somatic signs in

rodents including jumps, wet dog shakes, paw tremors, rearing, and grooming. Does the MOR in the PVT contribute to distinct behavioral phenotypes or merely the severity of somatic withdrawal signs? Using ex vivo brain slice electrophysiology in mice, we assessed how acute pharmacological activation of MOR in mouse PVT neurons stimulates hyperpolarization through G-protein coupled inward-rectifying K⁺ (GIRK) channels. Neuroadaptations due to chronic MOR activation led to a rebound increase in neuronal burst firing activity after withdrawal. Burst firing is regulated by T-type Ca²⁺ channels, which are activated then quickly inactivated by depolarization, and recover to the inactivated state during subsequent hyperpolarization. We determined whether a T-type Ca²⁺ channel antagonist reduced somatic withdrawal signs. This research will deepen our understanding of cellular and circuit mechanisms underlying adverse effects of opioid withdrawal.

Disclosures: V. Friedman: None. X. Liu: None. H. Yu: None. Q. Liu: None.

Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.25/KK13

Topic: H.08. Drugs of Abuse and Addiction

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Title: Preclinical investigation on biomarkers and therapeutic efficacy of deep brain stimulation for polysubstance use

Authors: *J. M. ROJAS CABRERA¹, S. VETTLESON-TRUTZA², K. SCHEITLER³, C. D. BLAHA³, Y. OH³, H. SHIN³, K. H. LEE³;

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Abstract: Polysubstance use, primarily stimulants like amphetamine adulterated with illicitly manufactured fentanyl, has led to an alarming rise in fatal overdoses. Their distinct pharmacologic actions complicate clinical management, whilst absence of real-time neurobiological monitoring during drug use limits the development of biomarker-informed interventions. To address this gap, we utilized the multifunctional apparatus for voltammetry, electrophysiology, and neuromodulation (MAVEN) platform which enables simultaneous local

field potential (LFP) acquisition and dopamine (DA) concentration recordings to monitor changes within key reward-processing brain areas in response to acute polysubstance use. Additionally, we tested the hypothesis that **deep brain stimulation (DBS) of the medial forebrain bundle (MFB)** mitigates the dopaminergic surge induced by polysubstance use, thereby mitigating a critical aspect of its addictive potential. **Methods:** A recording and stimulating electrode were implanted into the core of the nucleus accumbens and MFB, respectively, of urethane-anesthetized rats. In the drug-response group (n=6), basal extracellular DA levels were recorded pre- and post-drug administration (fentanyl, 10µg/kg + amphetamine, 2mg/kg, i.v.) with multiple cyclic square wave voltammetry (M-CSWV) while LFPs were recorded concurrently. In the DBS-group (n=5), MFB-DBS (130Hz, 0.5ms PW, 0.25mA) was continuously applied 10 mins pre- and 20 mins post-drug injection while tonic DA levels were recorded. **Results:** Accumbal DA significantly increased by 52.8 ± 12.1 nM after drug administration from baseline levels of 150 ± 34.2 nM. LFP recordings showed a significant decrease in theta (4-8Hz), beta (13-30Hz), and gamma (30-80Hz) power. MFB-DBS decreased DA levels by 11 ± 3.1 nM from baseline, while drug administration with concurrent DBS increased DA levels by 21.89 ± 2.2 nM. **Conclusion:** Our study reports potential neurochemical and electrophysiologic biomarkers for substance use disorders (SUD), particularly those involving polysubstance use. Furthermore, our exploratory use of DBS highlights its efficacy as an alternative therapeutic in cases of treatment-refractory SUD, although further preclinical behavioral work is warranted and ongoing.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR297.26/KK14

Topic: H.08. Drugs of Abuse and Addiction

Support: Intramural Research Program of the NIH

Title: Role of frontostriatal circuits in relapse to opioid seeking after voluntary abstinence

Authors: *Z. MA, Y. DUAN, S. LIN, H. NGUYEN, H. LU, Y. SHAHAM, Y. YANG; NIH, NIDA IRP, Baltimore, MD

Abstract: Introduction: High relapse rate during abstinence is a defining characteristic of drug addiction. We previously found that opioid seeking progressively increases after voluntary

abstinence induced by adverse consequences of oxycodone seeking (crossing an electric barrier). Functional MRI (fMRI) revealed that this effect is associated with changes in functional connectivity within medial orbitofrontal cortex (mOFC)- and dorsomedial striatum (DMS)-related circuits. However, as the resting-state functional connectivity results are correlational, understanding the causal roles of frontostriatal circuits in oxycodone seeking is needed for potential intervention.

Methods: In this study, we combined fMRI with either pharmacological manipulation or non-invasive transcranial magnetic stimulation (TMS) to study the causal role of frontostriatal circuits in oxycodone seeking after electric barrier-induced voluntary abstinence in rats. We first tested the effect of intracranial injections of the GABA_a and GABA_b receptor agonists muscimol and baclofen into either mOFC or DMS on oxycodone relapse (n=70 males, n=40 females). We then used fMRI to determine the effect of DMS inactivation on changes in regional cerebral blood volume (CBV) and functional connectivity of DMS. In addition, we investigated the effect of non-invasive TMS modulation of the frontostriatal circuits on relapse to oxycodone seeking (n=36 males).

Results: We found that inactivation of the DMS, but not mOFC decreased oxycodone seeking. Functional MRI data revealed that DMS inactivation decreased cerebral blood volume levels in DMS and several distant frontal cortical areas. Furthermore, functional connectivity of frontostriatal circuits significantly increased after DMS inactivation. In addition, non-invasive TMS modulation of the frontal cortex significantly reduced relapse to oxycodone seeking. Together, these results indicate a critical role of frontostriatal circuits in oxycodone seeking after voluntary abstinence, suggesting potential target for intervention.

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Poster

PSTR297: Opioids: Cell Signaling, Circuitry, and Neurophysiology

Location: SDCC Halls B-H

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Program #/Poster #: PSTR297.27/Web Only

Topic: H.08. Drugs of Abuse and Addiction

Support: NIH R01 DA015462

Title: Drug and money choice preference patterns in regular heroin users maintained on buprenorphine

Authors: *A. S. JHAND¹, M. K. GREENWALD²;

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Abstract: Background

About two-thirds of people treated for opioid use disorder (OUD) return to opioid use within the first-year post-treatment. Furthermore, many individuals continue to use illicit opioids while on

agonist therapy. Understanding determinants of opioid-seeking is vital to reducing recurrence of drug-use. In this work, we examined predictors of effortful responding for opioid and money rewards in a model simulating real-world choices.

Methods

We reanalyzed data from previously conducted controlled laboratory studies in which regular heroin-users were stabilized on buprenorphine 8 mg/day sublingual to suppress opioid withdrawal. During experimental sessions, participants could choose across 12 trials between units of hydromorphone (HYD, 1 or 2 mg IM) and money (\$2 or \$4). Reward amounts and ratios differed parametrically across sessions and studies. The effort required to earn each reward increased exponentially across trials. We examined individual differences between drug and money effortful responding (breakpoint difference) and latencies (choice speed difference).

Results

Faster latency (across within-session trials) to choose HYD (2 mg) vs. money (\$2) was associated with recent pre-experimental cocaine use ($n=25$ vs. no cocaine use, $n=9$; $p=0.016$), and choosing HYD on the first trial ($n=17$ vs. chose money first, $n=18$; $p=0.001$). Faster latency to choose HYD vs. money was associated with greater effortful responding for drug vs. money ($n=29$, $r^2=0.4$). Using regression analysis ($n=59$), initial choice for HYD ($p<0.001$, $r^2=0.5$) was a stronger predictor of greater effortful drug responding, compared to pre-experimental cocaine use ($p=0.37$) and relative reward magnitude (HYD ÷ money ratio, $p=0.35$). When predicting maximal effortful drug responding for a subset of subjects reserved for prediction ($n=20$), the regression model demonstrated acceptable fit (RMSE=0.32).

Conclusions

Our results highlight that participants who initially choose opioid over a monetary alternative and exhibit faster decisional speed for opioid than money engaged in more effortful opioid-seeking. These findings suggest that higher-magnitude contingency management coupled with cognitive therapies that promote deliberation could decrease opioid use.

Disclosures: A.S. Jhand: None. M.K. Greenwald: None.

Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.01/Web Only

Topic: I.02. Perception and Imagery

Support: CIHR Doctoral Research Award 181517

Title: Parkinson's Disease affects motor imagery of sequential actions: Evidence from neural oscillations

Authors: *K. J. M. LAMBERT¹, Y. Y. CHEN³, A. W. S. LEUNG¹, A. SINGHAL²;

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Abstract: Parkinson's Disease (PD) leads to significant deficits in the physical execution of action sequences. These deficits may stem in part from impaired processing of action representations, which are integral to the planning, monitoring, and execution of voluntary movement. Action representations can be examined using motor imagery. Here we examine whether PD affects imagery of action sequences at the level of behaviour and neural activity. In particular, 20 people with PD (6F; mean age = 67.85 Y; mean Montreal Cognitive Assessment score = 27.00) and 20 controls (7F; mean age = 66.00 Y; mean Montreal Cognitive Assessment score = 27.15) completed a motor imagery task while electroencephalographic activity was recorded. Our data analysis focused on activity in the mu rhythm (8-14 Hz) over the motor area, as suppression in this rhythm is associated with sensorimotor network activation, and the theta rhythm (3.5 - 7 Hz) over the temporo-occipital area, which is proposed to support sensorimotor integration. The PD group was significantly less accurate when imagining action sequences as compared to controls. Successful trials were accompanied by more mu suppression, with comparable levels of overall mu suppression between groups. Interestingly, the PD group exhibited significantly more theta activity over the right temporo-occipital region. This increase in theta oscillations may represent increased activity in the extrastriate body area, suggesting compensatory strategies employed by individuals with PD. People with PD and controls exhibit similar activation of the sensorimotor network during motor imagery. However, this activation may be less efficient in PD, leading to a reduction in accuracy when imagining action sequences and the compensatory engagement of visual regions. Taken together, the findings of the present study indicate that impaired processing of action representations may contribute to the sequencing deficits observed in PD.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.02/KK15

Topic: I.02. Perception and Imagery

Support: KAKENHI grant 24K14231

Title: Olfactory-visual cortical coupling during autobiographical memory: an EEG study

Authors: *Y. MASAOKA¹, M. MATSUI², N. KOIWA³, M. HONMA², A. YOSHIKAWA⁴, M. IZUMIZAKI²;

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Abstract: Specific odors are known to evoke vivid autobiographical and episodic memories. Our previous functional magnetic resonance imaging (fMRI) study demonstrated that odors linked to autobiographical memories (AM) elicited significant activations in the orbitofrontal cortex, hippocampus, parahippocampal gyrus, and visual cortex. Notably, the degree of activation in the visual cortex positively correlated with the intensity of memory retrieval and the subjective experience of mental time travel. These findings suggested that AM-related odors may induce visual imagery, thereby reactivating occipital regions. In the present study, we employed electroencephalography (EEG) to further examine the neural dynamics underlying odor-evoked autobiographical memory, with a particular focus on the visual cortical areas. EEG data were preprocessed using EEGLAB (MATLAB), and spectral power analyses were conducted using nonparametric cluster-based permutation tests implemented in the FieldTrip toolbox. This statistical approach, based on the method described by Pizzamiglio et al. and Maris and Oostenveld, effectively controls for multiple comparisons in time-frequency domain analyses. Our results revealed a significant increase in beta-band (15-30 Hz) activity in the posterior visual cortex during presentation of AM-associated odors, compared to control odors without memory relevance. This enhancement of β oscillations suggests heightened visual cortical engagement, potentially reflecting the generation of odor-induced visual imagery during memory recall. Previous animal research has reported that 20-40 Hz oscillatory activity emerges in the entorhinal cortex during odor-guided spatial memory tasks. These findings, together with our results, suggest that functional coupling between the entorhinal and visual cortices may be enhanced during odor-evoked retrieval of spatially embedded autobiographical memories. This study was approved by the Ethics Committee of Showa University School of Medicine and conducted in accordance with the Declaration of Helsinki (approval number: 3327). The authors declare no conflicts of interest.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.03/KK16

Topic: I.02. Perception and Imagery

Title: Self-efficacy as a potential factor influencing both implicit and explicit sense of agency.

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Abstract: Sense of agency (SoA) constitutes a fundamental aspect of human daily subjective experience, without which humans would be like philosophical zombies (Chalmers, 1996), or mere observers of the world, devoid of the feeling of interacting with the environment. The concept of SoA typically encompasses the range of feelings ‘before’, ‘during,’ and ‘after’ an action. Studies on SoA have predominantly focused on the ‘during’ phase of an action (Haggard, 2017). This study, in contrast, focused on the feeling before an action, particularly the confident feeling of ‘I will get my desired outcome’ here referred to as ‘self-efficacy.’ Self-efficacy reflects the subjective sense that ‘I can manage my actions and make what I want to happen’. Intuitively, actions carried out with this feeling are likely to be accompanied by a strong SoA. We conducted a series of experiments to investigate whether such a connection could be found. Simple gambling tasks were employed, which were designed to make participants experience different levels of self-efficacy depending on payoff probabilities. Four identical options, or one option yielding a reward with a 0.25 probability, were presented to the participant. The participant could bet to open a box for a reward by paying a cost or decline to bet without any costs. There were alternatives to the above conditions, in which the options were given a color coding to indicate high and low probabilities. The color matching was learned before the experiment. We presupposed that when an option indicating high probability was shown, or when four boxes were presented, the participant would feel relatively higher self-efficacy than in the other cases. Following the presentation of the options, the participant reported the degree of self-efficacy and then selected one option by pressing the target button. The outcome was delivered after a delay of 100-900 ms. The participant was asked to report the degree of SoA (explicit measure of SoA) and to evaluate the delay (implicit measure of SoA). Self-efficacy partially affected explicit and implicit SoA. We also analyze the relationship between implicit and explicit measures of SoA, some of which were found to be inconsistent with previous research (Imaizumi, Tanno, 2019; Schwarz et. al., 2019).

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.04/KK17

Topic: I.02. Perception and Imagery

Support: NIH R01 MH131595

Title: The neural mechanisms of aligning spatial perspectives

Authors: *S. O. MURRAY, B. LEONARD, K. WOODARD, J. A. PYLES, A. STARR;
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Abstract: The ability to adopt a spatial perspective different from one’s own is crucial for navigation, social interaction, and collaborative behavior. Visual perspective-taking (VPT) tasks

require understanding how objects appear from another viewpoint and presumably depend on mental rotation processes. A central debate is whether viewer-centered transformations involve abstract spatial computations or embodied sensorimotor simulations. To address this, we used functional MRI while healthy adults ($N = 24$) decided whether a cube was left or right from the standpoint of either an avatar or an empty chair, across systematically varied angular perspective alignments (0° , fully aligned, to 180° , fully unaligned, in 60° steps). Reaction times (RTs) rose approximately linearly with angular differences in perspective (slope = 1.10 ms/deg, $p < 10^{-9}$), indicating increasing mental rotation demands. Whole-brain general-linear modelling revealed activity that scaled with angle in dorsal premotor cortex/area 6a, supplementary-cingulate eye fields (24dv/SCEF), inferior frontal operculum (FOP4), intraparietal sulcus (IPS1/LIP), and cerebellar lobule VI/Crus I. Moreover, individual differences in fMRI magnitude in these regions predicted individual differences in the magnitude of RT increases. Responses did not differ between avatar and chair conditions, indicating that social inference was unnecessary. Functional localizer scans further revealed activations in body-selective regions in parietal cortex. Together, the data show that VPT recruits a motor-parietal-cerebellar network in direct proportion to the mental rotation required, supporting embodied—rather than purely geometric—accounts of perspective alignment.

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Poster

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Topic: I.02. Perception and Imagery

Support: Vision Science Training Program: Cornea to Cortex training grant (5T32EY015387-22)
National Science Foundation, Mind, Machine, and Motor Nexus program grant (221190)

Title: Measuring Intention - A Path Forward in Intentional Binding and the Sense of Agency

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Abstract: The sense of agency is the perceptual experience of authoring our volitional actions. It offers a valuable framework for assessing perceptions of control and the quality of interaction for human-in-the-loop systems. Intentional binding (IB) is a quantitative measure of the sense of

agency where individuals perceive temporal compression between a volitional action and its sensory outcome. Recent studies have questioned the validity of IB, claiming that binding could arise due to causal expectations. We ran 38 participants through a forced-two-choice psychophysics experimental paradigm and controlled sensory outcomes between intentional and unintentional movements. Participants discriminated time durations between pairs of auditory tones coinciding with paired active and passive movements (experimental group, N = 19) or two passive movements (control group, N = 19) executed on a Kinarm. Our results showed that there were no differences in time compression biases between the experimental and control groups - supporting recent literature that intentional binding is independent from movement intention. Furthermore, we found that the temporal precision between the groups was significantly different - a novel metric we term **intention consistency**. Intention consistency links intention and action; it is a direct measure for the internal sense of agency. When intention consistency is combined with causal binding, we can measure outcome intention: the external sense of agency. Prior work has shown that the planning and execution of motor plans in the pre-Supplementary Motor Area and the Supplementary Motor Area can lead to an increase in temporal precision. Future work should investigate differences in motor cortex areas involved with intentional and unintentional motion to further validate our results. This work can be used to measure intention in human-in-the-loop systems including assistive devices, exoskeletons, tele-operated robotics, semiautonomous vehicles, AI chat agents, and other human input dependent technologies.

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Poster

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Location: SDCC Halls B-H

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Program #/Poster #: PSTR298.06/KK19

Topic: I.02. Perception and Imagery

Support: CREST Grant Number JPMJCR23P1

Title: Factors Associated with Changes in Older Adults' Sense of Agency: An Exploratory Study

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Abstract: The sense of agency (SoA), the experience of controlling one's body and environment, is known to be altered in older adults compared to younger adults; however, the underlying factors remain unclear. This study examined whether SoA in older adults is associated with the amount of activity and extent of care. Participants were 21 older adults requiring care (85.7±6.8 years), 12 healthy older adults (80.6±5.8), and 13 younger adults

(29.7±6.1). The amount of activity was assessed by sedentary time (≤ 1.5 METs), low- (1.6-2.9 METs), and moderate-to-high intensity (≥ 3.0 METs) activity. The extent of care was assessed using care level, independence in daily living, and the Functional Independence Measure (FIM). SoA was measured using a dot-control task on a laptop touchpad, where participants' input was blended with prerecorded movements in 10% increments across 11 conditions (0-100%). Each participant completed 110 trials, indicating "Yes" or "No" to whether they felt in control. Logistic curves were fitted to "Yes" response rates to compute slope (sensitivity) and point of subjective equality (PSE; judgment criterion). Associations between SoA indices and the amount of activity and extent of care were examined using frequentist and Bayesian correlation analyses. In the care group, frequentist results (Spearman's r , 95% CI) showed significant correlations between slope and the amount of activity: sedentary time ($r = -0.74$, CI: -0.94 to -0.39), low- ($r = 0.75$, CI: 0.37 to 0.94), and moderate-to-high intensity activity ($r = 0.63$, CI: 0.21 to 0.90); and the extent of care: FIM ($r = 0.61$, CI: 0.17 to 0.84) and independence in daily living ($r = -0.69$, CI: -0.84 to -0.40). PSE also correlated with FIM ($r = 0.65$, CI: 0.27 to 0.83) and care level ($r = -0.58$, CI: -0.80 to -0.22). In the healthy group, only the correlation between PSE and moderate-to-high intensity activity was significant ($r = 0.72$, CI: 0.48 to 0.96). Bayesian results (posterior mean, 95% CrI) in the care group showed similar associations: slope with sedentary time (-0.54, CrI: -0.79 to -0.20), low- (0.50, CrI: 0.15 to 0.77), and moderate-to-high intensity activity (0.39, CrI: 0.02 to 0.70); and with FIM (0.44, CrI: 0.12 to 0.70) and independence in daily living (-0.52, CrI: -0.75 to -0.22). PSE was also moderately associated with FIM (0.40, CrI: 0.09 to 0.66) and independence in daily living (-0.36, CrI: -0.63 to -0.04). These findings suggest that SoA in older adults is linked to both the amount of activity and extent of care, and that the patterns of association may differ between older adults requiring care and healthy older adults.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

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Program #/Poster #: PSTR298.07/KK20

Topic: I.02. Perception and Imagery

Support: CAS Grant XDB1010302
CAS Grant YSBR-041
CAS Grant 173211KYSB20200021

Title: Self-organized context dependent processing in neural networks

Authors: *G. HAO;
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Abstract: Context dependent processing (CDP) enables flexible responses to stimuli based on varying circumstances. It is a crucial aspect of higher cognition that supports primates' adaptive behavior in dynamic environments. However, the structural requirements for its emergence remains poorly understood. The current study bridges this gap by investigating what network architectures enable the spontaneous development of CDP in artificial neural networks (ANNs). We demonstrate that networks with two parallel pathways that merge via cross-product self-organize towards CDP (we termed SOCDP), with spontaneously emerging functional modules for processing contextual and sensory information, respectively, mirroring the key aspects of CDP in the primate brain. Further analysis revealed that the pressure to efficiently navigate high-dimensional information space—a challenge akin to overcoming the curse of dimensionality—is the driver behind the SOCDP. Furthermore, we identified crucial factors such as network size, structural asymmetry, and task complexity in influencing the emergence of functional specialization in the system. These results suggest that CDP can arise spontaneously given proper network structural predisposition, providing insights into both the computational principles underlying flexible cognition and the design of more adaptable artificial intelligence systems that can adapt to more dynamic environments.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

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Topic: I.02. Perception and Imagery

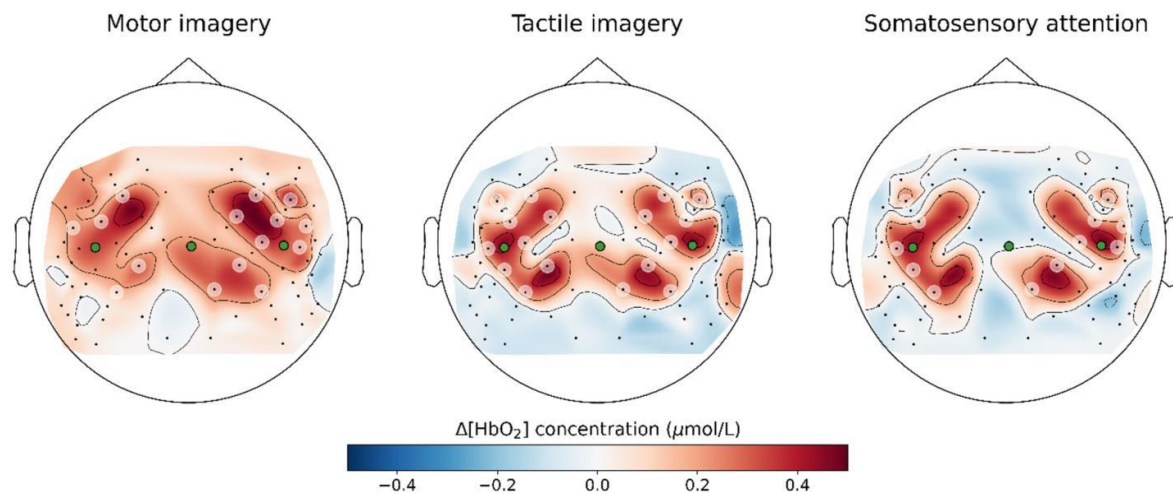
Title: Conscious control of cerebral hemodynamics through somatosensory attention, motor and tactile imagery: a functional near-infrared spectroscopy study

Authors: *A. KAPLAN¹, L. YAKOVLEV¹, N. SYROV¹, T. GATINSKAYA², N. KOMAROV², A. MIROSHNIKOV¹;

¹Biol. Fac., Lomonosov Moscow State Univ., Moscow, Russian Federation; ²SITA Sarl, Lausanne, Switzerland

Abstract: Conscious control of blood oxygen saturation in specific areas of the cerebral cortex is currently an underexplored resource for the targeted optimization of human cognitive abilities and stress resistance. One effective method for activating brain hemodynamics is through the mental representation of images associated with particular cognitive activities. Utilizing functional near-infrared spectroscopy (fNIRS), we examined hemodynamic responses (HRs) during kinesthetic motor imagery, tactile imagery, and somatosensory attention to determine how these distinct voluntary cognitive processes modulate hemodynamics in the sensorimotor cortex in the absence of overt movement or sensory input. The study involved 19 healthy volunteers (9 males, average age 24±5 years) and employed 74 fNIRS channels. Each participant completed 20 sensorimotor trials and 20 rest trials, each lasting 15 seconds, presented in random order. We

selected precentral and postcentral regions of interest (ROIs) using five channels each, corresponding to the locations of motor and somatosensory cortical regions, with the aid of the fOLD toolbox [Z. Morais et al. Scientific Reports, 2018, 8(1), 3341]. The main conclusions are summarized in the figure (see below): Grand average topographic projections of ΔHbO_2 . Gray transparent dots indicate statistically significant channels based on permutation t-tests (50,000 permutations, p-threshold = 0.0005). Green channels mark the reference locations of the C3, Cz, and C4 leads. 1. Motor imagery elicits similarly pronounced HRs in both precentral and postcentral ROIs in the contralateral hemisphere, while the ipsilateral hemisphere shows greater precentral activation; 2. Tactile imagery and somatosensory attention induce weaker precentral responses compared to motor imagery; 3. HRs exhibit a bilateral central distribution, particularly during tactile imagery. Thus, this study demonstrates that by manipulating mental images, it is possible to consciously control blood oxygen saturation in differentiated cortical areas of the brain.



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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.09/KK21

Topic: I.02. Perception and Imagery

Support: National Science and Technology Innovation 2030 Major Program (2022ZD0204802)
National Natural Science Foundation of China (T2421004, 31930053, 32400874)
Beijing Natural Science Foundation (5244044)

Title: Neural representation of the subjective uncertainty of prediction in the human brain

Authors: *T. HE¹, X. GONG², F. FANG²;

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²Peking Univ., Beijing.

Abstract: Prediction plays a crucial role in various cognitive processes, and numerous studies have successfully decoded the content of predictions in different domains. However, predictive processes inherently involve uncertainty, a subjective experience in making predictions. To date, the neural representations of the prediction uncertainty, which are essential for cognition and survival, have yet to be fully understood. In the current study, we recorded magnetoencephalography (MEG) signals and applied a generative model to investigate the neural representations of both the prediction content and its associated subjective uncertainty in humans. We found that trial-by-trial decoding errors, the difference between predicted and true target locations, were positively correlated with behavioral errors. Importantly, the decoded subjective uncertainty predicted participants' behavioral uncertainty in the predictive process. Furthermore, we characterized the temporal dynamics of the correlation between the decoded and behavioral uncertainty, showing that the decoded uncertainty tracked behavioral uncertainty over time. Together, our findings demonstrate that population activity in the human brain encodes not only the objective content of predictions but also the subjective uncertainty associated with them, both of which jointly guide predictive behavior.

Disclosures: T. He: None.

Poster

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Location: SDCC Halls B-H

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Topic: I.02. Perception and Imagery

Support: National Science and Technology Innovation 2030 Major Program
2022ZD0204804
National Natural Science Foundation of China 32200862, 32441106,
T2421004, 31930053, 32171039
High-performance Computing Platform of Peking University

Title: Learning Time by Touch: Plasticity and Generalization of Tactile Duration Discrimination Across Hands

Authors: *Y. GAO¹, H. QIN¹, J. LU², G. CHEN², Q. WANG²;

¹Sch. of Humanities and Social Sci., Beihang Univ., Beijing, China; ²Peking Univ., Beijing, China

Abstract: Precise estimation of subsecond durations is essential for perceptual organization, sensorimotor coordination, and interaction with dynamic environments. Extensive studies have revealed that temporal perception in visual and auditory modalities can be improved through

training, suggesting experience-dependent plasticity in our internal sense of time. However, whether similar plasticity exists in the tactile domain, and to what extent such learning generalizes across effectors, remains largely unknown.

To address this, participants were trained to perform a tactile duration discrimination task over five consecutive days. During training, participants received pairs of vibrotactile stimuli delivered to the index finger of either the dominant or non-dominant hand, and they were asked to make a two-alternative forced choice (2-AFC) judgment indicating which stimulus lasted longer. To assess learning and generalization, duration discrimination thresholds were measured on both hands before and after training.

Results showed that participants in both the dominant-hand and non-dominant-hand training groups exhibited significant improvements in temporal sensitivity on the trained hand.

Interestingly, we observed an asymmetric pattern of intermanual transfer: training the dominant hand led to modest improvement on the trained hand but resulted in complete transfer to the untrained, non-dominant hand. In contrast, training the non-dominant hand elicited greater improvement on the trained hand, but only partial transfer to the dominant hand. This asymmetry suggests that tactile perceptual learning of duration engages central timing mechanisms whose representations are modulated by sensorimotor dominance and task execution proficiency.

These findings provide behavioral evidence for the plasticity of tactile temporal perception and highlight how central-peripheral interactions support the generalization of learned temporal representations across effectors, offering insights into how the brain refines time-based judgments through experience.

Disclosures: Y. Gao: None. H. Qin: None. J. Lu: None. G. Chen: None. Q. Wang: None.

Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.11/LL1

Topic: I.02. Perception and Imagery

Support: NIMH R01 MH121520
CAMH Koerner Graduate Award

Title: Structural integrity of the vertical occipital fasciculus modulates face processing in body dysmorphic disorder

Authors: *J. P. DIAZ-FONG¹, N. YANG¹, V. GRACIE², H. J. PEEL¹, J. D. FEUSNER¹;

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Abstract: The vertical occipital fasciculus (VOF) is a white matter fiber bundle connecting dorsal and ventral visual systems, supporting global and local processing of complex visual stimuli. Disruptions in this integration may underlie perceptual biases in both neurotypical

individuals and clinical populations. We examined whether individual differences in VOF microstructure are associated with performance on the face inversion effect (FIE) in healthy controls and individuals with body dysmorphic disorder (BDD), a psychiatric condition characterized by distorted self-perception of appearance concerns that are minor or unnoticeable to others. We hypothesized that VOF microstructural integrity would show a stronger association with global interference during face inversion in controls than in BDD. Two independent datasets were analyzed: (i) 16 individuals with BDD and 19 controls completed a sequential matching task, judging whether two sequentially presented upright or inverted face stimuli were the same or different; (ii) 28 individuals with BDD and 28 controls completed a two-alternative forced choice (2AFC) match-to-sample task, selecting which of two simultaneously presented faces (upright or inverted) matched a previously viewed target face. The 2AFC task included both fast (0.5 s) and slow (5 s) stimulus presentation conditions. FSL's XTRACT tool was used to isolate the VOF from diffusion MRI, with microstructure indexed by mean fractional anisotropy (FA). We focused our analysis on the right VOF, as face processing is typically right hemisphere dominant. Response times (RTs) were analyzed using a linear mixed-effects model with group, orientation, and mean FA as predictors. Significant Group $\times$ Orientation $\times$ FA interactions were observed in both datasets. In the sequential matching task ($\beta = -.093$, $p = .024$), higher FA was associated with faster RTs in controls but slower RTs in BDD. In the 2AFC task, this interaction emerged only under fast presentation conditions ($\beta = -.184$, $p = .018$), suggesting that right VOF microstructure plays a role in early stages of face processing. These findings indicate that dorsal-ventral connectivity via the right VOF differentially modulates the FIE in BDD compared to controls. As expected, greater right VOF microstructural integrity was associated with increased global interference (larger inversion effect) in controls, whereas this relationship was reversed in BDD. These findings underscore the role of the right VOF in supporting the integration of global and local visual information during face perception, with distinct patterns in those with and without distorted self-perception of appearance.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

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Program #/Poster #: PSTR298.12/LL2

Topic: I.02. Perception and Imagery

Support: NIH Grant R37MH076136

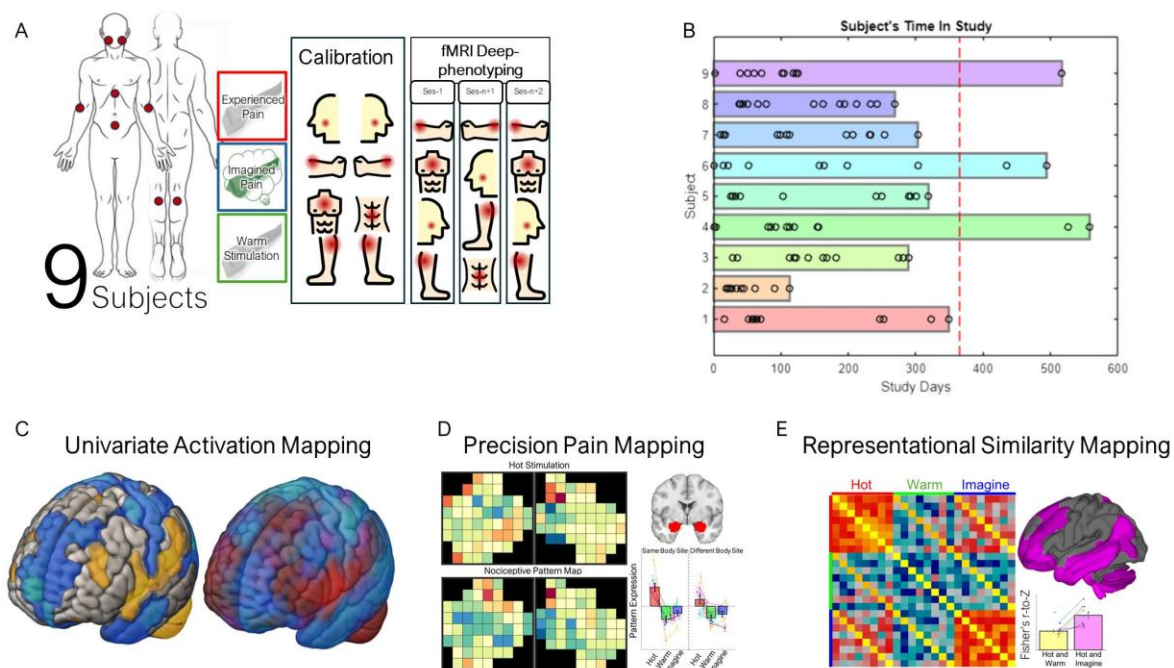
Title: Precision Functional Mapping of Imagined and Experienced Pain

Authors: *M. SUN¹, B. PETRE², K. BO³, C. I. BANGO⁴, H. JUNG⁶, T. D. WAGER⁵;

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Brain Sci., Dartmouth Col., Lebanon, NH; ⁴PBS, ⁵Psychological and Brain Sci., Dartmouth Col., Hanover, NH; ⁶Stanford Univ., Stanford, CA

Abstract: Imagination is a principal human capacity, conferring the ability to simulate sensations and situations that do not exist. We employed precision functional mapping of neural patterns underlying experienced and imagined pain across eight body sites, using fMRI in over ten hours of scanning for each participant (N = 9). Through univariate contrasts, pattern classification, and representational similarity analyses, we demonstrate that imagined and experienced pain co-activated nociceptive regions (e.g., thalamus) and non-nociceptive regions (e.g., supplemental motor area), but imagination did not re-activate somatotopic-specific nociceptive patterns reliably in any region or the Neurologic Pain Signature. Instead, imagined and experienced pain shared common representations across widespread cortical areas, particularly transmodal cortical areas such as dorsomedial prefrontal cortex, hippocampus, and BA47, but also subcortical areas such as the thalamus. Our findings suggest that imagination serves as a sophisticated cognitive simulation system, creating pain-like activity patterns across much of the brain. However, imagination does not activate somatotopic nociceptive representations, allowing the brain to differentiate real from simulated experience, but increasing the intrinsic difficulty of conjuring vivid, detailed mental images of pain.



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Poster

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Location: SDCC Halls B-H

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Program #/Poster #: PSTR298.13/LL3

Topic: I.02. Perception and Imagery

Support: NRF-2022R1A2C3008991
Singularity Professor Research Project of KAIST

Title: Aesthetic preference as an emergent property of hierarchical neural networks

Authors: *A. SONG^{1,2}, M. KANG², S.-B. PAIK²;

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Abstract: The golden ratio (GR) has been recognized for centuries for its presence in nature and art, and is often considered a hallmark of objective beauty. Both human preference and neural sensitivity to GR (Bartolo, 2021; Rizzolatti, 2007), as well as sensitivity to visual ratios in infants and non-human animals (McComb, 1994; Pallet, 2010) have been frequently reported. These findings suggest that a preference for the GR may arise independently of visual experience. However, no mechanism has yet been proposed to explain whether or how such a preference could emerge. To explore this, we first conducted a behavioral experiment to confirm the tendency for GR-based selection. Participants were shown GR and non-GR face images and asked to choose the ones they found more appealing. Results revealed a stronger preference for GR-structured faces compared to original versions. Given that the GR is defined as the limiting ratio of the Fibonacci sequence, we hypothesized that a preference for GR might emerge in feedforward neural networks through stochastic wiring processes. To test this, we designed a hierarchical network model inspired by early visual processing. Units in the input layer had Gaussian receptive fields (RFs), representing unit-length sampling akin to retinal encoding. In subsequent layers, RFs were sparsely combined through local convergence, allowing deeper units to integrate over larger spatial scales. These structural constraints reflect biologically inspired features such as local integration and sparse connectivity. We found that pairwise RF length ratios clustered around the GR, suggesting that sensitivity to GR can emerge intrinsically from the network architecture. This clustering effect was absent in fully connected network configurations, highlighting that biologically plausible wiring rules are crucial for enabling GR tuning. These findings suggest that aesthetic preferences may spontaneously emerge from hierarchical neural architectures in the brain.

Disclosures: A. Song: None. M. Kang: None. S. Paik: None.

Poster

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Program #/Poster #: PSTR298.14/LL4

Topic: I.02. Perception and Imagery

Support: NRF-2023R1A2C2007289

Title: Microsaccade Rates Increase Under High Visual Processing Load in Syncopated Rhythm Reading

Authors: *H. KIM, C.-Y. KIM;

Sch. of Psychology, Korea Univ., Seoul, Korea, Republic of

Abstract: Visual rhythm reading requires fine-grained temporal processing. Among musical rhythms, syncopated rhythms violate metrical regularity and disrupt internalized beat expectations, making them more difficult to process especially during silent reading without auditory feedback (Kim & Kim, 2023). We investigated whether the increased processing demand of syncopated rhythms is reflected in ocular indices, particularly microsaccades — i.e., small, involuntary eye movements occurring during fixation. Previous studies have shown that microsaccade rates vary with task demands, decreasing under sustained internal attention (Siegenthaler et al., 2014) and increasing when stimuli are visually complex or temporally unpredictable (Martinez-Conde et al., 2006). We measured microsaccades during the rhythm reading under central fixation without requiring overt search. We hypothesized that syncopated rhythms would elicit higher microsaccade rates due to greater visual processing load. Upon each trial, participants fixated on a central cross, under which four quarter notes appeared on a CRT monitor. Prior to the rhythm presentation, they tapped at a comfortable pace to establish their internal beat. Then, a visual rhythm — either metrically regular (non-syncopated) or syncopated—was presented for 300 ms, while participants continued to fixate on the cross. After the rhythm disappeared, they reproduced it by tapping the spacebar. Microsaccades were recorded using an eye tracker (Eyelink 1000, SR Research) and analyzed with participant-specific velocity thresholds. Results showed that overall microsaccade rates were significantly higher in the syncopation condition compared to the non-syncopation condition. Time-resolved analysis further revealed a significant increase between 10 and 50 ms from the rhythm onset ($p=0.03$). These results show that microsaccade rates are modulated by visual processing load in reading syncopated rhythm. The temporal characteristic of the signal suggests an early stage of processing even when visual input is structurally complex and temporally unpredictable.

Disclosures: H. Kim: None. C. Kim: None.

Poster

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NSF-BSF BCS2241417
ONR (N00014-24-1-2026)

ONR (N00014-19-1-2029)
DARPA (D19AC00015)

Title: Performance decreases for untrained orientation observed in dominant computational models but not humans are mitigated by divisive normalization in encoding processes of visual perceptual learning

Authors: *Y.-A. CHENG, Y. SASAKI, T. SERRE, T. WATANABE;
Brown Univ., Providence, RI

Abstract: Visual perceptual learning (VPL) refers to long-term performance improvements following visual experience. It is controversial whether VPL arises from plasticity at the level of neural encoding processes or downstream readout processes (Watanabe & Sasaki, 2015, Ann. Rev. Psych.; Doshier & Lu, Perceptual Learning, MIT Press, 2020). To address this issue, we compared how training on an orientation detection task alters performance in humans and in a well-established neural network model of the early visual cortex, which includes broad excitation, untuned inhibition, and readout components. We focused on performance changes in untrained orientations, which have been discussed only relative to the trained orientation. In the psychophysical experiment with humans, as predicted, the greatest performance enhancement was observed at the trained orientation. Enhancements tapered off as orientations deviated further from the trained one, until around 90 degrees, where no performance increase was observed. Performance at orientations approximately 90 degrees from the trained orientation remained unchanged. In the neural network model simulation, plasticity in the readout components always led to unexpected performance decreases at untrained orientations. However, the introduction of plasticity in untuned inhibition during neural encoding processes, leading to a divisive normalization effect, mitigated the performance decreases at untrained orientations. These findings suggest that divisive normalization plasticity may resolve the discrepancy between the results from human psychophysics and the initial computational model. Our results further suggest the involvement of neural encoding processes in VPL.

Disclosures: Y. Cheng: None. Y. Sasaki: None. T. Serre: None. T. Watanabe: None.

Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

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Program #/Poster #: PSTR298.16/LL6

Topic: I.02. Perception and Imagery

Support: NIH R01 MH132225
NIH R01 MH107797
NSF 1734907

Title: A Naturalistic Paradigm for Studying Food Cognition Using Intracranial EEG (iEEG) and Eye-Tracking Data

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Abstract: The most common modality for studying food cognition is fMRI, in which the neural activity of subjects is recorded while they view images of food and / or have flavored solutions delivered to their tongue. This experimental paradigm is able to capture neural correlates for some of the relevant aspects of food cognition, like visual representation and taste quality. However, the context this experimental paradigm creates is dramatically different from the natural context in which humans interact with food. It is well-known that context and behavior can modulate the neural activity underlying cognitive processes, thus key aspects of food cognition cannot be fully explored in these artificial settings. To fully characterize the neural correlates underlying food cognition requires methods that allow researchers to extract meaningful signal from data collected in real-world settings - for example, while a subject eats a meal. In this study, we sought to demonstrate the feasibility of such a real-world experimental paradigm for studying food cognition by training a classifier to predict whether or not a subject was looking at a food object using a noisy, real-world dataset. We collected over an hour of iEEG and eye-tracking data from an epilepsy patient while he ate a meal. The egocentric video was annotated with the subject's eye position and object labels generated by the object-tracking algorithm BoT-SORT. A "fixation" was defined as a period in which the subject's gaze remained on an object for longer than 300 milliseconds. A subset of these fixations were divided into two classes: 215 in which the subject was looking at a food object (positive) and 215 in which the subject was looking at a non-food object (negative). The 300 ms of neural data associated with these examples was then used to train a linear classifier (glmnet) using five-fold cross-validation. After training the model, the weights were extracted and analyzed to determine the electrodes and corresponding brain regions that contributed most to the model's performance. Our model was able to achieve fairly high accuracy (0.695) with a large effect size (0.961). Furthermore, analysis of the extracted model weights revealed that brain areas such as the insula, fusiform visual cortex, and the inferior temporal cortex - areas well-known to be involved in food cognition and object recognition - contributed most strongly to the performance of the model. The results of our study represent a proof-of-concept that meaningful signal related to the visual perception of food can be extracted from noisy datasets in real-world settings.

Disclosures: K. Chetnik: None. A. Alreja: None. A.S. Ghuman: None.

Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.17/LL7

Topic: I.02. Perception and Imagery

Support: European Research Council (ERC Starting Grant 101039152)

Title: The psychophysiology of human (self-) tickle sensations

Authors: *A. H. HOFFMANN¹, W. PLÉTU¹, K. KILTENI^{2,1};

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Abstract: Tickling is a common form of social play among humans and other primates. Despite its familiarity, the full range of responses to tickling remains poorly characterized, and it is still unclear how these responses change when individuals attempt to tickle themselves. To explore these questions, we developed a robotic device capable of applying controlled strokes to the foot sole with varying velocities and forces. The setup also enables self-tickling: participants draw on a digitizing tablet, and the robot replicates these movements on their foot sole. In experiment 1 (N=20), we address how people experience external ticklish sensations on their foot soles. We collected both explicit ratings of ticklishness and emotional experience (arousal, pleasantness, unpleasantness), as well as implicit psychophysiological (skin conductance, respiration, heart rate, facial EMG) and behavioral markers (audiovisual recordings). Our results show significant positive relationships between perceived ticklishness and arousal. Strikingly, stronger ticklishness was associated with more ambivalent emotions - simultaneously high pleasantness and unpleasantness ratings. Perceptual ratings also correlated with physiological indicators of arousal (heart and respiration rate, skin conductance) and unpleasantness (corrugator supercilii muscle activity), suggesting that tickling induces high-arousal states but ambivalent valence. Building on these findings, in experiment 2 we address how people experience self-applied tactile sensations on their foot soles. Specifically, we aim to understand why reactions to ticklishness (e.g. laughter, arousal) are suppressed when people try to tickle themselves. First, we introduce spatiotemporal mismatches (e.g., delays or spatial rotations) between their hand movement on the tablet and the touch on their foot sole. Pilot data (N=7) show that participants reliably detect such mismatches, and that the perceived causality between the movement and the touch decreases with greater perceived temporal and spatial deviations. Next we will examine how these changes in perceived causality influence perceptual and physiological responses to ticklish sensations, as those observed in experiment 1, using the Bayesian Causal Inference framework. Together, these two experiments will clarify how perceptual and physiological responses to tickling are shaped by the source of stimulation: whether it comes from the self or from others.

Disclosures: A.H. Hoffmann: None. W. Pléту: None. K. Kilteni: None.

Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

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Program #/Poster #: PSTR298.18/LL8

Topic: I.02. Perception and Imagery

Support: European Research Council (ERC) Starting grant TICKLISHUMAN 101039152
MSCA postdoctoral fellowship NeuroSelfTickle 101207511
NWO SSH Open Competition XS grant "The ticklish brain"

Title: EEG correlates of tickle perception in humans

Authors: *T. STEPHANI¹, K. KILTENI^{2,1};

¹Donders Inst. for Brain, Cognition, and Behaviour, Radboud Univ., Nijmegen, Netherlands;

²Dept. of Neurosci., Karolinska Inst., Solna, Sweden

Abstract: What makes a touch ticklish? Current theories stipulate that tickle reflects a discrete and ambivalent sensation tightly coupled to a drive to convulse and laugh. It typically arises when another person applies fast and strong repetitive pressure on specific areas of our body, such as the armpit, sides of the trunk, or the foot soles. However, the neural representation of ticklish sensations remains elusive. To establish a comprehensive neurophysiological model of tickle perception, we examined the neural dynamics of ticklishness using electroencephalography (EEG). Participants received tactile stimuli (i.e., strokes) at varying velocities, which had previously been shown to systematically differ in perceived ticklishness, applied to their foot soles using a robotic tactile stimulator. We examined whether the associated somatosensory neural responses differed between low and high perceived ticklishness (i) at touch onset, (ii) during, and (iii) after a stroke stimulus on the foot sole. To this end, event-related potential (ERP) analyses were conducted along with time-frequency decompositions in relation to a ~4.5 sec long back-and-forth stroke stimulus on the foot sole. A "post-stroke poke" stimulus after the tickle stimulus served to probe the system's state after the expected build-up of ticklishness. Moreover, we recorded muscle activity in the lower extremities using electromyography (EMG), electrodermal activity (EDA) at the fingertips, and face expressions using the Facial Action Coding System (FACS), to capture peripheral body behaviour related to ticklishness. At stimulus onset, ERP magnitude did not vary with perceived ticklishness. Crucially, ERP amplitudes after the application of a stroke decreased with higher ticklishness ratings. This suggests that the neural representation of ticklishness does not emerge immediately upon the stimulus' onset but builds over time, altering the somatosensory system's responsiveness to subsequent sensory input. Preliminary observations suggest an involvement of event-related alpha band activity in this dynamic build-up, which will be explored in further analyses. Altogether, these findings offer novel insights into the neural accumulation of repetitive somatosensory signals, leading - or not leading - to the enigmatic percept of ticklish touch. Eventually, understanding this mechanism may shed light on the neural basis of self-tickle cancellation (i.e., the inability to tickle oneself), a sensorimotor phenomenon that could have diagnostic implications for psychiatric and neurodevelopmental conditions such as schizophrenia and autism.

Disclosures: T. Stephani: None. K. Kilteni: None.

Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

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Program #/Poster #: PSTR298.19/LL9

Topic: I.02. Perception and Imagery

Support: European Research Council (ERC) Starting grant TICKLISHUMAN
101039152

Title: Characterizing and decoding tickle responses in humans: A multimodal approach

Authors: *Z. XIONG¹, T. STEPHANI¹, K. KILTENI^{1,2};

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Abstract: Tickling, a behavior commonly observed in social interactions already early in life, is a fundamental human experience that extends beyond laughter. Despite being so ubiquitous, its basic characteristics, including how a touch perceptually turns into a tickle and what psychophysiological responses define ticklishness, remain poorly understood. In three experiments (N = 36), we examined how the physical properties of a stroke applied on the foot sole determine its perceived ticklishness using a robotic haptic stimulation device. Experiment 1 revealed an inverted U-shaped relationship between stroke velocity and perceived ticklishness, with a moderate velocity (15 cm/s) rated as most ticklish, while both slower (1 cm/s) and faster (30 cm/s) velocities resulted in lower ratings. Experiments 2 and 3 replicated this pattern under varying stroke forces (0.5 N and 1 N), with the additional effect that higher forces increased ticklishness further. To capture the complex dimensions of tickle responses, we employed a multimodal approach, combining behavioral (facial and vocal expressions) and physiological measurements (electromyography, electrodermal activity, electrocardiogram, and respiration). Using support vector machines, we successfully decoded high versus low perceived ticklishness states within individuals (area under receiver operating characteristic curve, AUROC = 0.71) and across individuals (AUROC = 0.72), based solely on the collected behavioral and physiological data. Together, these findings reveal the stimulus properties and psychophysiological markers that constitute a ticklish touch, paving the way for further systematic investigation of the function of tickle perception in humans.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

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Program #/Poster #: PSTR298.20/Web Only

Topic: I.10. Human Learning and Cognition

Support: Co-Funded Brain Circulation Fellowship, 112C010
TUBA-GEBIP Award

Title: Experience-dependent changes in cortical function and structure following passive audiovisual associations

Authors: *F. KELES^{1,2}, P. DEMIRAYAK^{3,4}, H. KAFALIGONUL^{5,6};

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Abstract: Crossmodal associations play a critical role in shaping perceptual experience and guiding our actions. While such associations can be formed even through passive exposure to multisensory stimuli, the underlying neural mechanisms and experience-dependent reorganization of sensory processing remain largely unresolved. Using an audiovisual motion paradigm, we paired upward and downward visual motion (random dot kinematogram presented in the left visual field) directions with static tones of distinct frequencies. During the association phase, participants were passively exposed to these audiovisual pairings without any behavioral task or feedback. In the pre- and post-association test phases, we assessed the perceived direction of visual motion (with and without static tones), resting-state functional connectivity, and cortical thickness. After associations were formed between each motion direction and a static tone, the tones significantly altered perceived motion direction in line with the paired presentation during the association phase. More importantly, our findings indicated significant alterations in functional connectivity and cortical thickness. Functional connectivity between the stimulated visual regions in the right hemisphere (e.g., V1, V2, and V3) and higher-order frontal, temporal, and occipital cortices was increased following association. In contrast, the unstimulated (control) visual regions in the left hemisphere (e.g., V1, V2, and V3) showed much smaller changes. Additionally, area MT+ showed increased connectivity with occipital, temporal, and somatosensory areas. Consistent with the functional results, cortical thickness changes occurred exclusively in the right hemisphere and were also more pronounced in the association areas compared to low-level sensory areas. This pattern was consistent with reverse hierarchy theory, which posits that experience-dependent changes emerge earlier and more robustly in higher cortical areas before influencing primary sensory regions. Together, our findings highlight the brain's capacity to reorganize itself in response to multisensory input and sensory experiences. The findings also illustrate that audiovisual associations and sensory experiences formed without explicit feedback can lead to overlapping functional and structural changes in the adult human brain, providing important implications for perception and neural plasticity in daily life situations.

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Poster

PSTR298: Agency, Multisensory Experience, and Cognitive Integration

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR298.21/LL10

Topic: E.08. Multisensory Integration

Support: NIBIB R01EB026549

Title: Common representations underlie idiosyncratic neural topographies

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Abstract: Primary sensory brain areas contain innate stereotyped topographies like retinotopy and somatotopy, but association cortex shows more idiosyncratic organization. Using a twin sample from the human connectome project (N=988) and functional magnetic resonance imaging, we test the hypothesis that idiosyncrasies are the byproduct of convergent learning and subserve common functional representations.

We combined representational similarity analysis, full brain neuromaps and twin heritability models to investigate the spatial profiles and sources of representational and topographic similarity across diverse task conditions (emotion, gambling, language, social, relational, motor, working memory) and at rest. After parcellating the brain into discrete regions (N=518) we estimated regional task evoked responses and between-task representational distance matrices (RDMs), and equivalent metrics for individualized resting state network time-series. We found common representational geometry (high $\cos\theta$ of RDMs) with idiosyncratic topographies (low $\cos\theta$ of spatial patterns) in both task evoked responses and resting state network organization. Common representations with idiosyncratic topographies did not show consistent associations with a priori neuromaps of genetic or developmental markers (Markello, et al. [2022] Nature Methods), only network hierarchy, and were especially common in transmodal brain areas (association of topography with hierarchy is more negative than geometry and hierarchy, task geometry > topography: $\beta_{\text{std}} = 0.131$, $p = 0.0010$, rest geometry > topography: $\beta_{\text{std}} = 0.154$, $p = 0.0004$, permutation “spin” test). Additionally, while topography was heritable, response geometry was not ($h^2 = 0.04 \pm [0.02, 0.06]$, $CI_{.95}$, family-wise BCa bootstrap, additive genetic, common and unshared environment, ACE model), and may instead be learned.

Our results show that association cortex exhibits substantial idiosyncratic topographic organization but nonetheless supports a convergent representational code across individuals. Additionally, genetic influences significantly constrain cortical topographies yet appear to have less impact on emergent representational geometry, suggesting that higher-level functional organization is more plastic. Together, these findings show how similar functional solutions can emerge despite individual differences in spatial organization of the brain.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR299.01/LL11

Topic: I.03. Decision Making

Title: Sex dependent effects of cannabinoid drugs on punishment-driven risky decision-making

Authors: *S. C. LOWE¹, T. G. FREELS², A. LILEY³, D. B. GABRIEL⁵, J. LEONIDES MARTINEZ⁶, Z. MIKKELSON⁴, N. W. SIMON³;

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Abstract: Risky decision-making is a hallmark of multiple psychological disorders, such as substance use disorder and schizophrenia. Disorders characterized by aberrant decision-making are often associated with abnormalities in cannabinoid signaling; however, the precise impact of manipulating cannabinoid receptors on punishment-driven risky decision-making remains poorly understood. Additionally, previous preclinical research has focused primarily on male subjects. To address this, we evaluated the acute systemic effects of multiple cannabinoid drugs on punishment based risky decision-making in male and female Long Evans rats. We measured risky behavior with the Risky Decision-Making Task (RDT), wherein rodents choose between a small safe reward (one food pellet) and a larger reward (three food pellets) with a risk of receiving a mild shock which increases across five blocks of trials (0%, 25%, 50%, 75%, 100%). After rats achieved stability in RDT, we assessed multiple doses of different cannabinoid drugs: THC, the primary psychoactive compound in cannabis plants which activates CB1 and CB2 receptors; ACEA, a selective CB1 receptor agonist; and Rimonabant, a selective CB1 receptor antagonist. THC exhibited sex-dependent effects on risky decision-making. Female rats were unaffected by THC, whereas males demonstrated inflexible decision-making, as evidenced by a reduced slope across multiple doses. Rimonabant did not produce significant effects on risky decision-making (RDT), omissions, locomotion, or latency to choose in male rats. However, at higher doses, female rats exhibited increased risk aversion as well as a significant increase in omissions compared to vehicle. Additionally, female rats took significantly longer to select risky but not safe choices with high dose Rimonabant. In contrast, ACEA did not influence risky decision-making in either sex, but did selectively reduce latency to select the safe (but not risky) option in females, indicating sex-specific effects on motivational or attentional processes independent of risk evaluation. Collectively, these data suggest that risky decision-making is susceptible to systemic activation of the cannabinoid system by THC. Furthermore, males are more vulnerable to the cognitive effects of CB receptor activation, whereas females are more vulnerable to the cognitive effects of CB1 blockade. Understanding how the cannabinoid system regulates risky decision-making is critical for developing treatments to improve decision-making in psychiatric disorders and address cannabinoid misuse.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR299.02/LL12

Topic: I.03. Decision Making

Title: Sex-dependent effects of psychotomimetic and antipsychotic drugs on risky decision-making

Authors: ***J. WHITEHEAD**¹, J. LEONIDES MARTINEZ³, K. HUNDLEY¹, N. PATTERSON¹, S. LOWE¹, Z. MIKKELSON¹, N. W. SIMON²;

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Abstract: Schizophrenia is a chronic and debilitating illness that produces widespread disruption of mental functioning, with higher-order cognition being particularly compromised. Decision-making, which requires integration of reward value and risk of negative consequences, is one of these deficits. However, it is difficult to understand the association between schizophrenia and risky decision-making in humans because most research participants with schizophrenia are concurrently taking antipsychotic medications. Here, we aimed to disentangle the contributions of a psychosis-like state and antipsychotic drug exposure on risky decision-making. Rats were trained in the Risky Decision-making Task, which presents a choice between small, safe rewards and large rewards accompanied by a probabilistic risk of mild footshock. To induce a psychosis-like state and examine potential decision-making effects of schizophrenia, we tested the acute effects of two mechanistically distinct psychotomimetic drugs. MK-801, an NMDA antagonist commonly used as a pharmacological model of schizophrenia, increased risk-taking behavior selectively in males. Conversely, a 5-HT_{2A} agonist that produces perceptual disturbances reminiscent of positive symptoms of schizophrenia decreased risk-taking behavior in males. We also assessed the effects of the atypical antipsychotic risperidone on risky decision-making. In female rats, a dose-dependent decrease in risk-taking occurred with all doses examined. In males, only high dose exposure decreased risk-taking. Finally, we are currently examining the effects of joint antipsychotic and psychotomimetic administration on risky choice. Preliminary data suggests that joint treatment evokes increased risk-aversion without impacting punishment sensitivity. Collectively, these data suggest that risky decision-making is impacted in sex-specific fashion by both psychotomimetic and antipsychotic drugs, which has important implications for managing aberrant decision-making and risk processing in schizophrenia.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

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Topic: I.03. Decision Making

Support: AMED JP21wm0425017, 21wm0425008 , JP23tm0524001
JSPS KAKENHI 23K27360, 23H02669, 24K18223, 24K02218

Title: Pramipexole hyperactivates the external globus pallidus and impairs decision-making in a mouse model of Parkinson's Disease

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Toyoake, Japan

Abstract: Dopamine replacement therapy with dopamine D2/D3 receptor agonists is a commonly employed pharmacological strategy for the treatment of Parkinson's disease (PD). The dopamine D3 receptor (D3R)-preferring agonist pramipexole (PPX) is particularly associated with impairments in decision-making, including pathological gambling in patients with PD. However, the neurobiological mechanisms underlying these adverse effects remain elusive. Here, in a mouse model of PD, we investigated the effects of PPX on decision-making. PD model mice were generated using a bilateral injection of the toxin 6-hydroxydopamine into the dorsolateral striatum. Decision-making performance was assessed using a touchscreen-based Iowa Gambling Task (IGT), which evaluates choices between options offering varying levels of risk and reward, closely mirroring probabilistic decision-making in humans. Subsequent treatment with PPX increased disadvantageous choices characterized by a high-risk/high-reward outcomes in the IGT. This effect was blocked by treatment with the selective D3R antagonist PG-01037, implicating D3R-mediated signaling in the pathogenesis of impaired decision-making. In model mice treated with PPX, the number of c-Fos-positive cells was increased in the external globus pallidus (GPe), indicating dysregulation of the indirect pathway in the corticothalamic-basal ganglia circuitry. In accordance, chemogenetic inhibition of GPe neurons using a DREADD-based approach successfully restored normal c-Fos activation and rescued PPX-induced disadvantageous choices. These findings demonstrate that the hyperactivation of GPe neurons in the indirect pathway impairs decision-making in PD model mice. The results provide a candidate mechanism and therapeutic target for pathological gambling observed during D2/D3 receptor pharmacotherapy in PD patients.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

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Program #/Poster #: PSTR299.04/LL14

Topic: I.03. Decision Making

Support: CIHR Project Grant #190154

Title: Brain networks underlying approach-avoidance conflict processing in rodents and humans

Authors: N. R. STEWART, S. VALENZANO, J. ZHANG, Z. WU, S. CHU, R. ITO, *A. C. LEE;

Univ. of Toronto, Toronto, ON, Canada

Abstract: A number of cortical and subcortical structures, including the medial prefrontal cortex, nucleus accumbens, striatum, lateral septum, and hippocampus, have been suggested to subserve the resolution of approach-avoidance conflict (AAC), a scenario that arises when a stimulus is simultaneously associated with reward and punishment. Relatively little is known, however, about how these regions interact in the context of brain-wide functional networks during AAC decision making. To investigate this, the current study applied graph theory to rodent c-Fos and human functional magnetic resonance imaging (fMRI) data acquired in association with high AAC processing involving learned incentive values. Rats (n = 39) were first trained to associate individual cues with sucrose or shock. Following successful conditioning, they were exposed to a combination of these cues (i.e., high AAC), and c-Fos was then quantified in a number of a priori regions of interest (ROIs) implicated in value-based decision making, learning, and motivational behaviour. Neurologically healthy human participants (n = 39) completed a rule-based AAC task during fMRI, in which they approached or avoided pairs of indoor/outdoor scene images according to a reward/punishment rule associated with monetary gain or the consumption of an unpleasant food mixture (e.g., indoor = positive; outdoor = negative). Percent signal change during high AAC trials (i.e., a positive scene paired with a negative scene) was extracted in ROIs homologous to those chosen in rodents. Separate interregional correlation matrices were created for the rat c-Fos and human fMRI data and then submitted to network analyses. These revealed distinct communities of regions in rats and humans, with shared characteristics across species. For example, hippocampal subregions and the insula were identified as hub-like regions and network edges were largely consistent with known anatomical connectivity, including between the hippocampus, amygdala, medial prefrontal cortex, striatum, and lateral septum. Our study sheds novel insight into the brain networks underlying AAC processing and highlights the benefits (as well as challenges) of a cross-species methodological approach.

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Poster

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Program #/Poster #: PSTR299.05/LL15

Topic: I.03. Decision Making

Title: Brain-wide neural dynamics for linking value to action during economic decisions

Authors: *C. BAO, J. LI, O. M. GAULD, C. A. DUAN, J. C. ERLICH;
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Abstract: Economic decisions often arise in diverse and dynamic contexts, requiring the brain to flexibly map value evaluation and economic preference to distinct actions. This flexibility relies on coordinated interactions among neural circuits involved in sensory processing, valuation, decision-making, and motor control. To understand these mechanisms, it is crucial to determine where in the brain option values are computed and compared, how abstract economic choices are transformed into spatial motor plans, and how these computations are distributed across cortical and subcortical networks. However, disentangling these deliberation processes remains a significant challenge, as value representations are often temporarily intertwined with signals related to action preparation and execution.

To address these questions, we developed a novel head-fixed behavioral task in mice, where animals chose between a cued lottery and a small stable surebet. The lottery expected value changed from trial to trial and was indicated by an auditory cue. The task temporally dissociates abstract economic choice from motor planning using two consecutive phases: an option evaluation phase where only the lottery value but not the lottery location is cued, followed by a motor planning phase in which the lottery's spatial location is revealed. Using acute Neuropixels recordings, we simultaneously recorded neural activity across multiple brain regions connected with the anterior lateral motor cortex (ALM), a key area for memory-guided licking. After value cue onset, cue specific responses emerged quickly across several areas (agranular insular cortex, ALM, thalamus, dorsal striatum, hippocampus, orbitofrontal cortex (OFC), and superior colliculus (SC)). These responses were stable throughout the cue presentation period and remained robust in ALM and OFC even beyond cue offset. In many of these areas, including frontal and thalamic regions, these signals evolved into abstract economic choice signals, differentiating between lottery versus surebet choices. Despite the widespread distribution of non-spatial economic choice signals, only the ALM, OFC, and thalamus maintained these signals into the subsequent epoch when value-guided decision information needs to be integrated with location information to plan a spatial choice. Robust motor planning and execution signals were also localized to the ALM, OFC, and thalamus. Together, these findings highlight the ALM, thalamus, and OFC as key hubs that bridge value-based deliberation and motor execution during economic decision-making under risk.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

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Topic: I.03. Decision Making

Support: KAKENHI(JP20H03357)
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KAKENHI(JP22H05162)
JST(JPMJMS2294)

Title: Behavioral confidence reporting task for head-fixed rats

Authors: *S. SUGIMOTO, Y. TANAKA;
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Abstract: Confidence is a component of metacognition, and understanding how neural mechanisms support its behavioral expression in animals may enhance our knowledge of cognitive processes. Because animals cannot report their confidence verbally, previous studies have developed behavioral paradigms that indirectly infer confidence from observable behaviors. One example of such a task combines sensory discrimination with temporal wagering. In that task, freely moving rats report their confidence by how long they spend waiting at a reward port after making a choice (“postdecision wagering task” in Lak et al., 2014, *Neuron*; “behavioral confidence reporting task” in Masset and Ott et al., 2020, *Cell*). Building on these studies, we developed a confidence- reporting task for head-fixed rats using visual stimuli. In this task, the rats moved a cursor on a screen by pressing buttons on the left and right sides of their forepaws. Pressing the left and right buttons moved the cursor to the left and right, respectively. Each trial began with a cursor centered on the screen, followed by the appearance of a Gabor patch with varying contrasts on either the left or right side of the screen. Upon a correct cursor placement, a water reward was delivered after a random delay. After making a choice, the rat needed to return the cursor to the center to initiate the next trial. The duration from the choice to the cursor return was recorded as “time investment,” serving as a behavioral proxy for confidence. To measure time investment without the confounding effect of reward consumption, we included probe trials in which the reward was omitted from a subset of correct trials. After pretraining sessions (21.0 ± 7.9 sessions; mean $\pm$ SD), twelve rats were trained on this task and showed robust confidence indications after several additional sessions (10.3 ± 4.9 sessions; mean $\pm$ SD), as evidenced by significant differences in time investment between probe and error trials. The head-fixed task offers two major methodological advantages for investigating the neural basis of confidence. First, by minimizing movement-related noise, it is particularly well-suited for imaging experiments, enabling high-resolution behavioral measurements, such as pupillometry. Second, compared with freely moving conditions, it facilitates the integration of multiple recording modalities and neural manipulations. Thus, this task provides a promising platform to elucidate the neural dynamics underlying decision confidence.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

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Program #/Poster #: PSTR299.07/LL17

Topic: I.03. Decision Making

Title: Replicable modeling of opioid withdrawal-induced cognitive deficits in rats using cross-species translatable tools

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Abstract: Cognitive deficits are observed in persons who formerly used opioids chronically. These cognitive opioid withdrawal (OW)-induced deficits are untreated, despite being observed in late (protracted) withdrawal. Of concern, OW induces risky decision-making which correlates with relapse and poor clinical outcomes. Effective preclinical modeling of OW-induced cognitive deficits is lacking but required for delineating neural mechanisms for the development of targeted therapeutics. Here attempt to model OW-induce cognitive deficits in rats using cross-species translatable assays. For initial piloting a group (n=20; 50% female) of non-experimentally naïve Fischer F344 rats were utilized. A second cohort of experimentally naïve rats were subsequently utilized (n=14; female). Rats were trained to respond in operant chambers for milkshake reward, then exposed to an 11-day intermittent fentanyl protocol previously shown to produce dependence (0.1 mg/kg i.p. injection 2x/day) or saline (control). Rats were tested in the Iowa Gambling Task (IGT) to measure risk-based decision-making and the Progressive Ratio Breakpoint Task (PRBT) to measure effortful motivation. Tests occurred at baseline (Cohort 2 only), during fentanyl exposure (4 hr following injection), during acute OW (16 hr post abstinence), then every 3 - 5 days thereafter to assess how these functions change as a result of opioid exposure, and withdrawal therefrom. Across cohorts, there were no differences between treatment groups at baseline (Cohort 2 only) or during fentanyl exposure (ts<1.2, ns). Instead, acute OW produced robust IGT deficits compared to treatment (Fs>1.8, p<0.05), with no additional impact of sex (determined by Cohort 1). IGT deficits were also observed during protracted withdrawal, across both cohorts, indicating long-lasting impairments. Instead, there were no significant drug-exposure group differences in PRBT performance at baseline (Cohort 2 only), shortly following fentanyl exposure, or during the OW testing period. Our findings indicate that withdrawal from, but not exposure to, chronic intermittent fentanyl produces reproducible risk-based decision-making deficits in rats performing the IGT, consistent with OW-induced IGT deficits observed in the clinic. Neither fentanyl exposure, nor withdrawal therefrom, resulted in PRBT deficits, suggesting OW-induced IGT deficits were not a result of changes to effortful motivation to achieve reward. Hence, this model can be used to explore the impact of OW on other translatable cognitive tasks and leveraged to explore neural mechanisms contributing to withdrawal-induced IGT deficits.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

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Title: Neural oscillatory signals representing uncertainty in decision-making in the primate anterior insular cortex

Authors: *R. ITO^{1,2}, R. KATO², S. ISHII¹, K.-I. AMEMORI²;

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Abstract: Uncertainty in predictions often leads to unstable expectations and significantly influences decision-making. Several studies have suggested that neural activity in the anterior insular cortex (AIC) correlates with decision variables reflecting the degree of uncertainty. However, the mechanisms by which the AIC contributes to decision-making under uncertainty remain largely unexplored. To investigate this issue, we trained two macaque monkeys to perform the Balloon Analogue Risk Task (BART), a decision-making task involving reward uncertainty. In BART, the monkey inflated a virtual balloon by fixating on a central target (“pump up”), thereby increasing the amount of stored reward. When the monkey broke fixation and made a saccade to the peripheral target, the accumulated reward was delivered (“cash out”). Continued pumping carried the risk of a random balloon burst, which resulted in the loss of the stored reward. The timing of the burst was not informed to the monkeys. As a control, we randomly interleaved trials of Cued BART, in which the burst timing was visually signaled. First, we recorded local field potentials from the AIC during the decision period and quantified beta-band power (13 to 30 Hz). Mean beta power preceding the saccade was significantly more suppressed in Cued BART than in BART at many recording sites (36 of 61; two-sample t test, $P < 0.05$), indicating an uncertainty-dependent modulation of beta activity. In BART, beta power during the decision period correlated with cash-out timing at 17 of 50 recording sites (Pearson correlation, $P < 0.05$), whereas such correlation was observed at only 5 of 49 sites in Cued BART. The proportion of correlated sites differed significantly between the two tasks (chi-square test, $P < 0.01$), suggesting that AIC beta suppression primarily reflects the timing of decision-making under uncertainty. Second, we applied microstimulation to the AIC (200 Hz, 150-200 μ A, 1-2 s duration). We calculated cash-out times and derived deviations from the non-stimulated baseline. In the effective sessions (10/28 sites), microstimulation significantly altered the deviation of cash-out timing in BART relative to the preceding non-stimulated sessions (two-sample t test, $P < 0.05$). In contrast, stimulation had a significantly smaller effect in Cued BART (1/28 sites) (chi-square test, $P < 0.01$). In summary, beta suppression in the AIC predicted cash-out timing, and microstimulation influenced decisions only when the timing was not explicitly cued. These findings suggest that the AIC plays a causal role in decision-making under uncertainty, with beta oscillatory activity supporting the prediction of upcoming cash-out timing.

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Poster

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Topic: I.03. Decision Making

Support: NIH/NIMH R01MH128669

Title: Uncertainty coding in the dorsolateral prefrontal cortex

Authors: ***W. G. KERKHOFF**¹, W. R. STAUFFER²;
²Neurobio., ¹Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Real-world decision making almost always involves some form of uncertainty. Decision theory distinguishes between two types: **risk**, where the possible outcomes and their probabilities are known, and **ambiguity**, where the probabilities are unknown. We and others have described the behavioral and neural computations underlying individual risk attitudes. However, the mechanisms driving ambiguity attitudes remain poorly understood, in part because they require a fundamentally different cognitive process compared to risk. While risky uncertainty can be directly quantified based on experience or instruction, ambiguous uncertainty cannot. Consequently, decisions under ambiguity depend on subjective probability estimates. As a result, the mechanisms that drive attitudes toward ambiguity remain poorly understood, despite their importance for understanding real-world behavior. To investigate the cognitive basis of ambiguity attitudes, we developed a novel behavioral task—the method of distributional equivalents—for use in non-human primates (NHPs). This task dissociates reward magnitude and probability using a two-dimensional visual stimulus, with ambiguity induced by masking probability information. By comparing ambiguous choices with risky choices across low, medium, and high reward magnitude ranges, we identified the *distributional equivalent*—the risky option that choices reveal to be subjectively equivalent to an ambiguous prospect. The distributional equivalents directly revealed subjective probability estimates. To investigate the neural basis of those estimates, we performed single-unit electrophysiological recordings in the dorsolateral prefrontal cortex (dlPFC). Single units in the dlPFC demonstrate responses to risky and ambiguous stimuli, suggesting a role for the dlPFC in representing different types of uncertainty. This work will provide insights into the biological foundations of beliefs and decision-making, offering new perspectives on the mechanisms for ‘irrational’ choices.

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Poster

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RO1 EY017699
R01MH137624

Title: Hierarchical Decision-Making in Rhesus Monkeys and Humans: Strategy Profiling and Inter-Species Comparison

Authors: *J. HUANG, L. YING, S. KASTNER;
Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ

Abstract: Efficient decision-making in natural environments requires to simultaneously select among perceptual alternatives and update task rules. To investigate how primates perform such hierarchical choices, we designed a two-tier random-dot paradigm: (i) participants judge a the dominant color of a dot cloud consisting of red or blue dots presented under variable sensory noise conditions (i.e. mediated by color coherence), and (ii) mapping of the dominant dot color to images of one of three object categories (e.g., red→dog , blue→tree). The mapping rules changed unpredictably and covertly. We collected data from ~ 70 human subjects and, in parallel, trained five rhesus monkeys (4 females) using an automated in-cage touchscreen system that exactly mirrored the human task. Particularly the female monkeys demonstrated rapid improvement through mutual observational learning and spontaneously took turns using the touchscreen according to social rank, without conflicts. Both humans and monkeys exhibited robust post-error slowing: subjects showed increased reaction times in the subsequent trial, indicating rapid detection of errors and behavioral adjustment. By constructing trial-level behavioral models, we dissociated perceptual errors (caused by ambiguous color coherence) from rule-based errors (caused by failure to update mappings). Both species dynamically adjusted their confidence in whether the mapping had changed according to the sensory uncertainty of the dot cloud, reflecting hierarchical reasoning across perceptual and rule levels. Quantitatively, humans typically achieved asymptotic accuracy on the new mapping within approximately 10 trials after a switch, whereas monkeys required more than 30 trials to reach a comparable level of correct performance. Monkeys also exhibited significant perseveration on the previous mapping and confusion among valid options, suggesting more limited working-memory resources and processing capacity compared to humans. These findings imply that the core neural mechanisms supporting uncertainty-based hierarchical decision-making are evolutionarily conserved across primates. Nonetheless, species-specific differences in memory retention and processing speed reflect monkeys' reliance on a more basic decision-making framework, whereas humans may benefit from enhanced working memory and language.

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR299.11/LL21

Topic: I.03. Decision Making

Title: Internal Mechanisms of Loss Aversion Decision Making

Authors: *L. HIRT¹, P. SOKOL-HESSNER², G. FELSEN³, D. KRAMER⁴, J. A. THOMPSON⁵;

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Abstract: Loss aversion is the tendency to internally emphasize potential losses over gains and impacts everyday decision-making; however, the neural circuitry involved in this phenomenon is not well understood. Loss aversion has been implicated in poor long-term decision making and may contribute to disorders associated with impaired decision-making such as anxiety and depression. Previous fMRI results suggest an increase in amygdala activity when participants experience a monetary loss when engaged in a gambling paradigm. As an important node in affective processing, the amygdala is hypothesized to drive loss aversion behavior. One putative circuit that could support long term network modifications related to the status of wins and losses would be connections between the amygdala and hippocampus, implicated in forming associations with negative stimuli. The interaction between the amygdala and hippocampus warrants further exploration, as it may contribute to the maintenance of loss aversion by reinforcing negative and positive outcome associations. To expand our understanding of the internal mechanisms of loss aversion, the current study utilized internal electrophysiology (iEEG) in humans. We examined amygdala and hippocampus activity while the participants completed a validated gambling task that assessed loss aversion. We sought to explore the oscillatory activity during the outcome epoch, when participants who gambled learned whether they won or lost. As a first step toward examining amygdala-hippocampus interactions, we quantified activity in each region during the decision epoch. Comparison of FOOOF oscillatory activity between the targeted brain areas demonstrated that both power (PW) and peak frequency (CF) were significantly different ($p = 0.01$ and $p = 0.01$). Post hoc results demonstrated a significant difference in CF and PW for amygdala and anterior hippocampus ($p = 0.01$) and amygdala and posterior hippocampus ($p = 0.01$). Additionally, an ANOVA between trial type (if the participant gambled or not) by CF or PW had significant results ($p = 0.01$). Post hoc analysis revealed a significant difference in CF and PW when someone gambled versus when they did not ($p = 0.01$). These preliminary results highlight the importance of understanding the neural mechanism of loss aversion.

Disclosures: L. Hirt: None. P. Sokol-Hessner: None. G. Felsen: None. D. Kramer: None. J.A. Thompson: None.

Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR299.12/LL22

Topic: I.03. Decision Making

Title: Subjective behavioral thermoregulation: how individualized thermal comfort affects human decision making: preliminary behavioral and neuroimaging evidence

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Abstract: Environmental neuroscience explores how external physical conditions, such as temperatures, shape brain and behavior. Little is known about the effect of moderate temperature deviations (“operational temperature”, ~20°C to ~30°C) on decision-making and its underlying neural mechanisms. We present preliminary findings from a large-scale, multimodal study examining how individual optimal thermal comfort affects decision behavior and brain function. We conducted a preliminary pilot (N = 30, mean age = 28.1 [± 4.9], 9/21 Male/Female), including lab and MRI visits. In each session, participants underwent a rest phase to assess their baseline comfort temperatures, while watching a nature-scene video, followed by decision-making tasks. Both phases were repeated multiple times under varying temperatures. Physiological data, including heart rate and skin temperatures, were continuously recorded. These measures, combined with participants’ self-reported comfort rankings, were used to estimate individual optimal temperatures (IOT). Decision making tasks included (1) Mixed Gambles Task to measure loss aversion, (2) Delay Discounting Task for impulsivity, (3) Probability Learning Task to measure sensitivity to reward, and (4) Balloon Analogue Risk Task for risk-taking. Behavioral performance showed consistent marginal effects: deviations from IOT were linked to reduced loss aversion ($\beta = -.01$; $p = .065$), steeper discounting ($\beta = -.01$; $p = .073$), and increased risk-taking ($\beta = .16$; $p = .064$). These effects suggest thermal discomfort may compromise executive function, biasing behavior toward immediate, riskier choices. We applied drift-diffusion modeling (DDM) to examine underlying behavioral mechanisms in decision-making processes. The computational modeling of these tasks revealed an increased in threshold for evidence accumulation (non-decision time τ : [$\beta = -.01$; $p = .052$] ; decision boundary α : [$\beta = .01$; $p = .032$]), suggesting increased uncertainty in choices for sub-optimal temperatures. Structural MRI using T1-weighted scans, in a subset sample (N=20, mean age = 27.3 [± 4.7], 6/14 Male/Female), revealed associations between IOT and grey matter volume in the hypothalamus, and axonal density in cortico-spinal tracts, both regions related to thermoregulation. These results demonstrate the feasibility of our multimodal approach, suggesting that deviations in moderate temperature can alter decision-making, with a potential underlying neural mechanism. This work also sheds light on cognitive adaptability in the face of climate change and provides mechanistic insights into how environmental stressors shape human behavior.

Disclosures: A. Itzkovitch: None. T. Schonberg: None.

Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR299.13/MM1

Topic: I.03. Decision Making

Title: Aspects of uncertainty cognition in AGI/ASI capabilities and their Implications for AI regulation

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Abstract: Human perception of risk and benefit, central to behavioral economics and prospect theory (PT) (Kahneman & Tversky, 1979), reveals systematic biases such as loss aversion and context-dependent risk preferences. As Artificial General Intelligence (AGI) and Superintelligence (ASI) (e.g., Bostrom, 2014) approach potential realization, societal anticipation is mixed, reflecting diverse cognitive evaluations of their profound opportunities and risks (e.g., Zhang & Dafoe, 2020). A critical unexplored question is how established cognitive processes for assessing risks and benefits transfer to these novel, high-impact technologies, and how such transferred perceptions subsequently shape public demand for their regulation. Understanding this transfer is crucial, as it directly impacts how individuals and societies will likely approach the regulation and governance of these transformative technologies, offering vital insights for policymakers aiming to develop AI regulations that align with public cognitive frameworks, as exemplified by comprehensive initiatives like the EU AI Act (Regulation (EU) 2024/1689). Here we investigate how individual decision-making biases, typically observed in economic contexts and described by PT, transfer to the perception of AGI and ASI. We further examine how these transferred cognitive evaluations of risks and benefits correlate with attitudes towards the governance and regulation of AGI and ASI. An online survey was administered among Japanese-speaking participants. The questionnaire covered issues such as choice-based tasks derived from PT to assess individual biases in risk/benefit perception (e.g., Tversky & Kahneman, 1992; Toyoda et al., 2007), items measuring attitudes towards AGI and ASI, including perceived benefits, risks, timelines, acceptance, and views on AI consciousness/rights, support for development, and specific preferences regarding their regulation and governance, together with demographic data and AI familiarity. Analyses focused on how PT-derived metrics correlated with the perceived risk/benefit balance of AGI/ASI and subsequent support for different regulatory approaches. We discuss implications for neuroeconomic models of how humans adapt existing decision-heuristics to unprecedented technological advancements (Glimcher & Rustichini, 2004)

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Poster

PSTR299: Neural Mechanisms: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR299.14/MM2

Topic: I.03. Decision Making

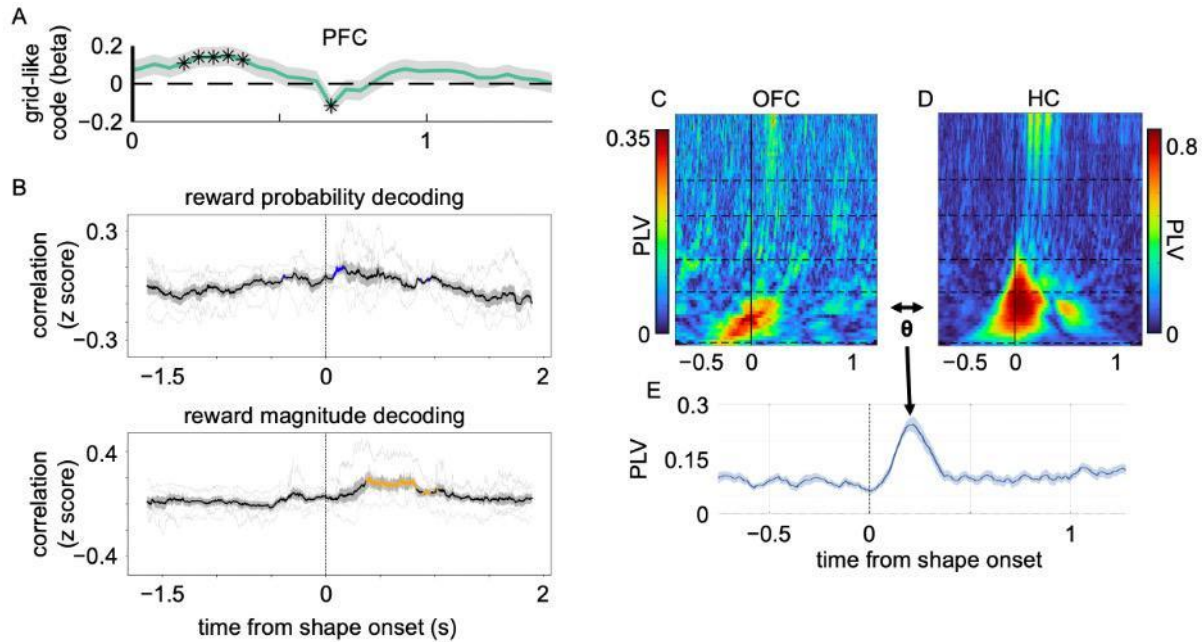
Support: 1R01MH123713-01

Title: Cognitive maps and theta oscillations in medial temporal lobe and prefrontal cortex during risky decision-making

Authors: *M. A. ORLOFF¹, S. A. PARK³, C. VERSTRAETE⁴, P. DOMENECH⁵, E. D. BOORMAN²;

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Abstract: Neuroeconomic studies propose that decisions utilize a common neural currency, where rewarding things such as juice, money, and social stimuli are encoded using a common neural value code in the brain. Within frontal cortex, such value coding has been identified in dorsal anterior cingulate cortex, orbitofrontal cortex (OFC), and ventromedial prefrontal cortex (vmPFC), among other regions. Our recent work shows that in addition to this brain valuation system, a cognitive mapping system emerges in a risky decision-making task in medial temporal lobe (MTL) and vmPFC and medial OFC (mOFC). Given the role of MTL in cognitive mapping and the PFC in both cognitive mapping and value-based decision making, we examine computations and communication between MTL and PFC in humans undergoing presurgical iEEG monitoring for treatment-refractory epilepsy. In this task, patients made binary choices between shapes, which varied along two continuous dimensions corresponding to reward magnitude and probability. We identify that a grid-like representation (a neural signature of cognitive maps) of decision vectors between options emerges in PFC during risky decision-making (A). Additionally, value information can be decoded (across all contacts including those in MTL and PFC) across subjects (N = 6) after viewing choice options (B). To examine hypothesized information transfer between MTL and PFC, we computed intertrial phase coherence (phase locking value, PLV) time-locked to the revelation of the second shape. We observed strong inter-trial coherence in both MTL and PFC—more specifically HC and OFC—in the theta band immediately following the revelation of the second shape (C and D). To test how information is transferred between these regions, we measured phase coherence *between* these regions in theta band. As hypothesized, and consistent with other studies, we observed a high PLV between HC and OFC after presentation of the second shape (E). Taken together, this suggests that theta coupling between HC and OFC supports information transfer for value-based choices.



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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.01/MM3

Topic: I.05. Working Memory

Title: Explainable Working Memory Performance Through the Lens of Machine Learning

Authors: Z. CHEN¹, P. JIANG², D. LUO¹, V. CHAUDHARY³, J. GERSTENBERGER⁴, S. LI^{3,4}, N. XIE¹, S. XU³, *S. PU⁴;

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Abstract: Working memory (WM) is central to cognitive flexibility and is closely associated with sustained activity in the prefrontal cortex (PFC). However, the computational mechanisms supporting this activity—and the minimal requirements for decoding task-relevant information—remain unclear. In this study, we present a comprehensive evaluation of minimalist machine learning (ML) models for decoding spatial WM signals from spiking activity recorded during oculomotor delayed response (ODR) tasks in rhesus monkeys. Using data from both the PFC and posterior parietal cortex (PPC) in juvenile and adult monkeys, we compared traditional

classifiers (such as KNN, logistic regression, SVM) to neural networks. When optimized, linear models performed similarly well as more complex networks in decoding cue location, particularly in well-trained animals. Remarkably, small, highly sparse subsets of neurons were sufficient to reach behavioral-level decoding accuracy, supporting the notion of sparse yet redundant population coding. Temporal analysis of model parameters revealed distinct encoding dynamics across task epochs. Training significantly improved the decodability of WM content, especially in the PFC, indicating experience-dependent plasticity in coding efficiency. Finally, we explored the alignment between the low-dimensional structure of our optimized ML models and the latent representations generated by shallow neural networks performing analogous memory tasks. This multi-method comparison uncovered partial geometric congruence, suggesting shared computational motifs between biological and artificial systems. Collectively, our findings offer a framework for decoding and interpreting WM representations using minimal computational tools. The results shed light on the distinct contributions of different brain areas to memory maintenance and contribute to our understanding of the efficiency and scalability of cortical coding in both biological and artificial intelligence.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.02/MM4

Topic: I.05. Working Memory

Support: MOE-T2EP30121-0010
MOE2017-T3-1-002

Title: No Evidence of Activity-Silent Working Memory Periods in Prefrontal Cortex

Authors: *C. LIBEDINSKY;
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Abstract: The lateral prefrontal cortex encodes working memory and motor preparation information; however, the underlying neural mechanisms are still debated. Neural network models relying on persistent neural activity have been challenged by the observation of periods of absent activity and information during memory maintenance, implying the existence of activity-silent mechanisms. Two key pieces of evidence support the idea of activity-silent mechanisms of memory maintenance. First, there is more informative spiking during gamma bursts, implying that activity-silent mechanisms may be necessary to maintain memory information between them. For this to be true, activity between gamma bursts should be uninformative. Second, the decoding of memory information in individual trials can drop to chance levels, implying that activity-silent mechanisms may be necessary to maintain

information during these periods. For this to be true, activity during these chance decoding periods should be absent, since the activity of a few selective neurons is sufficient to maintain memories in an attractor state. To test these hypotheses we recorded neural activity in male macaque (*Macaca fascicularis*) prefrontal cortex during a delayed-saccade task. We found that memory information is present in the spiking activity between gamma bursts, contradicting the gamma-burst model. Additionally, we did not find periods of silence of selective neurons longer than 150 ms (the temporal smoothing of NMDA receptors). In summary, our study challenges the activity-silent hypothesis of working memory maintenance.

Disclosures: C. Libedinsky: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: I.05. Working Memory

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The JPB Foundation
The Picower Institute for Learning and Memory
Research Program funded by the SeoulTech (Seoul National University of Science and Technology)
The Ministry of Trade, Industry, & Energy (MOTIE) of Korea (RS-2025-04812973)

Title: Working memory readout varies with prefrontal theta rhythms

Authors: *H.-B. HAN^{1,2}, S. L. BRINCAT¹, T. BUSCHMAN³, E. K. MILLER¹;

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Abstract: Increasing evidence suggests that attention varies rhythmically, phase-locked to ongoing cortical oscillations. Here, we report that the phase of theta oscillations (3-6 Hz) in the frontal eye field (FEF) is associated with temporal and spatial variation of the read-out of information from working memory (WM). Non-human primates were briefly shown a sample array of colored squares. A short time later, they viewed a test array and were rewarded for identifying which square changed color (the target). Better performance (accuracy and reaction time) varied systematically with the phase of local field potential (LFP) theta in FEF at the time of test array onset. Neural information analysis revealed a test array presented at the behaviorally optimal phase of theta led to readout of more sample array information. The optimal theta phase was earlier when the target was in the upper visual field than in the lower. This is consistent with theta “scanning” across the FEF, and thus visual space, from top to bottom. Consistent with this

prediction, we found that theta exhibited properties of a traveling wave across the FEF, with theta exhibiting a phase gradient of $\sim 0.5^\circ/\text{mm}$ across cortical space. This FEF theta was coupled, on opposing phases, to both cortical excitability (spiking) and beta (12-20 Hz) power. These results would be explained by a theta wave traveling across FEF, modulating the readout of information from WM.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

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Program #/Poster #: PSTR300.04/MM6

Topic: I.05. Working Memory

Support: NINDS R01NS113073
NIH R01 EY026924
NIH EY014800
Research to Prevent Blindness

Title: Optogenetic manipulation of visual inputs to FEF during visual working memory reveals dynamic gating of communication efficacy

Authors: *P. COMEAUX¹, L. NURMINEN², F. FEDERER¹, K. CLARK¹, A. ANGELUCCI¹, B. NOUDOOST¹;

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Abstract: Prefrontal control of sensory signals has been shown to be associated with a coherent oscillation in local field potentials (LFPs) between the two areas. For example, our group has shown that Beta band coherence increases between the Frontal Eye Field (FEF) and V4 during the maintenance of working memory (WM). This increased coherence is associated with an increase in efficacy of spike propagation from V4 to the FEF. These results implicate coherent oscillations as a potential means to alter the efficacy of signals transferred between cortical areas. We hypothesize that oscillation phase and coherence gate inputs from V4 to the FEF to increase the efficacy of communication during WM task completion, so to preserve LFP for analysis, we use optogenetic stimulation to induce activity from V4 projections in the FEF. Anterograde vectors containing channelrhodopsin-2 were injected into V4, allowing for the activation of V4 projections in the FEF when photostimulating the FEF. Photostimulation and simultaneous recording of spiking and LFP activity in FEF was performed using an opto-electrode. Using photostimulation in an awake, behaving rhesus macaque monkey performing a memory-guided saccade (MGS) task, we quantified the impact of V4 input to the FEF as a function of task epoch, memory condition, or neural state at the time of stimulation. FEF and V4 were first mapped to find sites with overlapping receptive fields (RFs) for photostimulation and recording. Spiking and LFP activity were simultaneously recorded from both areas. To understand how stimulation

of V4 input to FEF inputs impacts WM behavior we employed a task in which the animal needs to maintain a target in its WM, in the presence of a distractor. Photostimulation occurring during distractor presentation led to a greater probability of selecting the distractor as the WM target when the distractor was presented in the overlapping FEF and V4 RFs. This indicates that the strength of V4 to FEF inputs can gate the ability for sensory stimuli to enter WM. Optogenetic stimulation occurred during different epochs of the MGS task to compare the effect of varying cognitive states on the dynamic gating between V4 and the FEF. We have found pre-stimulation patterns of oscillatory activity in the FEF which predict a larger increase in LFP power in response to the stimulation of V4 projections during the maintenance of visual WM matching the RF location, indicative of dynamic gating of the FEF's response to visual input based on FEF LFP oscillations and the content of WM.

Disclosures: P. Comeaux: None. L. Nurminen: None. F. Federer: None. K. Clark: None. A. Angelucci: None. B. Noudoost: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

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Program #/Poster #: PSTR300.05/MM7

Topic: I.05. Working Memory

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NSF NRT Award #2152260 to WH
NSF NRT Award #2152260 to OS
Brain and Behavior Research Foundation to XC
NIH Grant EY014924 to XC

Title: Temporal Dynamics and Functional Specialization of Frontal Oculomotor Neurons

Authors: B. N. SEVAK¹, W. HU⁴, O. SOYUHOS², X. CHEN³;

¹Biomed. Engin., ³Dept. of Neurobiology, Physiology, and Behavior, ²Univ. of California, Davis, Davis, CA; ⁴UC Davis Ctr. for Neurosci., Davis, CA

Abstract: Oculomotor neurons in the frontal cortex have traditionally been categorized into functional subtypes such as - visual (sensory-driven responses), motor (eye movement-related responses), and memory-delay neurons (integrating sensory and motor information). Recent evidence indicates distinct roles for these neuron classes in cognitive control processes. Memory-delay neurons, in particular, not only maintain information during working memory but also have monosynaptic projections to the visual cortex, which may serve a crucial role in modulating visual cortical activity during attentional control.

This study examines the temporal dynamics of these neuronal subtypes, their responses to subthreshold microstimulation, and the subsequent effects on behavior. High-density neurophysiological recordings were performed in the frontal eye field (FEF) and supplementary

eye field (SEF) of a non-human primate engaged in a memory-guided saccade task. Subthreshold microstimulation was delivered during the memory epoch to assess its impact on neuronal activity.

Preliminary findings indicate that memory-delay neurons possess significantly longer intrinsic temporal scales compared to visual and motor neurons and exhibit sustained, increased firing following subthreshold microstimulation. These results underscore the specialized temporal and functional properties of memory-delay neurons and highlight their critical role in cognitive control mechanisms, with implications for understanding how targeted neuronal modulation influences behavior.

Disclosures: B.N. Sevak: None. W. Hu: None. O. Soyuhos: None. X. Chen: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

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Program #/Poster #: PSTR300.06/MM8

Topic: I.05. Working Memory

Support: NIH Grant 5R01NS119519-03

Title: Mechanisms in macaque prefrontal cortex for predicting the motion of occluded objects

Authors: *N. WATTERS¹, J. GABEL², M. JAZAYERI³;

²McGovern Inst. for Brain Res., ¹MIT, Cambridge, MA; ³Brain and Cognitive Sci., Massachusetts Inst. of Technol. Dept. of Brain and Cognitive Sci., Cambridge, MA

Abstract: The primate mind excels at making predictions about physical scenes with moving objects. From crossing a busy street to playing a sport, this capacity is on display at every turn. Cognitive theories have hypothesized that this “intuitive physics” underlies more abstract reasoning such as planning and imagination. However, the neural mechanisms by which the brain predicts the kinematics of occluded moving objects remain largely a mystery. In particular, three competing hypotheses remain unresolved. One hypothesis is simulation, under which the brain updates the evolving states of objects in real-time. A second hypothesis is delayed extrapolation, under which the brain instead tracks elapsed time that can be subsequently combined with a memory of previous states of moving objects to infer their current position. A third hypothesis is heuristics-based anticipation, under which the brain uses priors to precompute the expected state of objects when they will next be visible.

To resolve these competing hypotheses, we developed a task for non-human primates (NHPs) that requires predicting kinematics of multiple occluded objects as they move in a visual display. We trained two animals on this task and verified that they were able to remember and predict the positions and identities of multiple independently moving occluded objects. Using acute Neuropixels recordings, we measured large-scale activity of single neurons simultaneously in dorsomedial frontal cortex (DMFC) and frontal eye fields (FEF), areas that prior studies have

implicated in making predictions about dynamic scenes. We then formulated our hypotheses as computational constraints on neural activity and measured the degree to which each hypothesis was consistent with our data using encoding models. We found that neural activity in both FEF and DMFC shows an initial heuristics-based anticipation component, followed by subsequent simulation, and rejects our delayed extrapolation hypothesis. We used a suite of synthetic datasets with known ground truth to validate our methodology and reveal the limitations of alternative approaches such as decoding models. Lastly, our encoding models allowed us to infer the monkey's mental object tracking state on single trials from neural activity alone, and we found that this predicted the variability of subsequent responses, demonstrating that the latent variables inferred by our models are behaviorally meaningful.

Disclosures: **N. Watters:** A. Employment/Salary (full or part-time): Massachusetts Institute of Technology. **J. Gabel:** A. Employment/Salary (full or part-time): Massachusetts Institute of Technology. **M. Jazayeri:** A. Employment/Salary (full or part-time): Massachusetts Institute of Technology.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.07/MM9

Topic: I.05. Working Memory

Support: NIH Grant EY029438
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Title: Visuospatial working memory depends on the phase-locking of beta oscillations in macaque parietal cortex

Authors: ***M. ABDALAZIZ**^{1,2}, R. DOUDLAH², L. KRESSER², Z. ZHU², T.-Y. CHANG³, B. KIM², A. ROSENBERG²;

¹Univ. of Wisconsin - Madison Neurosci. Training Program, Madison, WI; ²Neurosci., Univ. of Wisconsin - Madison, Madison, WI; ³Sch. of Med., Natl. Def. Med. Ctr., Taipei, Taiwan

Abstract: Neural mechanisms of visuospatial memory are essential for behaviors that depend on the processing and storage of visual information for future interactions. Posterior parietal cortex (PPC) has a key role in the processing of visuospatial information, and beta oscillations (15-30 Hz) there have been linked to working memory, attention, and sensorimotor integration—making them a candidate mechanism for memory computations. To investigate the contributions of neurons in PPC to visuospatial working memory, we recorded spiking activity and local field potentials (LFPs) using linear arrays from two rhesus macaque monkeys. We targeted the caudal interparietal (CIP) area for its multiplexed roles in the visuospatial processing of objects and encoding of saccadic eye movements. To dissociate visually driven activity from memory signals

and motor planning, the monkeys performed memory-guided and overlap saccade tasks. In the memory task, a target was briefly presented (100 ms) followed by a delay in which the target location had to be remembered. After the delay, the monkey made a saccade to the remembered location. For the overlap task, the target remained visible throughout the trial, allowing the monkey to saccade to a visible target. Consistent with a role for beta in sensorimotor processing, we found induced 18-25 Hz oscillations in both tasks that manifested as transient bursts in the LFPs. However, beta bursting occurred more frequently and with greater power in the memory delay period, suggesting an additional role in working memory maintenance. In addition, a 100 ms period of transient beta phase-locking at the start of the memory delay was a prominent feature in the population firing rates and LFPs, reflecting precise phase alignment across recording channels and trials. This period may mark a temporal window in which mnemonic representations are stabilized through neural synchronization triggered by the target's disappearance, signaling that the location would need to be recalled. This phase-locking was absent in the overlap task. Supporting that beta phase-locking moderates the visuospatial memory of the target location, the accuracy of saccades, but not the saccade dynamics, depended on the phase and amplitude of the oscillation. These findings support a role for beta phase-locking in mnemonic rather than motor processing. These findings demonstrate that external cues and cognitive demands, such as the need to recall information, moderate endogenous beta oscillations in the parietal cortex to support visuospatial working memory.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

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Program #/Poster #: PSTR300.08/Web Only

Topic: I.05. Working Memory

Support: MOE AcRF Tier 2 Award MOE-T2EP30220-0003
MOE AcRF Tier 1 Award RG110/24
MOE AcRF Tier 1 Award RS01/19
MOE AcRF Tier 1 Award RG19/21

Title: Aging impairs the predictive relationship between network centrality and working memory properties of the prefrontal neurons

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Abstract: The prefrontal cortex (PFC) is essential for higher cognitive functions and is characterized by marked neuronal heterogeneity, with individual neurons displaying diverse activity patterns. To investigate the basis of this variability and its functional relevance, we used

calcium imaging in the medial PFC (mPFC) of mice and examined degree centrality—a measure of a neuron's connectivity within local circuits—during resting-state periods. In young adult mice, we found a strong association between connectivity strength and functional specialization: neurons with high degree centrality showed consistent and stable selectivity for action plans during memory-guided tasks. This indicates that intrinsic connectivity is a key determinant of functional heterogeneity within prefrontal circuits. However, this relationship weakened in middle-aged and older mice, suggesting that aging disrupts the organization and function of prefrontal networks. A computational model incorporating age-related declines in synaptic plasticity successfully recapitulated these age-dependent changes. In young mice, degree centrality also predicted cross-modal action-plan selectivity, a predictive relationship that diminished with age. Moreover, spatial analyses showed that highly selective neurons tended to cluster in young adults, but this spatial organization progressively broke down with aging. Together, these findings demonstrate that aging alters network connectivity, functional architecture, and spatial organization in the mPFC, providing important insights into the neural basis of cognitive decline.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

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Program #/Poster #: PSTR300.09/MM10

Topic: I.05. Working Memory

Support: NIH R01NS120987

Title: Prefrontal dopamine transients and oscillatory activity in D1 receptor-expressing neurons during working memory

Authors: *V. W. KILONZO, Y.-C. KIM, M. A. WEBER, N. S. NARAYANAN;
Neurol., Univ. of Iowa, Iowa City, IA

Abstract: Dopamine in the prefrontal cortex is critical for executive functions such as working memory. However, dopamine dynamics during executive functions are understudied due to the challenging nature of accurately and rapidly sampling prefrontal dopamine in behaving rodents. Recent advances in GPCR activation-based dopamine (GRAB DA) sensors that fluoresce with low concentrations of dopamine allow for temporally precise sub second sampling of dopamine dynamics in the mouse prefrontal cortex. Here, we injected the GRAB DA sensor variant gDA3m and Cre-dependent ChrimsonR into the prefrontal cortex of D1-Cre mice. We then implanted a combined optogenetic and multielectrode recording array to optogenetically tag D1 dopamine receptor-expressing neurons while simultaneously recording prefrontal dopamine dynamics via fiber photometry and neuronal ensembles with electrodes during a delayed non-match to sample working memory task. We focus on prefrontal dopamine dynamics, neuron

spiking activity, and low-frequency oscillations associated with working memory delay-related activity. Our work highlights how neuromodulatory dopamine signals in the prefrontal cortex engage specific D1-dopamine receptor populations to orchestrate neuronal activity and network rhythms underlying working memory.

Disclosures: V.W. Kilonzo: None. Y. Kim: None. M.A. Weber: None. N.S. Narayanan: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.10/MM11

Topic: I.05. Working Memory

Title: Hippocampal input-driven plasticity of prefrontal interneurons reveals a circuit basis for impaired spatial working memory.

Authors: *S. E. SILVERSTEIN;
NINDS/NIH, Bethesda, MD

Abstract: Hippocampal-prefrontal interactions are critical for spatial working memory (SWM), but how long-range inputs from the ventral hippocampus (vHPC) engage medial prefrontal (mPFC) inhibitory microcircuits to support SWM remains unclear. Here, we combined *in vivo* optical stimulation of vHPC inputs to mPFC with calcium recordings of discrete mPFC interneuron populations in male and female wild-type C57BL/6J mice. This approach revealed class-specific response profiles and plasticity. Repeated vHPC input stimulation persistently depressed activity in vasoactive intestinal peptide (VIP)-expressing interneurons and potentiated activity in somatostatin (SST)-expressing interneurons, while minimally depressing parvalbumin (PV)-expressing interneurons. As opposed to the highly plastic interneurons, CaMKII α -expressing putative pyramidal neuron responses were relatively small and stable. *Ex vivo* whole-cell electrophysiology and computational modeling revealed that these divergent effects likely arise from a primary weakening of monosynaptic vHPC input onto mPFC VIP interneurons. Leveraging this plasticity to inform vHPC-mPFC circuit interactions that support SWM, mice were subsequently trained on a delayed non-match-to-sample SWM task. We found that prior vHPC stimulation elevated mPFC VIP interneuron activity during the delay epoch in early training—blunting a dip normally associated with correct choices. This aberrant activity profile predicted poorer learning performance and was recapitulated in Df(16)A^{+/-} mice, a model of 22q11.2 deletion syndrome with known SWM deficits and reduced vHPC input to mPFC VIP interneurons. These findings cell-type-specific plasticity between vHPC inputs and mPFC interneurons and highlight how altering long-range inputs onto VIP interneurons may contribute to working memory dysfunction.

Disclosures: S.E. Silverstein: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.11/MM12

Topic: I.05. Working Memory

Support: McGovern Institute
Simons Foundation
HHMI

Title: Neural dynamics for compositional generalization in 2D vector addition

Authors: *C. TANG¹, M. JAZAYERI^{1,2};

¹MIT, Cambridge, MA; ²Howard Hughes Med. Inst., Boston, MA

Abstract: Compositional generalization, the ability to understand novel combinations of familiar stimuli, is crucial for navigating an environment filled with exponentially varied stimuli. For instance, if an individual in the SfN poster hall knows where [Row 2, Poster 5] and [Row 4, Poster 1] are, they can readily march to [Row 4, Poster 5]. However, the neural basis underlying compositional generalization remains poorly understood. Here, we trained two monkeys on a task that requires compositional generalization and recorded the neural activity in the dorsomedial frontal cortex (DMFC). The monkeys need to solve a vector addition task ($start + vector = target$) in a 2D coordinate system. In each trial, the monkey sees two kinds of stimuli: first, a circle at some coordinate as the *start*; and second, a flow field around the fixation point indicating the *vector* (*defined by both direction and length*). Finally, the monkey needs to make a saccade to the summed coordinate (*target*) to get the reward. Critically, during the behavioral training, the monkeys were trained only on a subset of $start + vector$ combinations (the training set), with a group of held out combinations never exposed to them (the testing set). We then evaluated the monkeys' generalization ability on the testing set. Monkeys demonstrated successful compositional generalization under some testing conditions and failed under others, allowing us to identify the neural signatures associated with successful generalization. Specifically, when monkeys generalized successfully, neural trajectories corresponding to identical vector directions remained largely parallel, indicating a factorized representation where vector encoding was invariant across different starting points. Conversely, failures corresponded with non-factorized neural representations, establishing factorization as essential for successful compositional generalization. Additionally, preliminary findings suggest two distinct neural dynamics underlying vector length encoding: ramping neural activity distinguished short from long vectors, while oscillatory patterns emerged as monkeys learned multiple discrete target positions along the same directional axis. Collectively, our findings demonstrate that compositional generalization in 2D vector addition tasks is supported by neural computations that factorize the *start* and *vector* components and encode vector magnitude, representing an effective 'coordinate-like' mental model of external spatial tasks.

Disclosures: C. Tang: None. M. Jazayeri: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.12/MM13

Topic: I.05. Working Memory

Support: NIH Grant 1R90DA060338 (TPCN Training Program)

Title: From recurrent structure to memory robustness: a dynamical analysis of a firing rate model

Authors: *Z. GUO¹, J. JARAMILLO²;
²Neurobio., ¹Univ. of Chicago, Chicago, IL

Abstract: Working memory (WM) refers to the capacity to temporarily maintain and manipulate information to guide behavior. This function is thought to rely on persistent neural activity—neural activity that represents and outlasts the duration of a stimulus to be remembered. Persistent activity has been observed in frontal, temporal, and parietal cortices during the stimulus-absent delay periods of various cognitive tasks. Critically, WM-related persistent activity is susceptible to interference from non-relevant stimuli or distractors during the delay period. In this project, we aim to identify the circuit-level mechanisms behind the robustness of WM persistent activity to distractors. We employed a recurrent neural circuit model derived from Wong & Wang (2006), in which selective populations interact via mutual inhibition and are sustained by slow NMDA-mediated recurrent excitation. The overall recurrent strength is defined by the composite parameter $J_S = J_E - J_I$, where J_E denotes self-excitation and J_I represents cross-inhibition. This architecture supports multistable attractor dynamics associated with memory maintenance and categorical decision-making (Wong & Wang, 2006; Murray et al., 2017). We simulated population responses to cue and distractor inputs in a reduced firing-rate framework and analyzed the resulting dynamics in a two-dimensional phase plane defined by synaptic gating variables. Increasing J_S expanded the basin of attraction surrounding target memory states, indicating enhanced resilience to interference. To quantify this change, we estimated the basin boundary area as a function circuit-level parameters, with J_S playing a dominant role. We further investigated how external input interacts with the attractor structure by segmenting the fitted boundary into functionally distinct dynamical regimes. In line with Murray et al. (2017), we observed that larger values of J_S require stronger perturbations to disrupt memory representations, revealing a trade-off between attractor stability and input sensitivity. Collectively, our results provide a low-dimensional framework for understanding WM robustness through the lens of attractor geometry.

Disclosures: Z. Guo: None. J. Jaramillo: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.13/Web Only

Topic: I.05. Working Memory

Title: Load- Dependent Neural Dynamics of Working Memory Capacity in Freely Moving Marmosets

Authors: *T. LO¹, S. VIJAYRAGHAVAN², L. E. MULLER³, J. MARTINEZ-TRUJILLO⁴;
¹Western Univ., London, ON, Canada; ²Western Univ., LONDON, ON, Canada; ³Dept. of Mathematics, ⁴Dept. of Physiol. and Pharmacol. and Psychiatry, Western Univ., London, ON, Canada

Abstract: Working memory (WM) is a crucial cognitive function that supports the temporary retention and manipulation of information essential for goal-directed behavior. This capacity is inherently limited and varies across species, tasks, and sensory modalities. To investigate the behavioral and neural constraints of WM capacity, we trained two common marmosets on a touchscreen-based delayed non-match-to-position (DNMP) task. In each trial, one to four spatial stimuli were presented sequentially, and subjects were required to select the novel location in each memory-guided response. Behavioral performance declined with increasing memory load, consistent with capacity limits observed in other primate species. To probe the neural correlates of this behavior, both marmosets were implanted with wireless multi-shank electrode arrays targeting lateral prefrontal cortex (area 46). Spike activity was recorded during task performance and analyzed across three key task epochs: pre-touch (delay), post-touch (selection), and post-reward (feedback). Many neurons exhibited load-sensitive tuning to specific screen quadrants, with spatially selective responses distributed broadly across the electrode array. This supports the notion that spatial representations in prefrontal cortex are maintained through a distributed rather than strictly retinotopic organization. To uncover low-dimensional structure in these neural dynamics, we applied principal component analysis (PCA) and visualized the resulting latent trajectories across memory loads. Population activity occupied distinct manifolds depending on memory load and trial phase, suggesting that prefrontal ensembles flexibly reconfigure as task demands increase. These load-dependent shifts in neural manifold geometry reflect the evolving demands of spatial memory maintenance and selection, potentially constraining behavioral performance as capacity limits are approached. Understanding how such neural dynamics support WM in marmosets provides a foundation for comparative models of cognitive function and its disruption in neuropsychiatric disease.

Disclosures: T. Lo: None. S. Vijayraghavan: None. L.E. Muller: None. J. Martinez-Trujillo: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.14/MM14

Topic: I.05. Working Memory

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Title: Reusable modular architecture enables flexible cognitive operations

Authors: *Y. OSAKO¹, G. R. HELLER², T. OSAKI³, T. BUSCHMAN⁴, M. SUR⁵;
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Abstract: Abstract: Flexible behaviors emerge from the coordination and reuse of multiple distinct cognitive processes. Theoretical studies have posited that artificial neural networks achieve this flexibility through modular architectures, where small functional modules, each specialized for specific computations, are reused across tasks. Despite its theoretical significance, empirical evidence for reusable modular networks in animal brains has been scarce. Here, we provide evidence for such a network architecture in the mouse brain during a delayed match-to-sample with delayed report (DMS-dr) task. Using population-level and geometric analyses, we identified reusable subspaces for each cognitive operation involved in the task, including stimulus processing and memory maintenance. Model-free clustering analysis revealed functionally distinct clusters in medial prefrontal cortex (mPFC) and posterior parietal cortex (PPC), each selectively contributing to a different subspace. Recurrent neural networks fit to the observed neural dynamics demonstrated that silencing specific clusters disrupted specific computational functions rather than specific representations, highlighting the modular and reusable organization of these networks. To causally test how this reusable modular architecture contributes to behavior, we are developing a two-photon microscopy-compatible platform for selective inhibition of identified neuronal populations. Our all-optical approach combines two-photon calcium imaging using red-shifted (RCaMP3) or green-shifted (GCaMP6) indicators with targeted optogenetic stimulation of neurons expressing a blue light-sensitive opsin (GtACR2) in awake mice. We thus expect to dissect online how modular neuronal subpopulations are reused for flexible brain function and behavior.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.15/MM15

Topic: I.05. Working Memory

Title: Optogenetic manipulation of thalamocortical circuits during working memory

Authors: ***R. MILTON**^{1,2}, **L. BARTLETT**^{1,2}, **R. OLSON**^{1,2}, **A. SONNEBORN**^{1,2}, **A. I. ABBAS**^{1,2};

¹Oregon Hlth. Sci. Univ., Portland, OR; ²Veterans Affairs Portland Hlth. Care Syst., Portland, OR

Abstract: The mediodorsal thalamus (MD) and medial prefrontal cortex (mPFC) are anatomically and functionally interconnected regions implicated in the maintenance of working memory (WM). While activity in this circuit has been associated with WM-related processes, it remains unclear whether the relevant information is primarily encoded in persistent spiking activity or stored in more latent, activity-silent forms. To address this, we trained mice on a delayed match-to-sample (DMTS) task involving randomized port locations and variable delay periods, requiring flexible encoding and retention of spatial information. After achieving stable performance, animals were implanted for dual-site electrophysiology and optogenetic manipulation of MD→mPFC projection neurons. These projections were activated with laser pulses (5ms at 5mW) at different frequencies during the sample or delay phase of the task to assess causal contributions to WM performance and neural encoding formats. Stimulation during the delay period produced frequency-dependent behavioral effects, with lower frequencies generally associated with modest improvements in accuracy. Importantly, we observed that stimulation during the delay phase elicited structured, trial-specific spike responses in mPFC and MD that were absent in non-stimulated trials. Using a phase-based analysis, we quantified the difference in evoked spike timing patterns between different trial types and found evidence for stimulation-dependent reactivation of task-relevant information. These results suggest that the MD→mPFC pathway supports WM not only through sustained activity but also by maintaining information in a form that can be selectively revealed by targeted input. Our findings highlight the dynamic nature of WM representations and provide a framework for using temporally precise stimulation to access otherwise hidden neural states.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.16/MM16

Topic: I.05. Working Memory

Title: Layer-specific and temporally structured dynamics in the mouse prefrontal cortex underlying working memory processing

Authors: *S. KO¹, J. BAE², M. W. JUNG¹;

¹KAIST/IBS, Daejeon, Korea, Republic of; ²Dept. of Brain and Cognitive Sci., MIT, Boston, MA

Abstract: The laminar organization of the neocortex is a fundamental feature of the mammalian brain, yet how distinct cortical layers contribute to higher-order cognitive functions such as working memory remains incompletely understood. To investigate this, we conducted multi-laminar recordings using Neuropixels probes in the medial prefrontal cortex of mice engaged in a delayed response task, in which an LED cue was presented on either the left or right lick port, followed by a delay period after which the animals were required to choose the matching port to obtain a water reward.

Deep-layer neurons showed stronger directional selectivity across all task epochs—sample, delay, and choice—indicating a temporally stable representation of directional information. In contrast, superficial-layer neurons showed enhanced correct-error selectivity specifically during the late delay period, suggesting a role in decision-related processes. Cross-correlation analyses revealed dominant intra-laminar interactions within deep layers during the sample and choice epochs, while superficial-layer interactions peaked during the late delay. Notably, inter-laminar coupling emerged mid-delay and intensified toward the end of the delay period.

Together, these findings suggest a dynamic division of labor across cortical layers: deep layers are primarily involved in maintaining stimulus-direction encoding, while superficial layers contribute to accurate decision formation. These functions are associated with temporally organized intra- and inter-laminar interactions, highlighting the importance of laminar circuit dynamics in the prefrontal cortex underlying working memory processing.

Disclosures: S. Ko: None. J. Bae: None. M.W. Jung: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

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Program #/Poster #: PSTR300.17/MM17

Topic: I.05. Working Memory

Support: NSF NCS DRL
NIH NINDS T32

Title: Transformations from memory to action in prefrontal and motor cortices

Authors: *A. CHANDRASEKARAN¹, M. MCDONNELL², C. KI², B. M. YU⁴, A. BATISTA⁵, S. M. CHASE², M. A. SMITH³;

¹Univ. of Pittsburgh Ctr. For Neurosci., Pittsburgh, PA; ²Neurosci. Inst., ³Biomed. Engin. and Neurosci. Inst., Carnegie Mellon Univ., Pittsburgh, PA; ⁴CMU, Pittsburgh, PA; ⁵Bioengineering, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: We are constantly inundated by information. To navigate a rich sensory world, it is essential to select and maintain relevant information in memory to guide our actions. What neural mechanisms prevent irrelevant, distracting input from interfering with our memory? What goes awry when we succumb to distraction? To study this, we trained a rhesus monkey to perform a memory-guided reach task with distractors. On each trial, a target was shown, followed by a distractor at an independently chosen location. After a delay, the monkey had to reach to the target while ignoring the distractor. We recorded from neurons in prefrontal cortex (PFC), a key node in working memory maintenance and distractor suppression, and premotor (PMd) and primary motor (M1) cortices which are involved in motor planning and execution. We assessed target and distractor information in each area by training decoders to predict target and distractor location from neural activity at each time point during the delay. We found that target information appeared in all areas shortly after target onset and was present throughout the delay period. Conversely, distractor information was neither universally nor persistently present. In PFC, distractor information appeared transiently after distractor presentation but decayed quickly. PMd showed similar trends but had much weaker distractor information compared to PFC, while M1 contained nearly no distractor information.

How are both target and distractor information arranged within PFC to prevent interference? We found that PFC employed distinct codes for target location during the visual onset and memory periods. Both target and distractor transiently drove activity along a visual subspace. However, only the target information was then shifted to a memory subspace that persisted throughout the delay period. By selectively routing task-relevant information to the memory space, PFC population activity shielded target representations from incoming distractors. To understand what happens when the animal succumbs to distraction, we separately analyzed error trials where he reached to the distractor location. We found that target information in the memory subspace of PFC, PMd, and M1 was overwritten by the distractor information. Further, we found that target information in PMd before the distractor onset was weaker on these error trials compared to successful trials. This enabled us to predict, on a trial-by-trial basis, when such errors were more likely to occur. In summary, we provide evidence for how prefrontal and motor cortical activity selects relevant information to guide motor planning while suppressing irrelevant distracting input.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

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Program #/Poster #: PSTR300.18/MM18

Topic: I.05. Working Memory

Support: R01MH115042

Title: Neural synchronization supports spatial attention and selection from working memory

Authors: *P. IAMSHCHININA¹, M. PANICHELLO², T. BUSCHMAN³;

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Abstract: Adaptive behavior relies on prioritizing task-relevant information, whether by attending to sensory inputs from the environment or selecting items held in working memory (WM). Previous research has identified several neural mechanisms used for attention, including neural synchronization (Buschman et al., 2012). However, less is known about how items are selected from WM (van Ede & Nobre, 2023). Recent neurophysiological evidence suggests that attention and selection share a common mechanism (Panichello & Buschman, 2021). Like attention, selection enhanced memory representations and transformed them into a neural subspace that guided behavior. Here, we investigate the role of synchrony in WM selection, posing two questions: 1) Does attention and selection recruit the same synchronized neural ensembles in the prefrontal cortex (PFC)? 2) Does selection modulate synchrony patterns encoding WM, and do these patterns reflect the observed representational transformation? To answer these questions, we analyzed local field potentials recorded from multiple brain regions. Two rhesus macaques switched between tasks requiring them either to attend to an “upper” or “lower” item or select one of the items from WM. We found that an ensemble of electrodes in PFC synchronized at ~3-11 Hz when animals attended to a specific location and it also synchronized when the same item was selected from WM. This suggests synchrony may be a generalized mechanism for both attention and selection. Next, we found that the pattern of synchrony varied depending on the remembered item. Synchrony increased when the item was selected, consistent with prior attention research. WM selection also affected the similarity of item-specific patterns: before the selection, the synchrony patterns for the “upper” and “lower” items were uncorrelated presumably to reduce interference. After the selection, item-specific information generalized across locations. Altogether, our results introduce a possibility that transiently synchronizing location-specific ensembles may prompt population-level representational shift during spatial attention and WM selection.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

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Topic: I.05. Working Memory

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Title: A new mathematical transform for large-scale spike patterns reveals how ketamine shapes dorsolateral PFC circuits during cognition

Authors: *A. BUSCH^{1,4}, M. ROUSSY¹, B. W. CORRIGAN⁵, R. A. GULLI⁶, R. BUDZINSKI^{2,4}, J. C. MARTINEZ-TRUJILLO³, L. MULLER^{2,4};
²Mathematics, ³Dept. of Physiol. and Pharmacol. and Psychiatry, ¹Western Univ., London, ON, Canada; ⁴Fields Lab. for Network Computation, Fields Inst., Toronto, ON, Canada; ⁵Biol., York Univ., Toronto, ON, Canada; ⁶Zuckerman Inst., Columbia Univ., New York City, NY

Abstract: Ketamine can provide rapid symptom relief for Major Depressive Disorder (MDD) (Yavi et al., *Discov. Ment. Health*, 2022). The best current hypothesis for ketamine's rapid efficacy is that it re-organizes cortical circuits in the frontal lobe to reshape or reset cognitive patterns. Evidence at the cellular level suggests that ketamine may promote neuroplasticity (Yavi et al., *Discov. Ment. Health*, 2022). Evidence at the network level suggests ketamine alters the functional connectivity of networks implicated in MDD (Crisanti et al., *J. Affect. Disord.*, 2020). However, despite ongoing clinical trials, a major gap exists between ketamine's effect at the receptor level and how it could shape local microcircuits to affect cognition. Working memory (WM) — a core executive function supporting active thought (Baddeley, *Rev. Gen. Psychol.*, 2013) — is frequently impaired in MDD, making WM a key focus in this field. In previous work, we found that sequences of spiking activity across 100s of neurons in dorsolateral PFC (dlPFC) of non-human primates (NHP) encode target locations held in working memory during a virtual reality (VR) WM task (Busch et al., *Nat. Commun.*, 2024). We have now developed a new mathematical tool, called the multi-sample Discrete Helix Transform (ms-DHT), that characterizes intricate patterns of spikes across neurons, beyond simple sequences. When applied to spikes from NHPs in the WM VR task, the ms-DHT reveals fine-scale structure that allows more detailed predictions of behaviour, such as the specific errors subjects make in this task (Busch et al., *arXiv*, 2024). In separate recording sessions, low doses of ketamine (0.2-0.7 mg/kg) were systemically administered to NHPs performing the VR-WM task, temporarily disrupting WM performance (Roussy et al., *Mol. Psychiatry*, 2021). We now use the ms-DHT to study the impact of ketamine on dlPFC circuit function. At first, a distinct spike pattern encoded each target location. After ketamine administration, each target was remapped to a new spike pattern — but distances between spike patterns were preserved by this remapping. This demonstrates the specific spike patterns used by dlPFC for each target change after ketamine, but the relationships between patterns are preserved. Critically, the reorganization of spike patterns during the WM delay persists even after behavioural performance recovers, revealing a long-lasting effect of ketamine in reshaping dlPFC circuit dynamics specific to WM. These results demonstrate, for the first time, how ketamine can reshape a dlPFC circuit essential for cognition,

potentially generating new and experimentally testable hypotheses about its efficacy in treating MDD.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.20/NN2

Topic: I.05. Working Memory

Title: Compositional Compression of Sequence Working Memory Using Geometrical Primitives in Macaque Prefrontal Cortex

Authors: *H. ZHANG^{1,2}, Y. LI³, Z. TIAN^{1,2,5}, G. FU¹, Y. XIE⁴, L. WANG^{1,2};

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Abstract: The mechanism through which relational structures facilitate the compressed neural coding of long sequences in working memory remains unclear. To address this, we trained macaques on a visuospatial delayed sequence working memory task that involved compressing four-item sequences using inferred geometrical primitives. Using two-photon calcium imaging in the prefrontal cortex, we recorded thousands of neurons. We demonstrated that the neural state spaces lacked representations of individual items but contained subspaces dedicated to primitives during memory maintenance. This compressed coding persisted despite changes in the spatial layouts of items. Moreover, we also observed neural dynamics of these primitives directing the decompression of the code back to individual items during memory retrieval. These findings lay the groundwork for understanding how the brain encodes long sequences in a compressed format and decompresses them during retrieval.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

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Program #/Poster #: PSTR300.21/NN3

Topic: I.05. Working Memory

Title: Constructing hierarchical control of sequence working memory in the macaque prefrontal cortex

Authors: X. LI¹, *J. LI², B. MIN³, L. WANG⁴;

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Abstract: The hierarchical organization of actions is essential for executing complex behaviors. However, the neural codes of hierarchical structure and the interaction between different hierarchy levels remain under-explored. In this study, we train monkeys to perform a novel “sequences of sequences” task, where the order of item-level spatial locations in the sequence working memory (SWM) were controlled by a higher rule-level sequence, constructing multi-level hierarchies. Behavioral results show that monkeys successfully learned the hierarchy, enabling them to execute a significant number of item-level SWM tasks consecutively. Electrophysiological recordings from the prefrontal cortex revealed shared orthogonal memory subspaces across tasks with varying rules, indicating reusable codes for item-level sequences. The rule-level sequence followed dynamical neural codes over a larger timescale, evolving according to the hierarchical structure. We observed a grid-like map of rules that transiently emerges during the inter-trial interval, which indicates the monkeys knew their location within the hierarchy. The two levels interact during the input period, when the rule signal intervenes the entry process of SWM. We further identified neuron populations that account for this control process. Finally, we examined the spatial arrangement of neural codes and discovered a rostro-caudal gradient in prefrontal subregions, with more abstract information (i.e., rule) encoded in the anterior regions. Together, these findings offer a clear insight into how the primate brain supports hierarchical representation and interaction of working memory.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

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Topic: I.05. Working Memory

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the CAS Strategic Priority Research Program XDB1010202

Title: The Decomposition of Conscious Access: Geometry of Working Memory for Masked Sequences in the Prefrontal Cortex

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Abstract: Conscious perception integrates sensory inputs with internal models, yet the neural mechanisms remain elusive. This study explores the role of the prefrontal cortex (PFC) in conscious access using a delayed-masked-sequence reproduction task in monkeys, where stimulus intensity varied from 0 to 100%. Employing two-photon calcium imaging and electrophysiological recordings, we found that conscious access to sequences involves a two-step process: initial continuous activity in a sensory subspace, followed by sustained, all-or-none metastable activity in rank-selective working memory (WM) subspaces within the PFC. Remarkably, the PFC circuit could be volitionally activated without sensory input (e.g., at 0% contrast), akin to mental imagery, underscoring the role of top-down control. We propose that this compositional architecture—binding transient sensory inputs to structured internal models via rank-selective subspaces—represents a general computational strategy for conscious cognition. This mechanism may extend beyond sequence working memory to domains like perception, language, and action planning, where transient inputs are integrated into coherent internal representations. Furthermore, these sequential compositional dynamics may elucidate the unity of conscious experience, providing a unified framework for understanding consciousness as an emergent property of structured internal control.

Disclosures: H. Zhou: None. L. Wang: None.

Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR300.23/NN5

Topic: I.05. Working Memory

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Title: Frontal cortical ripples associated with mixed selectivity neurons mediate sequence construction in macaques

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Abstract: When closely timed consecutive stimuli enter working memory (WM), the brain concatenates them into sequences that could be both faithfully reproduced and reorganized as needed. In both rodents and primates, brief bursts of high-frequency local field potential (LFP) oscillations, or “ripples”, are found across the brain, often accompanying the formation of long-term sequential spatiotemporal associations during sleep/rest periods. Recent works on neocortical ripples further suggest that they may serve a similar binding function during waking, but whether and how ripples support the formation of sequential associations in WM remain largely unexplored. Since sequence item representations have previously been identified in the macaque frontal cortex, we trained two monkeys on a WM task involving visuospatial sequence reproduction, and recorded frontal cortical LFP alongside single unit spikes with multi-channel electrode arrays during task performance. We found that, in the case of two-item (stimuli S1 and S2) sequences, neurons with mixed selectivity for both items after S2 onsets were preferentially derived from those best representing S1, and could be used to decode sequence identities. Crucially, ripples tend to co-occur with mixed selectivity neurons on the same channels post-S2-onset, and ripple occurrence rates could in turn predict these neurons’ contributions to WM. Similar results were also observed with three-item sequences. Furthermore, sequence re-ordering after initial construction during encoding was also linked to the emergence of additional ripples and mixed selectivity neurons. Overall, these findings suggest that frontal cortical ripples may support WM sequence construction, reminiscent of their roles in long-term memory functions.

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Poster

PSTR300: Central and Prefrontal Mechanisms of Working Memory in Animals

Location: SDCC Halls B-H

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Program #/Poster #: PSTR300.24/NN6

Topic: I.05. Working Memory

Title: Prefrontal dopamine and serotonin dynamics during delay-dependent decision making

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Abstract: The ability to represent and maintain information across delays is critical for working memory (WM) and higher-order cognition. Prior research has implicated monoamine signaling, particularly dopamine (DA) and serotonin (5-HT) within the prefrontal cortex (PFC), as playing an important modulatory role in delay-dependent processes. However, the phasic dynamics of prefrontal DA and 5-HT during delay-dependent decision-making remain largely unexplored. To investigate this, we expressed genetically encoded fluorescent GPCR Activation Based (GRAB) sensors for DA and 5-HT in the medial PFC and used fiber photometry to record neuromodulator dynamics during performance of two novel touchscreen-based tasks. In the rodent Self-Ordered Working Memory (rSOWM) task, mice were trained to touch 2-3 visual stimuli on each trial, earning milk rewards when choosing each unique stimulus location. Revisiting a stimulus location within a trial resulted in an error and timeout. Trials varied by stimulus number, spatial separation, and delay interval (0 or 6 s). In the rodent Delay and Reward Discounting (rDRD) task, mice choose between two visual stimuli, each associated with a different combination (16 total) of reward magnitude (15, 25, 35, 50 μ L) and delay (0, 2, 4 or 6s). In both tasks, DA and 5-HT exhibited opposing phasic dynamics following key behavioral reinforcement-related events (e.g., stimulus choice and reward collection). Interestingly, monoamine dynamics differentially predicted choice outcomes across tasks. In rSOWM, elevated DA levels in medial PFC during the delay period were associated with subsequent correct choices, selectively on more difficult trials (e.g., 3 stimuli, 6s delay). 5-HT dynamics were not related to rSOWM choice outcomes. In contrast, elevated 5-HT in medial PFC was associated with increased selection of immediate (no-delay) rewards. DA dynamics were not predictive to DRD response choice. Our findings of opposing phasic modulation of DA and 5-HT in the medial PFC during task reinforcement-actions are consistent with recent findings in striatum and suggest a potential functional interaction between the two monoaminergic systems in guiding reinforcement-driven behavior. Moreover, these results reveal task-specific and dissociable roles for phasic DA and 5-HT signaling in medial PFC, supporting their contributions to delay-dependent cognitive processes: spatial working memory and reward discounting.

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Poster

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Topic: I.11. Language

Support: NSF DRL 2219739
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Title: Exploring Whole-Brain Neuroanatomical Divergence Across Great Ape Species

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Abstract: Humans are a member of the great ape clade. Other extant great apes include chimpanzees (*Pan troglodytes*) and bonobos (*Pan paniscus*), to whom humans are most closely related, and orangutans (*Pongo spp.*) and gorillas (*Gorilla spp.*), to whom humans are more distantly related. These species offer valuable insights into the evolution of brain structure and function, providing a comparative framework to understand the neural substrates that underlie human cognition. This study investigates structural differences in brain organization across all five extant great ape species (orangutans, gorillas, bonobos, chimpanzees, and humans). Our dataset includes 180 scans, comprising both structural and diffusion-weighted imaging (DWI) data, which were preprocessed and analyzed using FSL and ANTS software packages. To facilitate comparisons of structural features, brain templates and atlases were created for each species. Within- and between-species variation in gray matter morphology and white matter connectivity was assessed, with probabilistic tractography employed for the connectivity analysis. The results are interpreted in the context of phylogenetic divergence within the great ape clade and the socioecological adaptations unique to each species. By comparing these species, valuable insights are gained into the distinctive features of the human brain and the evolutionary processes that have shaped its neural architecture.

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Poster

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Topic: I.06. Social Cognition

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Title: Chemogenetic inhibition of the default mode network selectively reduces attention to social interaction in macaque monkeys

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Abstract: The default mode network (DMN) is a large-scale brain network comprising regions such as the posterior parietal cortex (PPC) and posterior cingulate cortex (PCC). In humans, the DMN has been implicated in internally oriented processes, including self-referential thought and social cognition, and its dysfunction is associated with psychiatric conditions such as autism spectrum disorder and schizophrenia. However, the causal contributions of the DMN to social cognition remain poorly understood due to the complexity of the network. To address this, we chemogenetically manipulated key DMN nodes in nonhuman primates and assessed the impact on network connectivity and social attention. Four macaque monkeys received injections of adeno-associated virus vectors to express either an inhibitory designer receptor (hM4Di; N=2) or a combination of hM4Di and an inhibitory designer channel (PSAM4-GlyR) in the PPC and PCC nodes. Systemic administration of the corresponding agonists—deschloroclozapine (DCZ) and uPSEM817 (0.1 mg/kg)—induced targeted chemogenetic inhibition, confirmed by significant reductions in local BOLD signals ($p < 0.001$) using pharmacological MRI. Resting-state fMRI showed that inhibition of either PPC or PCC alone produced only limited reductions in DMN functional connectivity. In contrast, simultaneous inhibition of both nodes substantially disrupted intra-DMN connectivity (27% reduction, $p < 0.05$), indicating that PPC and PCC contribute synergistically to maintaining DMN activity. Functional connectivity within other large-scale networks remained unchanged, highlighting the specificity of DMN suppression. To assess behavioral consequences, two monkeys were tested using a free-viewing paradigm in which they explored three categories of visual stimuli: background scenes, a single monkey, and two monkeys engaging in social interaction. DMN suppression did not affect gaze allocation to background scenes or single monkeys. However, during social scene presentation it selectively altered gaze patterns—reducing gaze toward interaction regions between monkeys while increasing gaze toward individual monkey faces. These results provide causal evidence that DMN activity is critical for guiding attention toward socially relevant interactions, supporting its role in a core aspect of social cognition.

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Poster

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Program #/Poster #: PSTR301.03/NN9

Topic: I.06. Social Cognition

Title: Flexible V4-dlPFC communication enhances context-appropriate visual encoding in dlPFC

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Abstract: The brain has a remarkable ability to assign different meanings to the same visual input depending on context—for instance, perceiving a friend as a competitor in an exam but as a companion at a social gathering. Such flexibility enables adaptive behavior and requires the brain to dynamically reconfigure how it processes and routes visual information. In primates, visual cortex (area V4) and dorsolateral prefrontal cortex (area dlPFC) work together to process complex visual information and enable context-appropriate decision-making. However, it remains unclear how these areas communicate when visual input must be interpreted differently depending on moment-to-moment context. To investigate this, we recorded neuronal population activity from V4 and dlPFC in two freely moving rhesus macaque pairs during a social cooperation task. Wireless neural and eye-tracking data were collected while monkeys worked together to earn food rewards. We defined the task period as the social context and the inter-trial interval as the non-social context. Within each context, we analyzed population responses during fixations on the partner, reward system, or non-social stimuli. We used linear discriminant analysis (LDA) to classify visual cues based on population activity and compared decoding performance across contexts. Decoding accuracy in dlPFC was significantly higher during engagement, while V4 showed modest differences. To assess encoding changes, we tested how well a decoder trained in one context generalized to the other. We observed greater drops in decoding performance in dlPFC than V4, indicating stronger context-dependent representational changes in dlPFC. Additionally, using residual-based reduced-rank regression, we found that V4→dlPFC communication was selectively enhanced during engagement, but only for social cue processing. Our results indicate that while V4 maintains relatively stable encoding, engagement-dependent communication reshapes and enhances dlPFC representations of social environment. That is, moment-to-moment dynamic communication from V4 to dlPFC enables flexible re-representation of visual information in prefrontal cortex, which is a mechanism playing a key role in guiding context-appropriate decision-making.

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Poster

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Topic: I.06. Social Cognition

Title: Behavioral Strategies for Cooperation in Unequal Reward Conditions in Freely Moving Macaques

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Abstract: Cooperative behavior in primates often depends on more than just equal outcomes. In real-world social settings, individuals must navigate imbalanced reward environments and uncertain returns when deciding whether to cooperate with a partner. We designed a naturalistic, freely moving task, by examining how macaques cooperate under varying reward conditions, including trials in which one monkey receives little to no benefit. The behavior of pairs of monkeys was recorded by an overhead camera while oculomotor events in one of the animals were measured by a wireless eye tracker. Behaviorally, we found that the delay to initiate cooperation was significantly longer in selfish and altruistic conditions compared to fair ones (1-2 s in fair vs. 54-93 s in altruistic/selfish conditions), suggesting increased internal evaluation or sensitivity to partner's behavior. To identify the strategies macaques use to support or resist cooperation, we used the information from synchronized video and pose tracking to capture detailed social dynamics. We observed specific behaviors that appear to promote cooperation, including sustained proximity to the apparatus, re-engagement after partner disengagement, and sustained social fixations. In contrast, avoidance behaviors such as turning away, leaving the apparatus, or avoidance of partner fixation often occurred in trials with unequal outcomes. To quantify how these patterns unfold over time on a trial-by-trial basis, we applied Markov models and transition-based analyses to test how past partner behavior predicts future behavioral responses. Pose tracking allowed us to extract continuous measures of orientation, distance, and movement, which we used to characterize approach, disengagement, and protest-recovery sequences across conditions. These findings indicate that macaques use flexible, context-dependent strategies during cooperation and adapt their social behavior based on both recent outcomes and their partner's actions. This work provides a framework for identifying how primates coordinate behavior in naturalistic environments, even in the face of unequal reward.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

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Topic: I.06. Social Cognition

Support: 1U01NS108680

Title: Neural dynamics of unfairness and cooperation in non-human primates

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Abstract: Cooperation and sensitivity to inequity are essential features of primate social structure, promoting group cohesion and long-term survival. These behaviors rely on the ability to detect and integrate social context, yet the neural mechanisms that support this integration in naturalistic settings remain poorly understood. To address this gap, we investigated how social variables such as reward equity and partner outcome are encoded and communicated across cortical areas during cooperative interactions. Using a novel task in which pairs of macaques visually interact and move freely, we recorded wireless neural activity from visual area V4 and dorsolateral prefrontal cortex (dlPFC), alongside wireless oculomotor and behavioral tracking, while monkeys cooperated for varying amounts of reward. To assess coordination, we computed cross-correlations of button-press behavior across trial types. Peak cross-correlation values were highest in high-equal (4:4 reward) trials, lowest in low-equal (1:1 reward) trials, and intermediate in unequal conditions, suggesting stronger behavioral synchrony when cooperation is maximally rewarding. Eye-tracking revealed that fixation durations were significantly longer in high-equal and disadvantageous conditions ($p < 0.05$), consistent with increased visual monitoring when cooperation is either highly motivating or socially complex. We next asked whether fixation behavior reflects task conditions at the neural level. Using fixation-aligned neural activity, we decoded condition identity from both dlPFC and V4, with pairwise classification accuracy significantly above chance. Interareal analyses revealed stronger subspace communication between V4 and dlPFC in fair conditions compared to unfair ones, suggesting enhanced integration of visual and social information prior to cooperative behavior. Together, these findings show that macaques adapt both their behavior and cortical processing based on social context, using coordinated actions, visual attention, and network-level dynamics to support flexible cooperation even in the face of unequal outcomes.

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Poster

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Program #/Poster #: PSTR301.06/NN12

Topic: I.06. Social Cognition

Support: R01MH128190

Title: Social gaze encoding in the prefrontal cortex and the amygdala depends on mutual interactivity states during naturalistic gaze interactions

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Abstract: In humans and nonhuman primates, gaze interactions play a crucial role in shaping social dynamics. However, it remains unclear how the neural representations of social gaze behavior depend on the levels of mutual interactivity during social gaze exchanges. To investigate this, we simultaneously monitored the gaze behaviors in six unique pairs of macaques (M1 and M2) as they participated in naturalistic social gaze exchanges without any task constraints. During these gaze interactions, we recorded single-neuron activity from multiple brain regions in the prefrontal-amygdala networks including the anterior cingulate cortex, dorsomedial prefrontal cortex, orbitofrontal cortex, and the basolateral amygdala. We first verified that the gaze behaviors of M1 and M2 were indeed dependent on each other in a task-free setting. We found that the product of the probability of M1 looking at M2's face, $P(M1_{\text{face}})$, and that of M2 looking at M1's face, $P(M2_{\text{face}})$, does not equal the observed probability of mutual face-viewing, $P(M1_{\text{face}} \& M2_{\text{face}})$, indicating that the act of looking at each other's faces were not independent between the pairs of macaques. Notably, sizable proportions of neurons in these brain regions exhibited differential responses to face fixations made by M1 depending on whether the fixation was made during the periods of high or low mutual interactivity (periods of high or low occurrences of mutual face fixations). Further, when we projected the activity timeseries of each recorded neuron corresponding to M1's face fixations during high and low interactivity periods onto principal components, the trajectories during high-interactivity fixations were shifted in space compared to those during the low-interactivity fixations. Our findings thus indicate that both single-neuron representations and population dynamics of social gaze in the prefrontal cortical areas and the amygdala critically depend on the state of mutual interactivity. Taken together, our findings demonstrate that neural representations of social gaze in the primate brain can be dramatically altered by the state of social engagement.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

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Program #/Poster #: PSTR301.07/OO1

Topic: I.06. Social Cognition

Title: Investigating the Internal States of Social Decision-Making in Macaques via Latent Variable Modeling

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Abstract: Understanding social decision-making requires the dynamic integration of contextual information and internal motivational states. To uncover latent behavioral dynamics underlying prosocial and antisocial choices, we applied a Generalized Linear Model- Hidden Markov Model (GLM - HMM) to trial-by-trial choice data from macaques performing the social reward allocation task. On each trial, actor monkeys chose between rewarding themselves (Self) and both themselves and a partner monkey (Both), and also between rewarding the partner (Other) and no one (None). The task context varied between Self/Both and Other/None, enabling dissociation of prosocial versus antisocial preferences in distinct social frames. To understand the latent social states underlying prosocial and antisocial decisions, we trained the GLM - HMM on over 1000 trials of sequential monkey choice data, covarying by the two contexts. The model inferred multiple latent states that were stable over time and selectively engaged during behaviorally distinct phases. In the Self/Both context, one state strongly favored the prosocial "Both" choice, while others favored the antisocial "Self" choice. On the other hand, in other/none context, these states were reconfigured to favor the prosocial "other" decision over the antisocial "none" option. These context-dependent findings are demonstrated using emission probabilities that revealed a general increase in prosocial behavior when shifting from Self/Both to the Other/None context.. The emission and transition patterns suggest that the monkeys adjusted their preferred decisions depending on the task conditions. These findings demonstrate that other-regarding social decisions are governed by a small number of context-sensitive latent states. By combining HMMs with interpretable GLM emissions, we reveal how internal dynamics and external context jointly structure social behavior, which can help provide novel insight into the neural mechanisms of social cognition.

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Poster

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Topic: I.06. Social Cognition

Support: R01MH128190

Title: Investigating inequity sensitivity and its underlying facial movement dynamics in monkeys

Authors: *S. BAEK¹, M. INCEBOZ², G. QI¹, S. W. CHANG¹;

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Abstract: Understanding how nonhuman primates evaluate unequal outcomes provides critical insight into the foundations of social cognition. While it is known that monkeys can detect and respond to imbalances in reward distribution, it is less understood how inequity sensitivity

unfolds in a temporally structured setting that requires dynamic, moment-by-moment behavioural adjustments. To address this, we developed a novel paradigm in which two rhesus macaques sat face-to-face and viewed visual cues indicating one of four reward distribution scenarios: (1) high self-reward, (2) high other-reward, (3) high mutual-reward, and (4) low mutual-reward. Following the visual cue, a target prompted the monkey to choose whether to proceed with the trial, with the reward delivery contingent on the simple saccadic choice. Response latency and rate served as the behavioural indices of inequity sensitivity. To dissociate social comparison from simple reward valuation, we included a non-social control condition in which a monkey performed the identical task in the absence of the partner. In these control trials, responses were primarily driven by absolute reward size: conditions with high reward outcomes for the subject (high self-reward, high mutual-reward) elicited similar performance, as did the conditions with low reward outcomes for the subject (high other-reward, low mutual-reward). In contrast, performance in the social trials reflected sensitivity to relative outcomes between the two monkeys, with slower responses and low response rates on disadvantageous inequity trials. These findings suggest that monkeys incorporate social reference frames into reward-based actions, showing behavioural signatures of inequity that emerge specifically with respect to a conspecific. Building on these results, the current study moves beyond coarse behavioural metrics to examine facial expressions and subtle movements as a window into primates' internal states. Using fine-grained facial coding and decoding methods, we assessed whether and how perceived inequity is reflected in facial movement dynamics, offering deeper insight into the affective and cognitive processes underlying social evaluation. Preliminary facial movement analysis indicates that certain features, particularly the ears and eyes, play a key role in distinguishing between different inequity conditions during the anticipation period after the visual cues but before reward deliveries. Together, these findings open new avenues for exploring detailed facial movement dynamics in relation to the affective dimensions of social evaluation in nonhuman primates.

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Topic: I.06. Social Cognition

Support: Kavli Neuroscience Discovery Institute Fellowship

Title: Vector space coding of social networks in primate amygdala and entorhinal cortex

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Abstract: Social networks are fundamental to primate life, shaping survival, alliance formation, and competition. To behave adaptively, primates must track who affiliates, threatens, or submits to whom over time. The medial temporal lobe (MTL)—including the amygdala and entorhinal cortex—is known to support memory and social cognition, but it remains unclear whether neurons in the primate MTL encode structured information about others' social roles and relationships. We investigated this by recording from the amygdala and entorhinal cortex of two rhesus macaques during passive viewing of photographs of conspecifics from their home, neighboring, and unfamiliar groups. Interpersonal relationships were quantified through three months of video using ethogram-based behavioral coding, capturing directed frequencies of affiliative, agonistic, and submissive behaviors within each group. We found that many neurons in the amygdala and entorhinal cortex encode social knowledge about interpersonal relationships, including both those involving the subject monkey (egocentric) and those between other monkeys (allocentric). Overwhelmingly, this took the form of rate coding of behavioral frequencies of the group monkeys relative to a single individual. This form of coding was spread across monkeys and behavior. Together, these neurons represent the group's social network as a high-dimensional vector space. Each monkey occupies a point in this space defined by its relationships to all the other monkeys.

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Title: Modes of social cooperation in the sequential spatial memory task in rats

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Abstract: Animals often coexist in shared environments where individuals have a similar ability to freely alter the state of the surrounding world. In such contexts, social strategies directly affect success in acquiring resources, as a lack of cooperation may impede each other's actions. Common social strategies include cooperation, competition driven by conflicting interests, direct conflict, and spatial or temporal segregation, while unintentional lack of coordination can also emerge and hinder efficiency. In the present study, to test social strategies in rat pairs, we developed an automated, interactive experimental environment in which rats perform a sequential spatial memory task. This open-field environment (64 × 64 cm) features a single reward-dispensing area and nine equally spaced floor buttons (arranged in a 3 × 3 grid), which rats can press using a fraction of their body weight. The system operates using an Arduino controller and a PC to manage floor switches, the reward dispenser, an amplifier with a speaker for playback of feedback sounds, and an LED array that signals reward delivery. Rats (n = 16) were individually trained to memorize a specific sequence of three button presses and became experts in the task. Expert rats were then introduced into the environment in pairs: eight pairs of cagemate experts for nine days, and all 112 possible unique intercage expert pairs for three days each. Additionally, we tested interactions in 28 pairs of experts with naïve cagemates and 28 pairs of experts with naïve rats from different cages. Behavioral system data were combined with position tracking from video recordings processed using DeepLabCut, allowing us to determine each rat's behavior. Our data suggest that, despite individual task proficiency, cooperation in a social context is a distinct skill that rats must develop and refine to succeed in an environment with a single reward-dispensing area. We observed different modes of operation, including cooperation, temporal segregation, and conflicts, which were resolved in various ways. We evaluated the effects of conflict on dynamic changes in social strategies. Additionally, we examined whether rats, over time, generalized this social skill to perform the task effectively with different partners, and whether extended social experience influenced their individual performance in the same task. Such testing may be useful for identifying individuals with reduced ability to operate in a social context during sequential instrumental tasks, and for studying social hierarchies emerging in an interactive, dynamic environment.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR301.11/OO5

Topic: I.06. Social Cognition

Support: NIH 1R34DA059718-01
NIH 7 RF1 DA056377-02
DA043443

Title: Understanding social cooperation in rats with multi-agent reinforcement learning

Authors: *A. M. JOHNSON¹, S. W. LI¹, H. KIETZMAN¹, D. BACKER PERAL¹, R. A. CAUCHON¹, K. PELIAS¹, J. TAYLOR², S. SAXENA¹;

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Abstract: Cooperation is an important, yet complex skill for social animals. To cooperate, animals must coordinate their actions to receive a reward which requires them to have a common goal and adapt their behavioral policies to each other. Experimental work has shown that social animals, such as rats and marmosets, learn to cooperate in tasks where both animals must press a lever within a set amount of time to receive a reward. In these tasks, visual occlusion and familiarity between the animals cause a significant decrease in cooperative ability. These experimental controls give considerable insight into the factors that are important for successful cooperation; however, it is not yet well understood how animals integrate various streams of social and non-social information to cooperate. To better understand how animals achieve cooperative success, we created a model of cooperative social behavior in animals using multi-agent deep reinforcement learning. The model was trained in a goal-driven manner; its aim was to maximize reward in a virtual environment that mimicked the experimental conditions of the cooperative lever pressing task. This model was able to consistently achieve high reward in the cooperation task and replicate key behavioral metrics observed in the rats' performance of the task such as a decrease in latency to cooperate, in re-pressing behavior, and in the latency between the first press and the cooperative press throughout the course of training. The constraint of partial observability was then added to the virtual environment: instead of receiving information about the position of the other agent at every timestep, a given agent receives this information only if they choose to "gaze" at the other agent. Agents were able to successfully cooperate even under this condition, implying the presence of an internal social model that predicts the other agent's actions. The gazing behavior of the agents provides a hypothesis for how animals collect the visual information necessary for successful cooperation and allows comparisons between the model's performance and animal behavior during visual occlusion. Training a neural network in a goal-driven way enables comparisons between the network activity of the model and the recorded neural activity of the animals during successful cooperation. These comparisons can give insight into the computations being performed in medial prefrontal cortex circuits that are necessary for cooperation.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

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Program #/Poster #: PSTR301.12/OO6

Topic: I.06. Social Cognition

Support: DA043443

Title: Investigating social cooperative learning in rodents exposed to early life adversity

Authors: ***R. A. CAUCHON**, H. W. KIETZMAN, N. NOUAILI, G. S. THIBODEAU, B. CHO, R. BONOMI, S. W. LI, J. R. TAYLOR;
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Abstract: Early-life adversity (ELA) is a significant risk factor implicated in a broad spectrum of neuropsychiatric disorders. Despite this, many individuals exposed to such adversity display notable resilience, particularly within social domains. Rodent models, exhibiting robust and diverse sociobehavioral repertoires, similarly reveal considerable variability in behavioral responses following exposure to ELA. It is known from both rodent and human literature that the timing, duration, and frequency of developmental insults critically influence adult behavior. Utilizing a novel semi-naturalistic social decision-making paradigm to measure cooperative behavior in rats, we assessed how ELA affects cooperative learning. Here, we implemented a limited nesting and bedding (LBN) paradigm from postnatal day (PN) 2-9, a chronic social isolation (CSI) paradigm from P28-42, neither, or both, in male and female Long Evans rats (n=62). In adulthood, we tested Pavlovian and instrumental learning prior to testing social cooperative learning. There was no impact of stressor on Pavlovian or instrumental learning ($F > 1$). ELA significantly impaired cooperative learning, as almost 80% of animals in the control condition learned to cooperate, compared to 60% of animals exposed to LBN or CSI and 50% of animals exposed to both stressors ($p = 0.0173$). We attempted to rescue cooperative behavior with either a behavioral intervention - a “tutor session” (n=16) with a good cooperator- or a pharmacological one -3,4,-methylenedioxy-methamphetamine (MDMA, 5mg/kg, i.p., n=22). MDMA was co-administered to cagemates either 24hr or 30min prior to cooperative testing. However, ELA-induced cooperative deficits were persistent, as non-cooperators did not improve performance following either intervention. Future studies will focus on delineating the specific neural circuitry underlying ELA-induced cooperative deficits and identifying biological and environmental factors that contribute to resilience in individuals exposed to developmental stress.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

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Program #/Poster #: PSTR301.13/OO7

Topic: I.06. Social Cognition

Support: DA043443

Title: Social Cooperation in Rodents Drives Corticolimbic Plasticity

Authors: A. BAGDAS¹, *S. W. LI², H. W. KIETZMAN², R. A. CAUCHON², N. NOUAILI², R. BONOMI², H. SANCHEZ², J. R. TAYLOR²;

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Abstract: Cooperation, a key social behavior requiring continuous appraisal of self and others, presents a promising framework to investigate prosocial-specific neural mechanisms underlying socio-affective disorders. Here, we developed a novel cooperation task in rodents to study how social coordination modulates neural circuits and whether repeated cooperation enhances plasticity. Male Long Evans rats (n=16) were trained to press levers for food individually before being paired in an open-field apparatus requiring cooperation. PET imaging was performed pre- and post-intervention, 90 minutes of repeated cooperation or non-cooperative reward seeking. Synaptic vesicle glycoprotein 2A (SV2A) binding potential, a marker of synaptic density, was assessed in the anterior cingulate cortex (ACC), anterior insula (AI), orbitofrontal cortex, striatum, and amygdala. We then used fiber photometry (n=11) to record real-time activity of ACC neurons projecting to the AI and basolateral amygdala (BLA), as well as projection-specific chemogenetic strategies to assess causation (n=16). Rats took an average of 4-5 days to learn to cooperate. Cooperative success was higher with familiar partners and visual access. Photometry revealed that ACC→BLA projections were activated during cooperative pressing, while ACC→AI projections were inhibited. DREADD-inhibition of the ACC→BLA projection vs. ACC→AI projection differentially affected cooperative success. PET imaging revealed significant SV2A increases in the insula and amygdala post-cooperation. Thus, repeated cooperation enhances social brain plasticity and engages distinct ACC circuits, highlighting its potential as a therapeutic intervention for socio-affective disorders. Future studies will explore neuromodulatory or neurofeedback interventions targeting these circuits to enhance prosocial functioning in clinical populations.

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Poster

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Alfred P. Sloan Foundation
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Title: Asymmetric social representations in the prefrontal cortex for cooperative behavior

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Abstract: Cooperative behavior, often structured by roles such as leadership and followership, is fundamental to social species. Despite its ubiquity, the cellular and circuit-level mechanisms underlying cooperation remain poorly understood. Here we introduce a novel cooperative foraging paradigm in mice, in which stable leader and follower roles emerge and reliably predict learning performance. Disruption of medial prefrontal cortex (mPFC) activity, particularly in followers, impairs cooperation and induces reciprocal shifts in the weighting of self- and partner-related cues during decision-making. One-photon calcium imaging reveals that the mPFC encodes both trial-by-trial leadership dynamics and a dynamic “social value map” of the partner's position, in a role-specific manner. These findings uncover neural representations of social roles and provide a framework for investigating the circuit basis of cooperative behavior in mammals.

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Poster

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Program #/Poster #: PSTR301.15/OO9

Topic: I.06. Social Cognition

Title: Multi-region neuronal recordings in dorsal cortex associated with differential social hierarchy competitions and naturalistic social interaction

Authors: *Z. WANG¹, Y. ZHOU¹, A. LI³, M. WANG⁴, W. ZHAO⁵, M. TIAN⁶, L. YUAN⁷, K. LI⁸, Y. ZHANG²;

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Abstract: Social hierarchy is the basic organization of animal society. The establishment and maintenance of individual ranks in such hierarchies depends on complex social calculations and adaptive behavioral strategies in the face of competition. While increasing insights have been gained into the contributions of subcortical circuits, the specific role of the dorsal cortex in coordinating dynamic behaviors and internal states associated with social competition remains inadequately characterized. We aims to investigate the neural underpinnings of social hierarchy. Our Airscope system enables cortical dynamics investigation during demanding social

interactions while minimizing behavioral disruption. In initial studies using Airscope with tube tests, we established stable hierarchies in Rasgrf2-2A-dCre;Ai148D mice. Dominant animals exhibited significantly elevated neuronal activity in specific dorsal cortical regions during encounter episodes compared to subordinate counterparts. Global analysis employing CEBRA, a non-linear dimensionality reduction technique revealed distinct low-dimensional embedding trajectories for winner and loser groups. Classifier models trained on first PCs achieved high accuracy in predicting tube test outcomes. To validate ecological generalizability, we developed a naturalistic behavioral paradigm observing differential spontaneous competition in a large home-cage arena. Social hierarchy retention was confirmed with neural patterns in spontaneous tube competition mirroring initial findings. Granger causality analyses further demonstrated differential cortical activation patterns across multiple brain regions during differential competition. Extending to autism spectrum disorder (ASD) research, we conducted comparative analysis of cortical dynamics in neuroligin-3 (Nlg3) transgenic ASD models versus wild-type controls during naturalistic social interactions. Airscope recordings demonstrated widespread functional dissociations: wild-type mice exhibited coordinated neuronal oscillations during social exploration, while Nlg3 mutants displayed disrupted synchrony and aberrant activation patterns across multiple regions. Machine learning classifiers achieved superior ASD discrimination accuracy and the anterior cingulate cortex (ACC) was identified as the most influential cortical region. In summary, we have, for the first time, provided multi-region neuronal recordings associated with differential social hierarchy competitions and naturalistic social interaction within ASD model and wild-type mice .

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Poster

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Program #/Poster #: PSTR301.16/OO10

Topic: I.06. Social Cognition

Title: Studying Rat Societies

Authors: *Y. PAN¹, D. G. TERVO²;

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Abstract: Animals have evolved to live and make decisions within complex societies, yet the exact nature of these societal social behaviors and the neural circuit mechanisms underlying them remains to be identified and understood. To investigate decision-making in societal contexts we constructed a “Rat City” -- an indoor naturalistic environment designed to simulate a societal setting for rats. We applied infrared red lighting and filter for camera to record rats’ activities 24/7 continuously. In the meanwhile, we used ultrasound microphone to record rats’ vocalization. We also developed software for data acquisition, archiving and organization. These

developments allow us to monitor rats' behavior continuously for weeks and track social interaction dynamics over the period. To test if the group identity of rats within Rat City would influence their social behaviors, we conducted an experiment where we had two groups of rats that differed according to their kin relationships, familiarity, and home territory. After isolating them on opposite sides of Rat City for seven days, we opened a door that connected these two sides, allowing the two groups to interact freely. We initially focused on aggression behavior by annotating who fought whom and who won the conflict. We found that rats would fight with each other, where the aggressive period peaked when the two groups were united. We found that the target of aggression were dependent on the pairwise identity of rats, group affiliation and pairwise social interaction history. We are repeating experiments on different cohort of animals to confirm behavioral observations, and are establishing electrophysiological recordings in Rat City to examine neural correlates of these societal social behaviors.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

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Program #/Poster #: PSTR301.17/OO11

Topic: G.01. Neuroethology

Support: CUNY, Interdisciplinary Research Grant

Title: Effects of Urban-Stressors on Behavior using a Naturalistic Mouse City

Authors: *G. K. HOTH¹, M. B. BELLAMY², R. TOLEDO-CROW², P. GROFFMAN², L. ABHILASH³, J. LIU¹, P. AYATA¹, J. P. CURLEY⁴, D. P. MCCLOSKEY⁵, S. MINGOTE¹; ¹Neurosci., ²Envrn. Sci. Initiative, Advanced Sci. Res. Ctr. CUNY, New York, NY; ³Gill Inst. for Neurosci. and Dept. of Biol., Indiana Univ., Bloomington, IN; ⁴Psychology, Univ. of Texas At Austin, Austin, TX; ⁵Dept of Psychology and Program in Developmental Neurosci., Col. of Staten Island, City Univ. of New York, Staten Island, NY

Abstract: Urban living and the constant exposure to numerous environmental stimuli have been increasingly linked to negative impacts on mental health, including higher rates of depression and psychosis. However, adequate environmental stimulation is essential for optimal social and cognitive development. This underscores a paradox: while some level of environmental stimulation is beneficial, excessive stimulation—such as the noise and light pollution characteristic of urban environments—can be harmful to mental health. Causal links between urban stressors and brain and behavioral dysfunction are hard to study in humans. Mice, like humans, are naturally social animals, and their brain function and behavior are highly sensitive to social and environmental stressors. This makes them an excellent model organism for studying the effects of urban setting. Exposure to light or air pollution are often conducted with mice housed in small, impoverished cages. To address this limitation, it is crucial to evaluate the

effects of urban-like stressors on mice housed in enriched, naturalistic environments. We built a mouse city, a large colony of twenty female mice (C57BL/6J) housed in five interconnected cages. We compared two setups: 'mouse city' under enriched conditions but no stressors (named rural environment), and 'mouse city' exposed to urban-like stressors (named urban environment). The stressors in the urban setting include light pollution, sound pollution, soil pollution, reduced cage space, and unpredictability of available food. Using Radio Frequency Identification (RFID) tagging system, animal behavior was tracked for three months, exploring changes in circadian rhythms, cage preference, social interactions, and reward-based learning. The artificial light and sound exposure in urban environment altered the circadian rhythms compared to the mice in rural environment. Exposure to the additional stressors in the urban-like housing also resulted in an elevated stress response exhibited by frequent changes in cage preferences. Interestingly, urban mice acquired the reward competition task more quickly than rural mice. Thus, these findings suggest that resource unpredictability drives motivation in the urban mice to compete more and boost task performance. Overall, the results indicate how environmental uncertainty drives behavioral shifts in mice and provide insights into how such urban stressors may impact social costs, resilience, and productivity in humans, providing valuable knowledge for optimizing urban planning. Future studies will investigate changes in brain functions between rural and urban settings.

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Poster

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Program #/Poster #: PSTR301.18/OO12

Topic: I.06. Social Cognition

Title: Ultrasonic Vocalization in Rats: The 25 kHz Call

Authors: *S. NEALE;
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Abstract: The phenomenon of ultrasonic vocal (USV) communication in rats has been well-documented, with observations generalized into two primary categories of signals: the 22 kHz and the 50 kHz call. In accordance with the assumption that signaling in animals is affective in origin, these signal groups have often been associated with affective states, aptly as general as the signal groups themselves. The 22 kHz call correlates with negative affect and is thought to serve as an alarm call, while 50 kHz signal correlates with positive affect and often occurs during play, positive social interactions, or reward consumption. It's important to note that these vocalizations are context-dependent and vary in their patterning, yet, despite this nuance, these generalizations have held. We have observed a previously undocumented 25 kHz call

synchronized with chattering, an acoustic, non-vocal behavior seen in rats under conditions of negative affect and high arousal, such as aggression or anxiety. This signal has been seen in pairs of rats undergoing an experiment in which one rat is confined in a transparent box that can be opened by their conspecific via lever-press. Rats that learn an association between lever pressing and liberation from confinement expressed the 25 kHz as a “call to action” to encourage helping behavior in the free conspecific. The 25 kHz differs from the 22 kHz signal in that appears to serve a distinct function, asking for help in situations where the vocalizer is in distress.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

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Topic: I.06. Social Cognition

Support: Peter and Carmen Lucia Buck Foundation
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Title: Individual Differences in Observational Learning in Rats

Authors: *E. J. MARKUS¹, E. BERI⁶, T. WRENN², M. JINDAL³, M. GARBIN⁴, Z. FAISAL⁵;
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Abstract: Learning through observation of a conspecific can play an important role for survival. The basic idea is that watching is better than doing. Observation of a demonstrator reduces the time, energy and exposure needed to learn through trial and error. The most obvious example is learning to avoid predators and dangerous situations indirectly rather than learning by actually experiencing the aversive event. However observational learning is also beneficial in other circumstances. The current study focuses on the role of observational learning in appetitive conditioning scenarios in which one animal learns the location of a food source from another. We use two conjoined chambers with a teacher rat being observed by a student rat for up to 80 independent observation training trials within a session. In the past the large number of trials was used to determine, in well trained rats, factors related to successful vs unsuccessful observing trials within a given session (Troha et al., 2023). The current study focuses on the process of “learning to observe” and variability in the degree of learning across students and with different types of teachers. There were differences in the rate of learning and asymptotic performance of student rats under social or non-social observation conditions. Initially, student rats avoided the location demonstrated by the teacher rat, and learning was faster in the non-social teacher condition. With further training, higher levels of performance were achieved in the teacher rat

condition. There were also differences in the behavior of student rats BEFORE the initiation of the task dependent on the social/non-social situation. When examining relative performance across observation sessions, performance of individual animals was stable with a given teacher. In addition, a student's relative performance correlated across rat teachers, and even generalized from the rat teacher condition to the non-social cue condition.

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Poster

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Program #/Poster #: PSTR301.20/OO14

Topic: I.06. Social Cognition

Title: Characterization of context-dependent social behavior with different motivational drives

Authors: D. KWAK¹, A. E. HARTEY², *B. C. CURD³, N. A. FROST⁴;

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Abstract: Social interactions consist of a diverse repertoire of behaviors, which are flexibly regulated over the lifetime of an organism. The manner in which an organism engages in social behaviors thus may manifest in a context-dependent manner which is influenced by competing motivational drives in different situations or environments. Thus, an organism may behave differently depending on its familiarity or comfort within a given location. In this study, we sought to characterize how social interactions change when mice interact within distinct contexts with varying degrees of novelty or familiarity. We hypothesized that competing motivational drives between environment exploration and social investigation induce social behavioral states across contexts. To test this, we imaged mice during social interactions occurring in enclosures of different sizes and familiarity. We then performed UMAP clustering of pose estimation inferred using a Social LEAP Estimates Animal Poses model trained in our laboratory which identifies multi-animal pose information within behavioral videos to identify individual mice during social interaction, and track their movements over time. We extracted 19 features from these inferred multi-animal pose estimations and performed UMAP clustering; this generated groups of frames clustered by inter- and intra-mice relationships. Comparison to manually labeled frames revealed clusters enriched for specific active and passive social behaviors which differed in their abundance, in a context-specific manner. These data demonstrate an unsupervised approach to analyzing social interactions and reveal context-dependent changes in the mouse social ethogram.

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Poster

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Topic: I.06. Social Cognition

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CUHK 7104252

Title: Social competition-driven modulation of social behavior in mice

Authors: ***R. GOH**^{1,2}, M. GAO^{1,2}, J. HUANG¹, W. YUNG², Y. KE¹;

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Abstract: Living in social groups inevitably leads to repeated interactions with various individuals, requiring the ability to recognize others as unique identities, integrate information accumulated from prior social encounters, and flexibly tune our behavior in response to the nature of past social experiences. While substantial research has focused on interactions between male-female or familiar-stranger pairs, less is known about the neural mechanisms underlying how competitive experiences shape social interactions. Here, we investigated the influence of competitive experiences on subsequent social behavior using an intergroup social competition model in mice. As an initial step to tackle this question, we employed a reward competition assay in which age-matched mice were trained to physically compete for water reward delivered via a single port during the presentation of a visual cue. The task consisted of two phases: (1) individual training of mice to associate a visual cue with reward delivery, and (2) subsequent cross-cage pairings of mice to compete for the same reward. Over two weeks of repeated competitions, mice exhibited progressively intensified competition-related behaviors, evidenced by a more than twofold increase in jostling time and a two-thirds reduction in the winning margin. To assess the effects of intergroup competition on social behavior, we then subjected mice to dyadic interactions involving either their competitors or familiar conspecifics with no history of competition. We observed that mice showed more persistent investigatory behaviors and heightened aggression towards their competitors compared to neutral noncompetitors. Further investigation using a conditioned place preference-avoidance assay demonstrated that interactions with noncompetitors were emotionally neutral, whereas competitive experiences induced place avoidance and negative emotional valence. These findings highlight the significant impact of competitive social experiences on the nature of subsequent social interactions.

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Poster

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Topic: I.06. Social Cognition

Support: JSPS KAKENHI 24K16868
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Title: Neural mechanisms of empathic concern toward conspecifics in nausea through olfactory signals in rats

Authors: *M. TOYOSHIMA¹, R. TACHIHARA¹, A. CHIBA¹, T. XIONG¹, P. DURVE¹, M. HORI¹, K. YAMADA²;

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Abstract: Empathy is defined as the ability to understand and share the feelings of others. It involves cognitive processes influenced by the affective states of others, as well as own responses to those emotional states. Empathy-like behaviors—such as emotional contagion, empathic concern, and consolatory responses—have been observed across various mammalian species, including rodents. Our research previously established that rats exhibit empathy-like behaviors toward sex-matched, unfamiliar conspecifics injected with the emetogenic agent lithium chloride (LiCl). In the three-chambered social affective preference paradigm, rats preferentially approached LiCl-treated conspecifics over those treated with saline, indicating that they can recognize and respond to the discomforted state of other rats. These behaviors appear to be mediated by olfactory signals, as rats' preference was consistently inclined toward the body odor of the distressed conspecifics. Furthermore, olfactory ablation induced by intraperitoneal injection of methimazole partially disrupted the preference behavior toward LiCl-treated conspecifics and their odors. Moreover, focal rats showed no preference during an encounter with unconscious conspecifics that were intraperitoneally injected with secobarbital. Additionally, male rats failed to display any preference towards LiCl-treated female rats when engaged in social behaviors involving distressed conspecifics of the opposite sex. We are currently investigating the role of the anterior cingulate cortex and its neural projections in mediating preference for LiCl-treated conspecifics and their odors.

Disclosures: M. Toyoshima: None. R. Tachihara: None. A. Chiba: None. T. Xiong: None. P. Durve: None. M. Hori: None. K. Yamada: None.

Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR301.23/OO17

Topic: I.06. Social Cognition

Support: JSPS KAKENHI 23K03023
JSPS KAKENHI 20K03488

Title: Effects of post-weaning stress and social environment on empathy-like behavior in adult rats.

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Abstract: This study investigated how different post-stress environments affect empathy-like behavior in adult rats that experienced early-life stress. Male F344/N rats were exposed to acute foot-shock from postnatal day 22 to 24 and then assigned to one of three conditions: group housing (G), social isolation (SI), or social isolation with positive emotional stimulation via tickling (T). In adulthood, rats were tested using the Social Interaction Test (SIT) and Emotional Discrimination Test (EDT) to assess social engagement and sensitivity to others' emotional states. Empathy-like responses toward unfamiliar conspecifics injected with lithium chloride (LiCl), an emetogenic agent, were assessed in both tests. T-group rats consistently showed short approach latencies and frequent 50-kHz ultrasonic vocalizations during tickling, suggesting a positive emotional state. As a result, G-group rats displayed stronger anxiety-related behavior in both tests, indicating more pronounced effects of early-life foot-shock stress. However, they also spent more time near distressed conspecifics in the EDT, suggesting that emotional sensitivity and contagion remained intact. Compared to the G group, both the SI and T groups showed more frequent contact and sniffing behaviors, indicating increased social motivation possibly resulting from isolation. However, SI rats kept greater distances from distressed conspecifics in the EDT, suggesting reduced social interest and empathy. In contrast, T rats moved closer to distressed conspecifics than to saline-injected control ones, implying that tickling may mitigate the negative effects of isolation and enhance emotional recognition. These results suggest that while early-life stress impairs empathy-like behavior, positive emotional experiences such as tickling can support the recovery or enhancement of social and emotional responsiveness, even under socially deprived conditions. The findings highlight the significance of environmental conditions following early adversity and support the use of social or emotional interventions to reduce the long-term impact of childhood stress on empathy-related behavior.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR301.24/OO18

Topic: I.06. Social Cognition

Support: JSPS KAKENHI 24K16868
JSPS KAKENHI 25KJ0661

Title: Prefrontal dysfunction impairs social affective behaviors in a rat model of schizophrenia

Authors: *R. TACHIHARA, M. TOYOSHIMA, T. XIONG, K. IGARASHI, M. HORI, K. YAMADA;

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Abstract: Schizophrenia is a severe psychiatric disorder characterized by a variety of symptoms, including social deficits. However, effective treatments for these impairments remain elusive due to an incomplete understanding of its pathology. Here, we investigated the neurobiological mechanisms underlying social affective deficits in schizophrenia using a rat model based on the glutamate hypothesis. To establish the model, Sprague-Dawley rat pups were subcutaneously injected with MK-801 (Control: saline only, Low: 0.2 mg/kg, High: 0.4 mg/kg), an NMDA receptor antagonist, twice daily during the neonatal period. In adulthood, their sociability was assessed through two behavioral tests. One was the Social Affective Preference (SAP) test, which measured approach behavior toward a distressed conspecific injected with lithium chloride. The other was the Social Interaction (SI) test, which observed behavioral responses during one-on-one interactions with a distressed conspecific. The MK-801 treated rats exhibited deficits in both the SAP and SI tests, with severity depending on the dosage of MK-801. Given the reported causal relationship between social deficits and cortical hyperexcitability, particularly in the medial prefrontal cortex (mPFC), we employed DREADD system to examine whether artificial inhibition could alleviate social deficits in the model rats. DREADD-mediated inhibition of the mPFC restored SAP performance in the MK-High group. Consistent results were observed following treatment with diazepam, an anxiolytic drug that enhances inhibitory signaling in the central nervous system. Conversely, artificial activation of the mPFC in control rats replicated the socio-affective deficits observed in the MK-High group. This study demonstrates that MK-801-induced model rats of schizophrenia show deficits in social affective behaviors, which can be linked to an excitatory/inhibitory imbalance in the mPFC. We are currently further investigating this relationship from behavioral, neuroscientific, and genetic perspectives.

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Poster

PSTR301: Behavioral and Neural Mechanisms of Social Interaction

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR301.25/PP1

Topic: I.06. Social Cognition

Support: NIH grant GM144276
University of Illinois College of ACES FIRE grant

Title: Synaptic proteome divergence in the prefrontal cortex of behaviorally tame and aggressive red foxes (*Vulpes vulpes*)

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Abstract: Social behavior is influenced by numerous factors, making it challenging to isolate the underlying biological mechanisms. The Farm-Fox Experiment at the Institute of Cytology and Genetics in Novosibirsk, Russia breeds three populations of foxes (*Vulpes vulpes*) that display genetically distinct behaviors (i.e., tame, aggressive, and conventional). Out of the potential biological systems that underly behavior, synaptic mechanisms stand out as they control neuronal communication. To determine the synaptic differences between the three fox populations, the prefrontal cortical extracts from sixteen, female foxes were collected. The synaptosomes were then isolated, digested, and tagged with individual tandem mass tags. Liquid chromatography tandem mass spectrometry (LC-MS3) was used to identify and quantify the relative abundance of the proteins. The results were broken down into protein groups and compared between the three populations using limma to determine differential expression (DE). ADGRB2 was found to be the most DE protein group, with 11-fold greater fold-change in aggressive foxes relative to the tame foxes. Notably, ADGRB2 is known to affect depression-like behavior in mice and is involved in the vascular endothelial growth factor signaling pathway, that is known to influence neurogenesis. Enrichment analyses of the DE protein groups found gene ontology terms and KEGG pathways that were enriched. Terms involved in ribosomal translation were enriched in all analyses of tame versus aggressive populations. Synaptic translation is important for synaptic plasticity and could therefore impact behavior. This study highlighted roles for neurogenesis and synaptic plasticity as mechanisms underlay the behavioral differences between tame and aggressive foxes.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

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Topic: I.08. Learning and Memory

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Johns Hopkins University Discovery Award
Johns Hopkins Kavli Neuroscience Discovery Institute Postdoctoral Distinguished Fellowship
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Title: Recalibration of hippocampal path integration gain without angular head velocity gain recalibration of head direction cells

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Abstract: Path integration (PI) is a spatial navigation process in which animals track their location by integrating self-motion cues. The PI gain of CA1 place cells is a key parameter that converts distance traveled in the world to corresponding distance traveled in the cognitive map. This gain recalibrates to new values when PI is placed in conflict with visual landmark navigation (Jayakumar, Madhav, et al., 2019). However, the locus of PI gain plasticity is unknown. A key input to the place cells is the head direction (HD) cell system, which sets the orientation of the cognitive map. To test whether HD cells also exhibit PI gain recalibration, we simultaneously recorded from place cells and HD cells in the thalamus and the cingulum bundle while 5 Long-Evans rats ran around a circular track inside a planetarium-style Dome VR apparatus. An array of visual landmarks was moved in the same or opposite direction as the rat's movement, causing an illusion that the rat was moving slower (or faster) than it actually was. The motion of the landmarks was controlled by an experimental gain, G , which specified the animal's speed in the landmark frame relative to the lab frame. In 42/53 sessions, the tuning of both place cells and HD cells remained anchored to the moving landmarks, establishing a sustained period where the rate of internal position update was entrained to G but in conflict with vestibular, proprioceptive, and motor-copy cues. Once the landmarks were turned off, the rate of update of place-cell activity, now solely driven by self-motion cues, was strongly correlated with the preceding changes in G , indicating that the PI gain had recalibrated based on the altered- G experience. Simultaneously recorded HD cells exhibited nearly identical gains as the hippocampal gain (Sueoka, Jayakumar, et al., SfN, 2024). The synchronized recalibration could result from plasticity occurring either i) in the angular head velocity (AHV) inputs onto the HD system, which entrains the hippocampal map (feedforward model), or ii) downstream of the HD system in the place or MEC grid-cell system, which then entrains the HD activity (feedback model). To dissociate these hypotheses, we analyzed head scanning behaviors, characterized by lateral head movements with minimal changes in body position. Given the absence of locomotion during the scans, HD cell activity was driven primarily by the AHV inputs. Strikingly, the gain decoded from HD cell activity during head scans in the absence of landmarks was close to 1, with no correlation to G ($r = -0.106$; $p = 0.441$). The lack of recalibration during

head scans supports the feedback model of PI gain recalibration under these experimental conditions.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR302.02/PP3

Topic: I.08. Learning and Memory

Support: NIH Grant R01 NS039456
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Title: Reward-evoked head scanning increases the likelihood of rapid hippocampal place field potentiation compared to spontaneous head scanning

Authors: *P. OZEL^{1,2,3}, G. RAO², V. MULLER², V. PULIYADI⁴, Y.-Q. ZHOU^{2,5}, J. J. KNIERIM^{2,3,4,5};

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Abstract: As animals navigate their changing environments, they rapidly integrate behaviorally relevant events onto an internal cognitive map in the hippocampus, supporting the “automatic recording of attended experience” necessary for episodic memory (Morris and Frey, 1997). Previous work demonstrated that an exploratory behavior known as lateral “head scanning” predicts the sudden appearance or strengthening of hippocampal place fields (Monaco et al., 2014). Building on this result, we asked whether modulating the saliency of information gathered during head scanning increases the likelihood that hippocampal place cells undergo rapid place field potentiation. Using Neuropixels 2.0 probes, we recorded hippocampal CA1 and CA3 neurons in Long-Evans rats running 25 laps clockwise on a circular track. We used intracranial stimulation of the medial forebrain bundle to deliver precisely timed reward events, which served to provide controlled, salient experiences and to motivate running. Across 642 place cells, 252 potentiated their firing fields during the session—either forming de-novo fields or strengthening existing ones. We classified behavioral events as reward-evoked head scans, spontaneous head scans, and reward-alone events. Monte-Carlo simulations confirmed that both scan classes predicted potentiation greater than chance. Notably, although reward-evoked scans were less frequent than spontaneous scans (1,543 vs. 4,925 events), they accounted for a twofold higher rate of field potentiation events: 1.8 % of reward-evoked scans (28/1,543) resulted in place field potentiation at the reward site on the next lap, compared to 0.9 % of spontaneous scans

(45/4,925). Although these rates were low (as expected), a bootstrap test revealed that this difference in field potentiation rate was highly significant ($p = 0.0039$). We propose that this form of behavior-induced, representational plasticity allows the hippocampus to rapidly bind salient information gathered during attentive behavior onto the cognitive map.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: I.08. Learning and Memory

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Kavli Neuroscience Discovery Institute Graduate Student Fellowship
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Title: Visual landmark-anchored directional signals differentiate rodent area 29e from neighboring parahippocampal areas

Authors: *G. SECER^{1,2,3,4}, B. KRISHNAN^{3,4,5}, J. J. KNIERIM^{3,4,6}, N. J. COWAN^{2,4,7};
²Lab. for Computat. Sensing and Robotics, ³Zanvyl Krieger Mind/Brain Inst., ⁴Kavli Neurosci. Discovery Inst., ⁵Dept. of Biomed. Engin., ⁶Solomon H. Snyder Dept. of Neurosci., ⁷Dept. of Mechanical Engin., ¹Johns Hopkins Univ., Baltimore, MD

Abstract: Spatial memory and navigation engage a network of regions in and around the hippocampus, including the medial entorhinal cortex (MEC), parasubiculum (PA), postrhinal cortex (PO), and dorsal presubiculum (PR), all of which have been extensively studied. In contrast, the spatial coding properties of Brodmann area 29e—an adjacent area with distinct anatomical and molecular features (Chen et al., 2022; Haug, 1976)—are largely unknown. To address this gap, we recorded single units from 29e along with PO, MEC, PA, and PR. Five rats ran laps on a 1.5 m diameter circular track in the presence of visual landmarks inside a virtual reality apparatus. In all 5 regions, the majority of cells displayed significant spatial tuning in similar proportions (29e 70/78; MEC 257/327; PA 175/228; PO 98/123; PR 31/43; $p = .116$). However, unlike the other regions, 29e neurons tended to increase or decrease their firing rates close to landmark locations, ($p < .001$). When landmark and background brightness were varied pseudo-randomly, 29e neurons' firing rates were more strongly modulated by landmark contrast than were firing rates of neurons in MEC and PR ($p < .001$). To test landmark control over the spatial firing, we rotated the landmark array around the track at various speeds determined by the rat's movement. A landmark locking score quantified the degree to which the firing fields remained bound to the rotating landmarks, resulting in a bimodal distribution when recorded

cells from all regions were merged. In one mode, largely consisting of low-speed rotation sessions, the spatial firing fields in all regions were tightly locked to the rotation of the landmarks, with no significant differences in landmark control ($p = .219$). In the other mode, largely consisting of high-speed rotation sessions, most 29e neurons maintained their spatial tuning tightly locked to the landmarks, whereas most neurons in other regions drifted relative to the landmarks ($p < .001$).

Finally, we assessed the spatial tuning properties of neurons as rats freely foraged in a 1.4 m x 1.4 m square arena. Cells were quantitatively classified as nondirectional place-modulated cells, allocentric head direction (HD) cells, and egocentric HD cells. The 5 regions differed in the proportions of these cells ($p < .001$), with 29e showing the fewest proportion of nondirectional, place-modulated neurons (15%) and the highest proportion of egocentric (28%) and allocentric (34%) HD cells.

Together, these findings identify 29e as a functionally distinct brain region that provides visual landmark-anchored, directional signals that are likely to play a role in binding the internally generated cognitive map to the external environment.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

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Topic: I.08. Learning and Memory

Support: NINDS Grant 5R01NS102537-07
NIH-NIMH Grant 1R01MH118926-01
Kavli NDI Distinguished Graduate Student Fellowship (B.K.)
Kavli NDI Distinguished Postdoctoral Fellowship (G.S.)

Title: Distinct physiological properties differentiate rodent area 29e from neighboring parahippocampal areas

Authors: *B. KRISHNAN^{1,2,3}, G. SECER^{4,2,3}, N. J. COWAN^{5,4,3}, J. J. KNIERIM^{2,6,3},
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Abstract: Brodmann area 29e has been considered by different anatomists as a part of retrosplenial cortex, presubiculum (PR), or parasubiculum (PA) (Brodmann, 1909; Rose & Woolsey, 1948; Stephan, 1975; Haug, 1976). Despite being recognized as a distinct anatomical subdivision, its physiological and functional properties remain largely uncharacterized in rodents. We performed simultaneous tetrode recordings in five areas - area 29e, medial entorhinal cortex (MEC), postrhinal cortex (PO), dorsal PR, and PA - as rats (n=5) freely

navigated two different environments: (i) a circular track in a planetarium-style virtual reality “dome” with visual landmarks and (ii) a square open arena. Across all recorded regions, power in theta (6-10 Hz) and mid-gamma (50-90 Hz) bands of local field potentials (LFP) were higher compared to background 1/f activity ($p < .05$). 29e and PR had lower theta band power compared to other regions ($p < .001$), but there were no regional differences in power in the mid-gamma band. Single neurons in 29e exhibited lower firing rates compared to other regions ($p < .05$). Compared to other brain areas, 29e neurons had lower theta rhythmicity in spike autocorrelograms and weaker coupling to LFP theta ($p < .001$). However, 29e neurons showed greater rhythmicity in the gamma band ($p < .05$). When the landmark array was rotated, 29e neurons with weaker spike-theta coupling were more strongly locked to landmarks than neurons with stronger coupling ($p < .001$). Similarly, 29e neurons that responded more strongly to changes in the luminance contrast of landmarks had weaker theta coupling ($p < .05$). The population of contrast-responsive cells was more strongly locked to visual landmarks when compared to 29e neurons without significant contrast responses ($p < .01$). In the open arena, among spatially modulated 29e neurons, egocentric-HD and nondirectionally modulated cells showed significantly weaker theta coupling than allocentric-HD cells ($p < .05$); correspondingly, 29e egocentric-HD and nondirectionally modulated neurons exhibited stronger landmark locking than 29e allocentric-HD neurons ($p < .05$). The relative lack of theta-band LFP and theta-rhythmic spiking, together with prominent gamma oscillations and gamma-rhythmic firing, support a visual processing role for 29e that is distinct from neighboring parahippocampal regions. Given the strong projections from area 29e to dorsal presubiculum (Chen et al., 2021; Chen et al., 2022), these findings identify 29e as a functionally distinct parahippocampal area that could serve as a route by which visual landmarks anchor an animal’s internal sense of direction to the external world.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR302.05/PP6

Topic: I.08. Learning and Memory

Title: Tau-mediated Disruption of Entorhinal-Hippocampal Connectivity Impairs CA1 Spatial Coding

Authors: L. WELZ¹, T. WONG¹, E. W. NICKERSON², E. BLOSS², *C. GRIENBERGER¹;
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Abstract: The entorhinal-hippocampal circuit is essential for spatial memory and is among the earliest sites of dysfunction in Alzheimer’s disease (AD). However, the mechanisms by which tau pathology disrupts this circuit remain poorly understood. Here, we investigated how expression of mutant humanized tau impacts information processing within the medial entorhinal

cortex layer III (MECIII)-CA1 pathway. MECIII-specific expression of humanized tau was achieved by pairing a MECIII-Cre line with Cre-dependent viral expression of constructs encoding humanized wild-type tau (hTau(Wt)) or humanized tau containing the disease-associated P301L mutation (hTau(P301L)). High-resolution light microscopy revealed that hTau(P301L) expression led to a significant loss of synapses specifically on the distal dendrites of CA1 pyramidal neurons. Electron microscopy revealed the accumulation of hTau at MECIII-CA1 synapses, and mass spectrometry showed that hTau(P301L) expression was associated with altered binding to proteins related to synaptic functions. In parallel, in vivo two-photon GCaMP6f imaging of CA1 pyramidal neurons during a treadmill-based spatial learning task showed a marked reduction in both the number and spatial selectivity of active place cells in hTau(P301L)-expressing mice. Ongoing analyses suggest that these functional impairments are linked to disrupted dendritic plateau potential generation and a consequent decrease in behavioral timescale synaptic plasticity (BTSP). Relative to bilateral MECIII hTau(Wt) expression, bilateral hTau(P301L) produced a spatial learning deficit evidenced by reduced lick selectivity for the reward location. As these changes occurred without gross neuronal loss or bona fide tangle pathology, our results implicate dysfunction in circuit-level plasticity mechanisms and overt synaptic disconnection as early drivers of tau-mediated cognitive decline. Together, these findings suggest mechanisms that promote synaptic maintenance and enhance BTSP as potential targets for intervention.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

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Topic: I.08. Learning and Memory

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Title: A right-to-left shift in CA3 input dominance underlies CA1 spatial map development and stabilization during learning

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Abstract: CA1 place fields underlie spatial maps supporting memory, yet how bilateral CA3 inputs shape these maps during learning remains unclear. Using two-photon calcium imaging and optogenetic inhibition in head-fixed mice navigating a virtual track, we examine left and right CA3 projections to right CA1 (CA1_R) as they learn about a novel environment. CA1_R spatial maps are initially inaccurate but gradually improve and stabilize after ~10 laps (early-phase), with later laps marking stability (late-phase). In the early-phase, both CA3 inputs drive CA1_R field emergence, but right CA3 inputs predominantly drive high-amplitude, reliable fields that contribute to developing maps. Later, left CA3 inputs predominantly maintain reliable fields and stable CA1_R maps. Axonal recordings reveal heightened right CA3 activity early, shifting to increased left CA3 activity later. These findings demonstrate a shift from right-to-left CA3 input dominance that mediates the progression from early map development during learning to later stabilization in CA1_R.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

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Program #/Poster #: PSTR302.07/PP8

Topic: I.08. Learning and Memory

Title: Unifying Directional Place Fields

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Abstract: The hippocampus encodes spatial information to support navigation and memory, with place cells exhibiting directional tuning in one-dimensional (1D) environments and non-directional activity in two-dimensional (2D) spaces. This dichotomy raises fundamental questions about the conditions shaping hippocampal spatial representations. We hypothesized that the directionality of place fields reflects the curriculum of sensory experience and its latent structure, and that spatial representations could be unified by systematically manipulating this input. Using computational simulations, we verify this hypothesis, and find that neural activity mirrors the latent structure of the linear track in the form of lines embedded in a high dimensional space. Under certain conditions these lines become parallel, which translates to non-directionality of place cells. To test this in vivo, we developed a novel behavioral paradigm in which mice traverse a linear track while being tethered to rotate throughout the track rather than traversing from one edge to the other, creating a continuum of sensory experiences associated with opposite directions. We find that this setup produces place fields with reduced directionality, reflecting a unified spatial representation. This approach enables causal

investigation into the mechanisms governing the emergence and plasticity of spatial codes in the hippocampus.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

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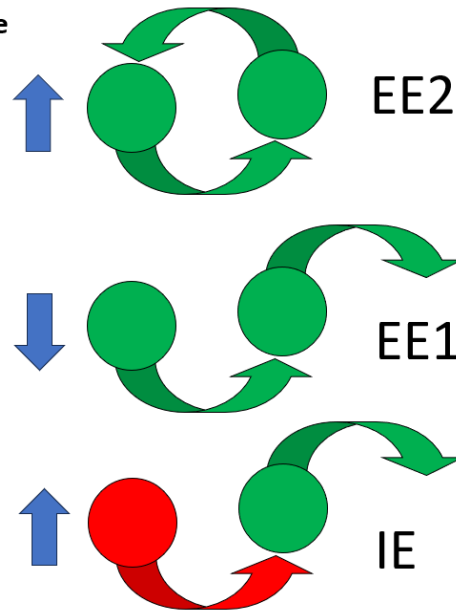
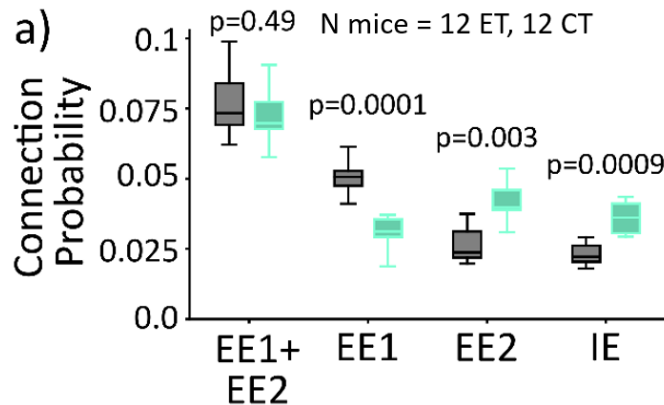
Title: Enriched experiences increase symmetrical connectivity and sparsity in higher-order association cortex, but not in primary sensory cortex

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Abstract: How does environmental enrichment (EE) alter neural connectivity and representations? Cognitive Reserve (CR), resulting from lifetime accumulation of knowledge, is the principal correlate of resilience against dementia and age-related cognitive decline. We studied CR in mice using EE as a proxy for knowledge acquisition. EE has been shown to improve memory and cognitive functions, but its effect on synaptic connectivity and population activity is understudied. Juvenile mice ran on either a control (CT) or enriched (ET) track for ten weeks. ET mice were exposed to a unique configuration of objects each day, providing a rich repertoire of experiences. After EE, mice were injected with a virus targeting the Parafacial Zone, enabling chemogenetic induction of slow-wave sleep (SWS; doi.org/10.1038/nn.3789). Single-unit recordings were performed using high-density silicon probes from the retrosplenial cortex (RSC) and primary visual cortex (V1). Recordings were performed for 2.5-3 hours during awake rest and at least 13 hours during induced SWS. These extended recordings allowed us to reliably assess functional excitatory and inhibitory synaptic connectivity. Analyzing short-latency time bins (0.8-4.8 ms) cross-correlograms revealed a substantial increase in reciprocal excitatory-excitatory (E-E2) connections in the RSC ET group, indicative of many more cell assemblies. This was accompanied by a corresponding decrease in unidirectional excitatory-excitatory (E-E1) connections and an increase in inhibitory (I-E) connections. These changes were observed in RSC, but not in V1. Furthermore, ET mice showed an increase in sparsity and orthogonality in population activity only in deep layers (layer 5/6) of RSC, but not in superficial layers (L2/3) or other brain regions (V1 or CA1). Overall, these results suggest that enriched

experiences reshape excitatory and inhibitory connectivity, reflecting formation of multiple recurrent cell assemblies that may underlie the increased representational capacity and behavioral performance conferred by accumulated knowledge.

Enrichment makes the excitatory synaptic matrix **more symmetrical** (more cell assemblies), and increases inhibitory to excitatory connections (compensates for increased feedback?)
No change in total excitatory connections (EE1 + EE2)



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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR302.09/PP10

Topic: I.08. Learning and Memory

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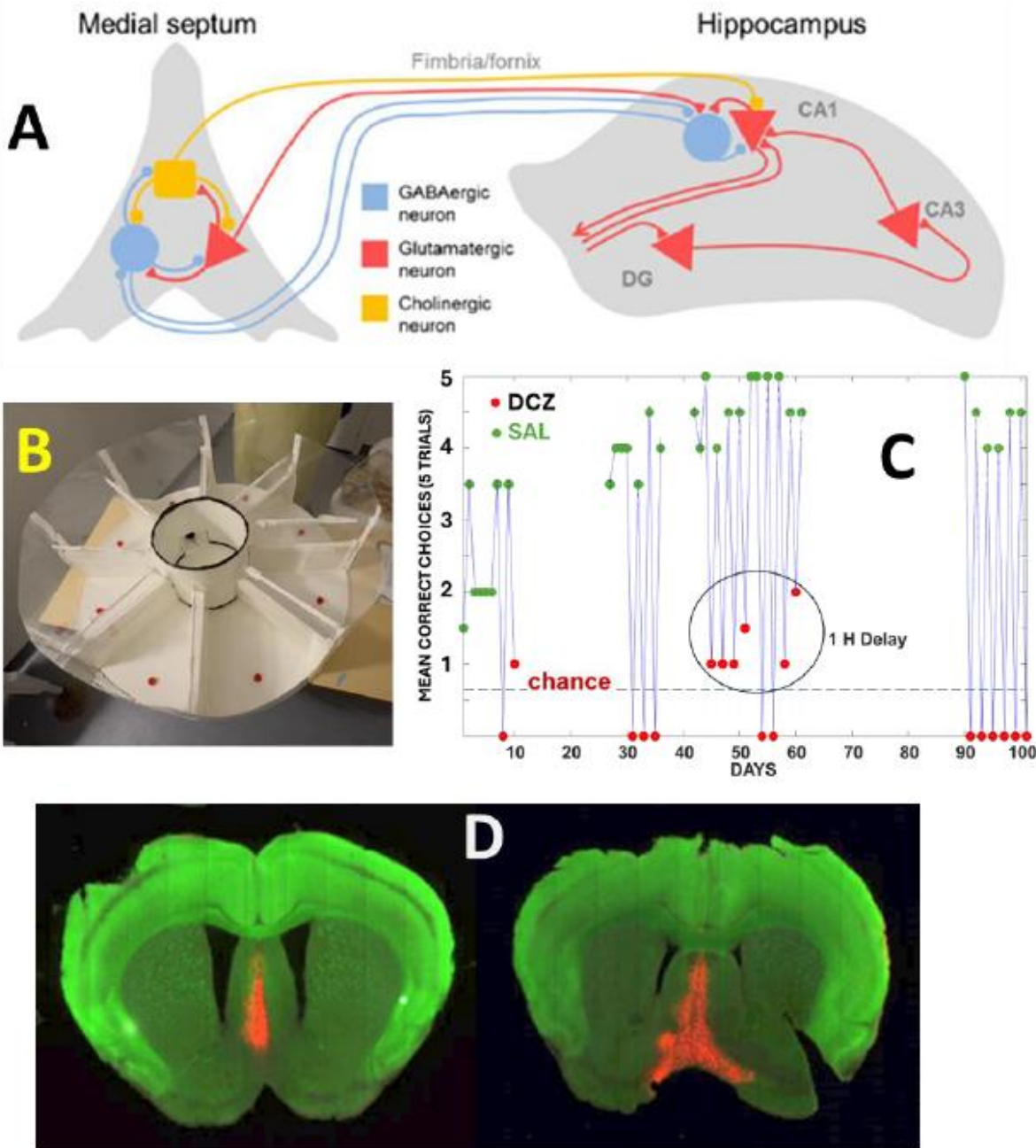
Title: Inhibition of medial septum interneurons with DREADDs attenuates hippocampal theta rhythm and impairs spatial memory while preserving hippocampal cell firing

Authors: J. D. FERBINTEANU¹, Z. ALIZADEH², J. L. SHOBE³, ***M. J. ECKERT**¹, B. L. MCNAUGHTON⁴;

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Abstract: Hippocampal place cells provide a cognitive map that is important for behaviour and memory formation. Place cell firing is modulated by the theta rhythm and also exhibit theta phase precession. Phase precessing spikes from different place cells form compressed ‘theta sequences’ within each theta cycle, which are thought to be important for sequence learning. One way to test this prediction would be to disrupt theta without altering the basic firing properties of place cells. Since theta depends on medial septum activity (MS), we used DREADDs to inhibit MS interneurons to assess the effect of reduced theta on behaviour and hippocampal cell firing. In 2 mice, we assessed the effect of DREADD activation on theta activity and hippocampal firing. Mice received viral injections in the MS to express a DREADD to suppress inhibitory neurons (<https://www.addgene.org/83896/>), and then trained to run on a 150 cm treadmill while viewing a virtual environment. Silicon probe recordings were performed in dorsal hippocampus / parietal cortex and ventral hippocampus / visual cortex while mice ran in the VR. Following injection of DCZ (0.5 mg/kg), theta amplitude was drastically reduced for up to 1 hr. Hippocampal unit activity was not grossly affected during theta disruption. Overall multi-unit activity was maintained and there was no systematic increase or decrease in individual unit firing. In our sample, there were relatively few cells with place fields prior to DCZ injection, but our data suggest that place fields are not significantly affected by theta disruption. Two separate mice with MS DREADD were tested on a spatial reference memory task on an 8-arm maze with the rewarded arm changed randomly each day. They were given 2, forced choice, “information” trials and then 5 trials of free choice. DCZ administered orally dropped performance to chance levels for about one hour. These preliminary data indicate that DREADD suppression of MS interneurons is an effective method to repeatedly disrupt hippocampal theta and spatial memory without affecting overall hippocampal activity.



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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

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Topic: I.08. Learning and Memory

Support: NIH R01 NS121764-02
NIH RF1 NS132041-01

Title: The influence of context on visual object learning and generalization

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Abstract: Naturalistic learning occurs within rich backdrop of contextual information. Despite this complexity animals learn to rapidly recognize content specific features such as visual objects across distinct contexts. However, how contextual variables such as location and visual stimuli contribute to this process remains unclear. Evidence suggests that during experience the hippocampus (HPC) generates context specific firing patterns that widely project to the neocortex (NC) which, in turn, makes heteroassociative 'links' to the contents of memory. To better understand how these 'links' are modified during learning, we have paired a novel virtual reality behavioral paradigm for head-fixed mice with large-scale silicon probe recordings that simultaneously target CA1 of HPC and two cortical regions: the visual and posterior parietal (PPC) cortices. During training (~1 month), mice learn to discriminate between dynamic visual objects (located at fixed positions) that have distinct features (rotating and changing colors) when traversing through static naturalistic landscape scenes (VR context). On recording day, we challenge mice with two distinct types of novel trials: 1) we place the familiar visual objects in a novel context with no change in position and 2) we keep the visual objects in the same familiar scene but 'swap' their positions so that mice encounter the objects in a novel location. Surprisingly, we find that task performance significantly improves only during the 'swapped' trial block, suggesting that mice become better at recognizing the objects specifically under these conditions. Indeed, we observe that object tuned neurons typically encode both visual and positional contextual information especially in the PPC. We are currently conducting trial specific analyses to test the hypotheses that the 'swapped' trials facilitate object specific attribute abstraction by reducing contextual interference. If true, this would suggest an inverse relationship between the strength of positional-attribute 'links' and the ability of mice to generalize visual objects.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

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NIH RF1 NS132041-01

Title: Regional and laminar differences in spatial context versus content coding across the mouse hippocampus, posterior parietal, and primary somatosensory cortices

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Abstract: The brain creates an internal representation of the world by extracting knowledge about the context (where) and content (what) of experiences. The indexing theory postulates that the hippocampus (HPC) generates a unique index code to associatively link the context with sensory content stored within the neocortex (NC) during an episodic experience. Recent studies have demonstrated sparse, place cell sequences in the superficial layers (L2/3) of multiple cortical regions. However, the functional role of deep layers (L5/6) of NC in context versus content coding remains elusive. To investigate this, we developed a multi-modal treadmill behavioral paradigm for head-fixed mice that integrates virtual reality (VR) environments with discrete tactile cues. On recording day, trial types included two distinct, familiar VR contexts and two novel tactile cues that were added and swapped in position. This allowed us to decouple cue activity from location-specific activity in a double dissociation design. Using high-density (512 channel) silicon probes, we simultaneously recorded activity across superficial and deep layers of the primary somatosensory cortex (S1), posterior parietal cortex (PPC), and dorsal CA1 of HPC in mice (N=6). We applied a generalized linear model on single-unit activity to quantify the relative contributions of context, position, and cue information and found that S1 neurons were more tuned to novel tactile cues, whereas PPC and HPC firing activity was better predicted by context or position. Using population vector correlation analyses, cue changes alone (while holding context and position constant) revealed that S1 had the highest tactile cue discrimination. In contrast, PPC exhibited greater selectivity to spatial position and was comparable to HPC. Laminar analyses revealed that superficial layers of PPC had more orthogonal representations and greater global remapping in response to VR context changes compared to deep PPC layers. Deep S1 tended to show stronger cue-detection activity than superficial layers, consistent with the idea that deep layers in primary cortices mediate bottom-up sensory processing. Finally, we found that PPC L5/6 was more strongly modulated by HPC theta oscillations and contained more phase-locked cells with higher mean resultant vector length and theta modulation index compared to S1 L5/6. Altogether, our data reveal regional and laminar differences in the coding of spatial context versus sensory content across the neocortical hierarchy and hippocampus to support episodic memory and the extraction of semantic knowledge.

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Poster

PSTR302: Spatial Coding in Hippocampal: Cortical Networks

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Title: Region-specific reactivation reflects context and content-dependent encoding of sensory cue changes during awake rest

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Abstract: Reactivation of neural ensembles during quiescent periods is thought to support memory consolidation. While this phenomenon has been widely observed in both hippocampal and cortical regions, the regional specificity and functional relevance of reactivation in response to sensory cue changes remain poorly understood. Here, we investigated how neural activity and offline reactivation patterns are influenced by the addition and swapping of tactile cues on a highly familiar treadmill belt (Demchuk et al., 2024), focusing on the hippocampus (HPC), retrosplenial cortex (RSC), and secondary motor cortex (M2). We observed that cue addition increased neuronal activity near cue locations more in M2 than RSC. Swapping the positions of tactile cues altered population activity in M2 relative to preceding familiar sessions, whereas RSC activity remained stable, highlighting a divergence in how these regions process environmental changes. Reactivation analysis using Principal component analysis (PCA) revealed that the introduction of cues increased the number of reactivated cells during post-trial rest periods (approximately 15 s) across all regions. However, reactivation patterns differed by regions: RSC showed a preferential increase in reactivation following cue addition, especially in early trials, suggesting a rapid, context-sensitive engagement. In contrast, M2 exhibited preferential reactivation for neurons encoding swapped cue locations immediately after cue-swap sessions, indicating more content-driven encoding. The increase in reactivation in M2 was largely accounted for by neurons encoding the locations of swapped cues. Together, these findings suggest that the cortical representation of sensory changes is dynamically reorganized during active behavior and awake rest in a region-specific manner. RSC may play a greater role in context-dependent memory encoding and consolidation, while M2 may support content-specific sensorimotor adaptation. These mechanisms may underlie the brain's ability to flexibly integrate environmental changes into memory-guided behavior.

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Poster

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Topic: I.08. Learning and Memory

Support: Swiss National Science Foundation (PR00P3_208598, X.Z.)
Synopsis Foundation (Grant Agreement 2022-CDA02, X.Z.)
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URPP AdaBD

Title: Synaptic, cellular, and network diversities govern membrane potential dynamics of CA1 pyramidal cells locked to awake hippocampal ripples

Authors: *T. LIM^{1,2,3}, P. M. JONAS⁴, F. HELMCHEN^{2,3,1}, X. ZHANG^{4,2,1};
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Abstract: Neuronal replay during sharp-wave ripples (SWRs) plays a critical role in memory consolidation, learning, and planning. While substantial evidence supports this functional relationship, the cellular mechanisms governing which neurons are recruited for replay remain scarce. To address this question, we performed whole-cell patch-clamp recordings of CA1 pyramidal cells together with local field potential recordings in the CA1 pyramidal layer of awake, behaving mice. Our findings reveal that CA1 pyramidal cells exhibit a pronounced depolarization followed by prolonged hyperpolarization around awake ripples. The variability in ripple-locked Vm depolarization is largely explained by pre-ripple Vm, participation, and cellular heterogeneity. The long-lasting hyperpolarization (~200 ms) at ripple offset is driven by high frequency ripples (~140 Hz) followed by a prominent network rebound. Moreover, a subset of CA1 pyramidal cells preferentially fires during awake ripples (SWR_{pref+}), while the majority do not (SWR_{pref-}). SWR_{pref+} neurons display significantly larger ripple-associated depolarizations than SWR_{pref-} cells. While both groups show similar hyperpolarization after high-frequency ripples with network rebound, SWR_{pref+} neurons are less inhibited following low-frequency ripples lacking a strong rebound. Thus, our findings suggest that intracellular Vm dynamics underlying neuronal participation during awake ripples are shaped by a synergistic interplay of synaptic inputs before ripples, cellular heterogeneity, and ripple diversity.

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Poster

PSTR303: Oscillations

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Support: JST ERATO (JPMJER1801)
The Institute for AI and Beyond of the University of Tokyo
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AMED Brain/MINDS 2.0 (24wm0625207s0101)
AMED Brain/MINDS 2.0 (24wm0625401h0001)
AMED Brain/MINDS 2.0 (24wm0625502s0501)

Title: Hippocampal ripple-contingent operant conditioning enhances memory

Authors: ***R. KOIKE**¹, N. MATSUMOTO^{2,3}, Y. IKEGAYA^{2,3};

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Abstract: Memory is the sculpted residue of wakeful encoding, refined through sleep-dependent neuronal reactivation, a phenomenon in which behaviorally relevant pyramidal cells in the hippocampus fire again. The neuronal reactivation often exhibits a form of temporarily organized and compressed sequences of pyramidal cell firing during ripples, high-frequency (110-230 Hz) transient oscillatory events that occur during sleep, rest, and immobility. In this sense, hippocampal ripples in sleep play a crucial role in consolidating memory acquired during wakefulness. Previous studies have applied neurofeedback—defined as monitoring brain activity and providing operant conditioning via contingent stimulation to guide the brain toward a desired neural state—to enhance memory by facilitating learning, recall, and consolidation. However, to our knowledge, no study has simultaneously delivered feedback during sleep and assessed memory performance during the awake state. We previously demonstrated that synaptic activity in hippocampal neurons could be operantly conditioned through neurofeedback, indicating the potential for reinforcing specific neural activity patterns during wakefulness. Based on this concept, the present study implemented real-time neurofeedback targeting sleep-associated ripples, combining classification based on ripple parameters with contingent reward stimulation to the medial forebrain bundle (MFB), to directly enhance hippocampus-dependent memory performance. Neural activity in the CA1 region of the hippocampus was recorded during natural sleep in Long-Evans rats aged 7 to 9 weeks, using insulated nichrome wires. Local field potentials (LFPs) and sleep states were analyzed in real time using a MATLAB program. When ripples were detected in the hippocampus, a 50 μ A current stimulus was applied to the MFB using two insulated nichrome wire electrodes less than 50 ms after detection. We assessed hippocampus-dependent memory performance using a novel object location (NOL) test, in which training and test sessions were conducted before and after neurofeedback sessions, respectively. Neurofeedback was applied until a total period of non-rapid eye movement (NREM) sleep reached 1 hour. The frequency of hippocampal ripples during sleep was increased after neurofeedback. Furthermore, daily neurofeedback sessions progressively improved NOL

performance. These results suggest that ripple-contingent operant conditioning may promote neuronal reactivation during sleep, thereby enhancing memory consolidation.

Disclosures: R. Koike: None. N. Matsumoto: None. Y. Ikegaya: None.

Poster

PSTR303: Oscillations

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Program #/Poster #: PSTR303.03/PP16

Topic: I.08. Learning and Memory

Title: Characterizing Curiosity through Electroencephalography

Authors: *A. NAKAMA, E. L. STEGEMOLLER;
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Abstract: Curiosity attracts curious individuals toward new information. Motivation for new information results in increased memory accuracy. Current research has used electroencephalography (EEG) to help understand the brain dynamics associated with the curious state of mind, but gaps remain. The trivia paradigm is the most commonly utilized technique employed to determine how changes in certain frequencies of brain oscillations are associated with the curiosity. However, timing of when the visual trivia question is recognized versus when brain oscillations alter in response to a curious stimulus has not been tightly controlled. Thus, this study seeks to standardize the time frame in which curiosity is realized by using auditory trivia questions to address this gap. After a screening phase to determine topics that participants were interested in, 64-channel EEG (Biosemi) was collected while trivia questions and answers were presented. The anticipation period, the time window between the question and answer, both presented verbally, was synchronized to the ending of the question and the beginning of the answer. The four-second time window was tightly controlled on each trial to obtain accurate timing. After presentation of trivia questions and answers, a memory recall test was obtained. The aims of this study are to determine which changes in power in EEG over frontal and midline cortical sites during answer anticipation and presentation relate to curiosity and memory accuracy. EEG data was filtered accordingly and analyzed in two different epochs. The first epoch included the four-seconds of anticipation and the second epoch for the four-second answer presentation. A Morlet wavelet analysis was applied to each epoch to determine when changes in power occurred over time. Electrodes of interest included frontal AF7, AF8, and midline FZ, and FCZ. Preliminary results showed differences in EEG power that were related to curiosity and memory performance. Data collection is ongoing to confirm results. Nonetheless, because timing between question and answer were tightly controlled, these results allow further clarity on which neural signatures are related to curiosity versus memory performance. This information can be used for innovative teaching strategies in the future.

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Poster

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Topic: I.08. Learning and Memory

Support: Office of Naval Research N00014-20-1- 2578

Title: Fronto-hippocampal synchronization in rapid spatial learning in humans

Authors: *M. GEVA-SAGIV¹, S. JUN^{5,6}, M. KRYVEN⁸, J. TENENBAUM⁹, E. D. BOORMAN², R. C. O'REILLY¹⁰, J. J. LIN³, I. SAEZ⁷, C. RANGANATH⁴;
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Abstract: Achieving goals in real-life situations often requires rapid learning, and then planning based on experience that suits the situation and desired outcome. Neurophysiological research in animal models has indicated that the orbitofrontal cortex (OFC) and hippocampus collectively support this behavior by mediating relationships between memory, actions, and outcomes. Little is known about how neurophysiological activity in the human brain supports interactions between the hippocampus and OFC in the service of goal-directed behavior. Hippocampal ripples are brief high-frequency oscillations reflecting synchronized neuronal firing and have been suggested as a window of elevated coordination between the hippocampus and OFC, and here we examined whether ripples might play a similar role during active, goal-directed behavior. We recorded intracranial electroencephalography (iEEG) to simultaneously track brain activity from both OFC and MTL areas while participants (n=10) searched for a hidden goal in a series of 2D mazes in an animated game. Our analysis contrasted high-significance events (planning and goal entry) with low-significance events, like moving in highly constrained paths. We found that gamma power in the OFC during high-significance events was strongly modulated by learning and memory demands. OFC gamma power significantly increased when planning trajectories and when reaching the goal position, but not during constrained moves. Second, we found that OFC gamma power was modulated by fast-frequency ripple oscillations detected in the medial temporal lobe (MTL) and that ripple-triggered gamma increases were associated with improved behavioral performance during memory-guided navigation. These findings suggest that OFC-MTL interactions during planning and attainment of goals support one-shot learning and memory-guided action. We propose that hippocampal ripples create a narrow window, orchestrating hippocampal-cortical communication required for successful goal encoding for future behaviors.

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Poster

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Support: BrightFocus Grant A2020321S

Title: Parafacial-zone mediated slow-wave sleep enhancement modulates physiological, molecular and behavioral plasticity

Authors: *H. S. GOMPF¹, C. ANACLET²;
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Abstract: Sleep deprivation negatively impacts all aspects of synaptic plasticity and memory consolidation. Our model of slow-wave sleep (SWS) enhancement allows us to test the role of deep sleep in these processes in the absence of other sleep / wake stages.

Here we use chemogenetics to stimulate GABAergic neurons of the parafacial zone of the brainstem to elicit at least three continuous hours of uninterrupted, high slow-wave activity deep sleep.

In adult, aged and aged Alzheimer's model (APP/Psen1) mice we enhance sleep between training and testing in the Barnes maze and novel object placement task. We also ask whether three hours of SWS enhancement affects hippocampal CA1 synaptic plasticity and track transcriptomic changes.

Generally "improved" measures of learning and memory are found in each measure, particularly in aged and aged Alzheimer's mice.

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Poster

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Title: Facilitation of associative memory encoding by LEC-mediated intercortical Gamma synchrony

Authors: *D. YUN, J.-S. GUAN;
ShanghaiTech Univ., Shanghai, China

Abstract: Memory encoding requires precise and coordinated communications between multiple brain regions. Neural oscillations of different frequencies provide temporal synchronization of neuronal population activity and are an important information-processing mechanism in the brain. Gamma oscillations are involved in associative memory encoding, however, the underlying neural mechanism remains unclear. Our previous study showed for the first time that optogenetic activation of intercortical Gamma synchrony mediated by the lateral entorhinal cortex (LEC) rescued contextual fear conditioning and spatial memory storage in hippocampus-lesioned mice. We hypothesize that the LEC receives reinforcement learning signals encoded by dopamine neurons in the ventral tegmental area (VTA) and then regulates the level of intercortical gamma synchrony to transmit reinforcement learning signals to various regions of the cerebral cortex, ultimately contributing to the formation of associative memory networks. In this work, we combined neural circuit tracing, multi-channel electrophysiological recording, two-photon Calcium imaging, and optogenetic manipulation to investigate the neural circuit mechanism of Gamma synchrony in facilitating associative memory encoding in terms of correlation and causality.

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Poster

PSTR303: Oscillations

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Topic: I.08. Learning and Memory

Support: JKST&IC/14/2023

Title: Impact of Acoustic Loop Music-Based Interventions on Cognitive Decline in Mild Cognitive Impairment and Alzheimer's disease: An Indian Home Care Approach

Authors: *D. S. AASEM;
Cognitive Musicology/Neuro Acoustics, Kashmir advanced Scientific research Ctr. KASRC,
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Abstract: Age-related cognitive impairments such as Mild Cognitive Impairment (MCI) and Mild Dementia (MD) are rising sharply in India, yet early-stage treatment options remain scarce, particularly in rural and underserved populations. Current pharmacological interventions provide limited benefits and are often inaccessible due to cost and availability. Addressing this gap, we propose the SM Model, a novel neuro-acoustic intervention utilizing structured, frequency-specific sound loops designed to stimulate brainwave frequencies linked to memory, attention, and emotional regulation. This personalized, home-based therapy was carried out at the Kashmir Advanced Scientific Research Centre (KASRC), a DSIR-accredited research institution that involved caregiver participation and differed from conventional music therapy by targeting precise cognitive functions through acoustic modulation. In a case-control trial, Pilot based study 200 participants (100 MCI and 100 healthy controls) underwent a 12-week intervention delivered via mobile applications or acoustic kits. Cognitive and emotional outcomes were assessed pre- and post-intervention using validated measures. Preliminary results reveal that the combination of personalized musical intervention with neuro-acoustic stimulation significantly enhances cognitive performance, emotional well-being, and patient-caregiver communication. The study aims to validate a scalable, low-cost approach that holds promise for integration into national dementia care frameworks and local health systems across India.

Disclosures: **D.S. Aasem:** A. Employment/Salary (full or part-time): Director at Kashmir Advanced Scientific Research Centre KASRC University Of Kashmir Hazratbal Jammu and Kashmir India.

Poster

PSTR303: Oscillations

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Program #/Poster #: PSTR303.08/QQ1

Topic: I.08. Learning and Memory

Support: NIH Grant 2K12GM133314-06

Title: Network States Across Threat Behavioral Outcomes

Authors: ***K. DORST**^{1,2}, J. MAGUIRE²;

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Abstract: Valence processing during threat discrimination will appropriately assign negative value to stimuli perceived as dangerous and positive value to those indicative of safety. Conversely, threat generalization broadly assigns negative value to a variety of stimuli. Generalization is useful for constantly changing environments or when the probability of a threat is unknown. Yet, deficits in valence processing produce pathological overgeneralization that occurs in neuropsychiatric disorders such as PTSD and anxiety. The neural substrates that mediate this interplay between discrimination and generalization are largely unknown. We use a systems-level approach to dissect the network states across a two-tone threat discrimination task.

Our first experiment utilized local field potential recordings of the basolateral amygdala (BLA), a known valence processing hub, to identify differences in oscillation dynamics across discriminators and generalizer mice. Preliminary results indicate a bias towards a “pro-fear” 2-5 Hz frequency in generalizers across both fear and safety tones that is not observed in discriminators. Concurrent work utilizes a holistic network approach to determine functional interactions between known hub regions for valence processing (i.e., the BLA) and others previously unidentified in this fundamental process. Future work will use artificial manipulations of these network states to bias behavioral outcomes across the task. These works have translational implications in our understanding of how brain states contribute to (mal)adaptive behavioral processes, with direct relevance to mechanisms of neuropsychiatric disorders where overgeneralization is symptomatic.

Disclosures: **K. Dorst:** None. **J. Maguire:** F. Consulting Fees (e.g., advisory boards); Scientific Advisory Board for SAGE Therapeutics, Inc..

Poster

PSTR303: Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR303.09/QQ2

Topic: I.08. Learning and Memory

Support: NIH Grant NS106447

Title: Investigating human memory signatures ex vivo

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Abstract: Memory related disorders are devastating conditions that disrupt higher order cognition and impose a profound emotional and financial burden. Higher order cognition requires multi-dimensional neuronal communication observed in the form of neural oscillations (local field potentials, LFPs), that serve as key signatures of memory related activity in the human brain. Disrupting cholinergic tone has been found to profoundly impair memory encoding in humans by acting on LFP activity. Despite the diverse treatment strategies, a limited understanding of human brain activity severely limits therapeutic development. Here, we will utilize live, cultured human cortical slices (ages 18-55; both sexes) to study memory relevant neural oscillations and their genetic signatures. Application of the cholinergic agonist carbachol was used to induce LFP activity during high-density microelectrode array (MEA) recordings. Immediately following MEA recordings, the same slices are then processed for spatial transcriptomics (VisiumHD, 10x Genomics). Preliminary results indicate specific electrophysiological dynamics of cholinergic modulation, specifically increase in power in the

slow theta band (2-5Hz), and an upregulation in memory relevant gene expression in human cortical tissue. Ongoing analysis aims to reveal laminar specific molecular signatures (i.e. differentially expressed genes) and laminar specific electrophysiology dynamics such as firing rate and LFP coherence properties. We then aim to integrate these data to correlate differential, cell type specific molecular contributions to electrophysiological characteristics in the human cortex. Combining electrophysiology with genomics is a multidisciplinary approach within the larger field of neuroscience to provide novel insights into mechanisms of human memory signatures. Our results highlight a novel framework for identifying gene level contributors to memory-relevant neural dynamics in the human cortex. This platform has the potential to uncover therapeutic targets for memory disorders and guide strategies for precision interventions such as gene therapy and neuromodulation.

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Poster

PSTR303: Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR303.10/QQ3

Topic: I.08. Learning and Memory

Title: α -tACS modulates reward-dependent pupil responses and corticostriatal connectivity

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Abstract: Introduction. Noninvasive brain stimulation can help clarify the neural basis of reward processing and potentially inform treatments for disorders involving reward dysfunction. However, widely used methods such as transcranial magnetic and electrical stimulation cannot directly stimulate deep-brain regions like the striatum. Here, we tested whether stimulating the ventrolateral prefrontal cortex (VLPFC)—a cortical region strongly connected to the striatum—could indirectly influence reward-related neural and physiological responses. **Methods.** In a within-subjects design, participants performed a card-guessing task involving monetary rewards for correct guesses and punishments for incorrect guesses. During the task, participants underwent functional magnetic resonance imaging (fMRI) and pupillometry while receiving concurrent 10 Hz transcranial alternating current stimulation (α -tACS). Stimulation targeted either the VLPFC or a control region (temporoparietal junction). We measured pupil dilation, brain activation (BOLD signal), and functional connectivity between the ventral striatum and dorsal anterior cingulate cortex (VS-dACC). **Results.** VLPFC stimulation increased pupil size during reward and punishment outcomes, indicating greater physiological arousal. At the neural level, α -tACS enhanced VLPFC activation during reward and suppressed its responses during

punishment. Stimulation also changed VS-dACC connectivity in a context-dependent manner. Importantly, stimulation-driven increases in pupil size during reward correlated positively with stimulation-induced changes in VS-dACC connectivity. Exploratory moderated mediation analyses suggested that stimulation influenced the degree to which striatal responses mediated the relationship between task outcomes and pupil size changes. **Conclusions.** Targeting VLPFC with α -tACS modulates local cortical activity and corticostriatal networks during reward processing, providing a promising noninvasive approach to influence reward circuitry.

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Poster

PSTR303: Oscillations

Location: SDCC Halls B-H

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Program #/Poster #: PSTR303.11/QQ4

Topic: I.08. Learning and Memory

Support: NINDS Grant K23NS110920
NSF CogNeuro Award 2148753

Title: Dissociating physiological ripples and epileptiform discharges with vision transformers

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Abstract: Two frequently studied hippocampal neural events are normal physiological ripples and abnormal interictal epileptiform discharges (IEDs). Despite distinct waveforms, IEDs are often misclassified as ripples by standard automated methods due to sharp edge artifacts, which have created challenges for studying ripples and IEDs independently. We leveraged recent advances in computer vision on time-frequency feature representations to enable more comprehensive dissociation. We retrospectively evaluated human intracranial recordings from 46 hippocampal depth electrodes across 17 patients with focal epilepsy, most with hippocampal seizure-onset. Using a common ripple detector, we identified 31,847 ripple/IED candidates and generated broadband spectrograms. Based on expert label overlap, a median of 3.9% per patient (range: 0-47.2%) were classified as IEDs. Those candidates were projected into a low-dimensional space and segmented with k-means to infer pseudo-labels. Independently, expert IED annotations were used for validation. Treating spectrograms as images, we trained vision transformer models for binary classification of IEDs vs. ripples. Expert-labeled training with 5-fold cross-validation yielded a mean AUC of 0.970 and a precision-recall score of 0.694. Leave-one-patient-out cross-validation using pseudo-labels for training and expert labels for testing achieved near-expert performance (mean AUC 0.966, range: 0.892-0.997), demonstrating generalizability. Attention maps showed models focused on triangle-like edge artifacts.

Spectrograms separated canonical IEDs vs. ripples better than raw or ripple-filtered waveforms. Canonical ripple and IED candidates emerged at opposite poles with a continuous landscape of intermediates in between. Transformer-derived attention maps revealed that models were tuned to triangle-like edge artifact spatial features in the spectrograms. Model-derived probabilities (i.e. of being an IED) for all candidates demonstrated continuous transitions between ripples vs. IEDs, as opposed to binary clustering. Vision transformers nevertheless perform virtually at human expert levels in dissociating these phenomena by leveraging time-frequency spatial features enabled by neural data spectrograms. This offers promising tools for advancing cognitive neurophysiology and optimizing closed-loop epilepsy therapies.

Disclosures: D. Zhang: None. J. Kleen: None.

Poster

PSTR303: Oscillations

Location: SDCC Halls B-H

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Program #/Poster #: PSTR303.12/QQ5

Topic: I.08. Learning and Memory

Support: NINDS Competitive Postdoctoral Fellowship Award
NIH/NINDS ZIA NS003144

Title: Direct electrical microstimulation evokes high-frequency ripple oscillations in humans

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Abstract: The development of effective neurostimulation treatments for loss of episodic memory depends on a detailed understanding of the neural signals that support memory processes and characterization of how stimulating the brain in different ways modulates these memory-related patterns. Direct electrical stimulation holds the potential to treat memory disorders; however, the mechanisms underlying the brain's memory-related neural circuits and how they respond to stimulation remain poorly understood. To address this, we collected human intracranial EEG recordings from neurosurgical epilepsy patients while delivering open-loop stimulation to the hippocampus and neocortical regions at a range of microampere-level amplitudes. We measured high frequency-oscillation events (80-120Hz), known as ripples, that have been shown to underlie memory encoding, consolidation, and retrieval while applying a range of stimulation sequences. We observed that high-frequency trains of microstimulation trains applied at 50-100uA at cortical and MTL locations increased local ripple rates. On each bipolar electrode pair, we conducted 5-minute stimulation sessions with alternating 30-second baseline and stimulation blocks (1 or 5 pulses at 100 Hz delivered once per second) at increasing amplitudes (1, 15, 25, 50, and 100 μ As), finding significant amplitude-dependent increases in

ripple rates during stimulation compared to baseline across patients. We observed significant amplitude-dependent changes in cortical ripple rates coinciding with stimulation of hippocampal regions. We found no significant differences in the duration and amplitude of electrically evoked and physiologic ripples across all patients suggesting evoked and physiologic ripples have similar morphology. Lastly, we found that spiking activity in the anterior temporal lobe increases as spectral power in the ripple band increases from stimulation implying evoked ripples also have underlying spiking activity. This work provides direct evidence in humans that low-amplitude hippocampal stimulation evokes local high-frequency ripple oscillations.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: I.08. Learning and Memory

Support: R01Mh122391
R01Mh139216
U19NS107616
SNSF Mobility Fellowship (P500PB_21440)

Title: Bridging scales of neural activity: predicting local field potentials from multi-area spiking activity

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Abstract: Local field potentials (LFPs) offer a valuable window into coordinated neural activity, reflecting the summed synaptic inputs and intrinsic membrane properties of neuronal populations. Understanding how specific LFP patterns arise from multi-regional spiking activity is crucial for deciphering neural communication across interconnected areas. We investigated the relationship between spiking activity in multiple areas, including V1, higher-order visual areas, visual thalamus, hippocampus, midbrain, and brainstem, and LFP in V1, using a deep learning framework. We trained deep neural networks on simultaneously recorded multi-regional spiking activity from mice performing a visual go/no-go change detection task on natural images, using single-unit spiking activity as input features. Different models were optimized to predict either the LFP in different layers of V1 or its power in separate frequency bands. We used integrated gradients to identify the most influential neurons and spiking patterns for predicting LFP in different V1 layers and frequency bands. Our results demonstrate that the model can accurately predict broadband and lower frequency V1 LFP from multi-regional spiking activity ($R^2 > 0.7$),

even generalizing to unseen spontaneous activity, with prediction accuracy decreasing at higher frequencies. Attribution analysis revealed that distinct brain areas contribute to specific V1 LFP frequency bands, with higher frequencies exhibiting a more local (V1-only) attribution. Furthermore, interneurons conveyed more information about the LFP than pyramidal cells, even after normalizing firing rates, likely due to their broader integration of neural inputs. This study provides a powerful, layer- and frequency-specific framework for linking microscale spiking activity to mesoscale LFP dynamics across interconnected brain regions within the visual system, offering new insights into the neural mechanisms underlying both task-related and spontaneous visual processing.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

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Topic: I.08. Learning and Memory

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Title: Dentate gyrus interneurons modulate winner-take-all network dynamics.

Authors: *T. HAINMUELLER¹, E. HEYNOLD³, N. PALEOLOGOS⁴, G. BUZSAKI²;
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Abstract: The hippocampus is a central hub in the brain and a key player in processing memories. Its functions are supported by the dentate gyrus (DG), which parses and segregates patterns of excitation from the entorhinal cortex and houses a unique cytoarchitecture, including powerful inhibitory circuits. Despite their importance, few studies have recorded identified DG interneurons in behaving animals. Here, we used electrophysiological recordings and bidirectional optogenetic manipulations to characterize the two major inhibitory neuron types in the DG: parvalbumin (PV) and somatostatin (SST) expressing interneurons. We employed machine learning to classify granule cells (GC), mossy cells (MC), PV- and SST interneurons in the same recordings based on electrophysiological criteria to study their synaptic interactions and how they shape spatial information and responses to entorhinal synaptic inputs during dentate spikes. We demonstrate a central role for SST interneurons in regulating the impact of entorhinal

excitation, while PV interneurons are critical for feedback inhibition. Finally, optogenetic manipulations revealed a paradoxical observation: instead of merely suppressing firing in the surrounding network, DG interneuron stimulation exacerbated preexisting ‘winner-take-all’ dynamics, including strong paradoxical excitation of a small group of excitatory neurons likely through polysynaptic network mechanisms. Such use-dependent modulation of winner-take-all dynamics may select content-bearing ensembles of DG principal cells during cognitive processes.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

Location: SDCC Halls B-H

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Topic: I.08. Learning and Memory

Support: R01 NS113071
Yoshida Scholarship Foundation

Title: Rapid brain state alterations during vocal interaction in the singing mouse (*S. teguina*)

Authors: *Y. FUJISHIMA¹, C. KEMERE², Y. ZHANG³, G. BUZSAKI⁴, M. A. LONG⁵;
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Abstract: The Alston’s singing mouse (*Scotinomys teguina*) produces loud, human-audible ‘songs’ lasting 6-10 seconds, with acoustic features unique to each individual. These mice use their songs as vocal markers of individual identity during spatial exploration of their home range (spontaneous singing) and as vocal responses to songs from distant conspecifics (counter-singing). This provides a unique opportunity to investigate the neural dynamics underlying ethologically relevant interactive vocal communication. Although previous studies have showed that the production of successful vocal response requires the orofacial motor cortex, how the brain integrates the external and internal factors to influence vocal responses remains unclear. Given that spatial and social information is believed to be computed in the hippocampal system, we recorded from the CA1-dentate axis of the dorsal hippocampus with silicone probes in freely moving singing mice. Our local-field potential (LFP) analysis shows increased theta power during attentive listening to a conspecific song. Unexpectedly, we find that when the mouse begins to sing, the LFP exhibits a steep decrease in theta power and an increased incidence of sharp-wave ripples, accompanied by a commensurate decrease in acetylcholine release in the hippocampus, characteristics typically associated with ‘offline’ behaviors such as non-REM sleep and reward consumption. These results indicate that singing mice rapidly switch between

complementary brain states when engaged in perceiving the song of a conspecific and self-vocalization. Further experiments and analyses will investigate how spiking activity, combined with neuromodulator signals in the hippocampal network, encodes relevant information to facilitate rapid transitions in brain states and behavior.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

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Topic: I.08. Learning and Memory

Support: NIH Grant 5T32GM136573-05

Title: CreER-dependent ChR2 labeling in CA1 and dentate gyrus of TRAP2 mice exposed to novel environments

Authors: *D. BEAM¹, H. JUN^{2,1}, G. BUZSAKI¹;

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Abstract: Targeted Recombination in Active Populations (TRAP2) mouse lines enable selective and long-term labeling of neurons that are active during a defined time window, via cFos-driven CreER expression and tamoxifen-induced recombination leading to Cre-dependent ChR2 expression. This allows for the tagging of neurons active during specific processes of interest, such as memory encoding. The physiological properties of the tagged neurons, however, remain poorly understood, and it is unclear which hippocampal cells are preferentially labeled and why. To characterize TRAP2-labeled and unlabeled neurons electrophysiologically, we first examined ChR2-EYFP expression in CA1 and dentate gyrus (DG) of mice exploring an enriched novel environment. TRAP2xAi32 mice (who had only ever previously been in the vivarium) received a tamoxifen dose of 50 mg/kg immediately before being placed in an enriched cage for four hours. One week later, the mice were sacrificed, and their brains were perfused, sectioned, and stained with an anti-GFP antibody. Histology demonstrated sparse labeling in the CA1 region compared to densely labeled DG cells. Increasing the tamoxifen dose to 75 mg/kg did not qualitatively alter this pattern, but a dose of 100 mg/kg resulted in enhanced CA1 labeling. These histological findings are supported by electrophysiological observations. Very few neurons in CA1 were light-responsive. In contrast, when the probe was lowered to the DG, ~20% of the recorded neurons responded to light, as demonstrated by both a significant increase in firing rate following 2 ms light pulses and sustained firing during 50 ms pulses. Given the known involvement of CA1 in encoding novel spatial environments, these data suggest that the TRAP2 system is either not sensitive enough to label CA1 cells encoding environments in the absence of salient stimuli (as is present in fear conditioning) or that we have not yet identified the right condition to drive

high CA1 expression. We therefore repeated the exposure to the novel environment, incorporating a female mouse in a centrally placed wire cage while a male mouse explored, with the hypothesis that this positive motivational stimulus would lead to greater neuronal labeling. In this pilot experiment, we observed increased CA1 labeling without a corresponding qualitative change in DG, suggesting that different regions of the hippocampus may be tuned to distinct inputs. Ongoing work aims to further characterize the physiological properties of TRAP2-labeled neurons across these regions.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

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Topic: I.08. Learning and Memory

Support: NIH Grant RF1DA056394

Title: Coordinated multiregional subspaces in the hippocampus contribute to context-dependent memory acquisition and consolidation

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Abstract: A core question in systems neuroscience is how the hippocampus flexibly routes information to support memory-guided behavior against changing contexts. Here, we leverage large-scale hippocampal-neocortical recordings and partial canonical correlation analysis to reveal that subspace rotations within the CA1 population space allow for differentially integrating DG-CA2-CA3 inputs and gating communication with the neocortex. These cross-regional subspaces are not static but adapt to the spatial context and reorganize across different environments. We demonstrate that intrinsic cellular and network properties, such as laminar position, excitability, and interneuron connectivity, constrain participation in these subspaces, suggesting a biologically grounded recruitment mechanism. Crucially, these same subspaces are selectively reactivated during post-maze NREM sleep, especially during hippocampal sharp wave ripples, where their reinstatement predicts replay of waking events. Our results also suggest a differential memory consolidation role within these subspaces, because only cross-regional cell pairs that were co-active in maze subspaces showed strengthened spike transmission

in post-maze periods. Overall, our results demonstrate an adaptive network substrate for context-dependent memory acquisition and consolidation.

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Poster

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Topic: I.08. Learning and Memory

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DFG HE 9600/2-1
FACES 2024

Title: The dentate gate in health and disease

Authors: *E. HEYNOLD¹, T. HAINMUELLER², I. SOLTESZ⁴, G. BUZSAKI³;
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Abstract: The dentate gyrus is the major gateway for information to the hippocampus. It aids in the selection and segregation of excitation patterns that arrive from other brain regions. A specialized cytoarchitecture controls this gating function and is constituted by powerful inhibitory interneurons. The two major classes of interneurons in the dentate gyrus are the dendrite-inhibiting somatostatin- and the soma-inhibiting parvalbumin-expressing interneurons. Excitation patterns arriving from the entorhinal cortex can be physiological, as in the case of dentate spikes, or pathological, as in interictal epileptiform discharges (IEDs) or seizures. Dentate spikes are associated with learning and memory consolidation and retrieval, whereas IEDs disrupt cognitive function. We investigated the differential role that distinct GABA-ergic interneuron subgroups play in the propagation of IEDs in mouse models of neurodegenerative diseases. The animals were chronically implanted with high-density silicon probes and optic fibers for optogenetic manipulation of different interneuron types. We further developed a closed-loop system that can reliably distinguish dentate spikes from IEDs and interrupt IEDs within microseconds by optogenetic activation of dentate gyrus interneurons while the mice performed different tasks. By performing multi-region electrophysiological recordings, we investigate the extent and patterns of propagation of IEDs and will correlate these spatiotemporal features with behavioral memory deficits.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

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Topic: I.08. Learning and Memory

Support: NIH Grant 1R01NS113782-01A1

Title: Dynamic patterns of propagation of hippocampal oscillations along the transverse axis in healthy and Alzheimer's disease mice

Authors: *N. PALEOLOGOS¹, M. VOROSLAKOS¹, J. GONZALEZ², A. MASLAROVA¹, D. AYKAN³, G. BUZSAKI¹;

¹Neurosci. Inst., ²Dept. of Psychiatry, NYU Grossman Sch. of Med., New York, NY; ³Sainsbury Wellcome Ctr., Univ. Col. London, London, United Kingdom

Abstract: The relationship between neuronal activity and oscillatory patterns in hippocampal local field potentials (LFP) has been extensively used to study memory. High-density recordings have allowed us to examine differences in the strength and timing of inputs to hippocampal sublayers and their association with spiking, providing critical information about the network dynamics underlying hippocampal functions *in vivo*. Despite technical advances, most studies only report the average frequency band-specific power and phase distributions across depths. However, these periodic LFP components are known to be highly dynamic along both the transverse and longitudinal axes of the hippocampus (Lubenov & Siapas 2009; Schomburg et al. 2014). Moreover, how these spatial patterns differ in diseases of hippocampal dysfunction is currently unknown. In the present study, we focus on the spread of extracellular theta (6-12 Hz) and gamma (30-150 Hz) currents across the medial-lateral axis of the dorsal hippocampus in mice. To record from all regions of the dorsal hippocampus simultaneously with high spatial resolution, we employed the SiNAPS probe which contains 1024 sites distributed across 8 shanks, spanning an area of 2x2 mm. Theta and gamma power were computed by bandpass filtering the broadband LFP signal, acquired at 20 kHz and downsampled to 1250 Hz, in their respective frequency ranges and applying the Hilbert transform to extract time-varying power and phases during wake epochs for each channel. Instantaneous power in theta and gamma bands was averaged across 10 ms bins to capture activity at the timescale of synaptic events. Spatial patterns of theta and gamma power across the dorsal hippocampus corresponded to changes in extracellular currents across distinct dendritic compartments, corroborated by current source density (CSD) analysis. Stable spatial profiles capturing the motion energy of theta and gamma activity spread within the hippocampus were extracted using principal component analysis. Loadings from the top 10 components (PCs) reproduced known patterns of activation from various intrahippocampal and hippocampal-entorhinal microcircuits. Peaks in each motion energy PC were correlated with distinct 2D CSD profiles and changes in spiking activity in discrete subgroups of hippocampal neurons. To contrast these physiological activity patterns, we performed similar implantations and analyses in an APP/PS1 mouse model of Alzheimer's

disease (AD). This approach revealed AD-specific differences in the prevalence and magnitudes of theta and gamma current distributions and associated spiking patterns across the hippocampal transverse axis.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

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Program #/Poster #: PSTR304.08/QQ13

Topic: I.08. Learning and Memory

Support: MH122391

Title: Hippocampal ripples are coupled to transient shifts in autonomic tone

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Abstract: Rationale: The hippocampus is traditionally studied in the context of memory, yet it also influences autonomic and metabolic functions. To explore this brain-body interaction, we simultaneously recorded hippocampal activity and heart rate in freely behaving mice. Heart rate variability (HRV) provides a fast, non-invasive readout of autonomic nervous system activity, capturing real-time changes in sympathetic and parasympathetic tone.

Methods: To measure how hippocampal sharp wave ripples (SPW-Rs) are coupled to autonomic tone, we simultaneously measured heart rate (lead-II ECG) and local field potentials from the CA1 region of the hippocampus in freely moving mice. We collected data while the animals remained in their home cage. The total recording time ranged from a couple of hours to a full 24-hour session. To detect and analyze the heart rate variability, PhysioZoo software was used. The sleep state of the mice during the recordings was classified as non-REM, and REM sleep, wake, and micro-arousal states.

Results: We analyzed high-resolution HRV aligned to SPW-Rs. Time domain metrics (RMSSD, cardiac sympathetic index, and cardiac vagal index) revealed a transient, state-dependent shift in autonomic tone following SPW-Rs. Specifically, we observed a consistent increase in parasympathetic activity post-SPW-Rs, most pronounced during NREM sleep. Frequency-domain analyses further demonstrated that SPW-Rs were associated with significant modulations in both low-frequency (LF) and high-frequency (HF) power, indicating coordinated sympathetic and parasympathetic responses.

Conclusions: Our results indicate that hippocampal SPW-Rs are consistently followed by

transient increases in parasympathetic tone, as measured by high-resolution heart rate variability metrics. These findings suggest that SWRs may actively modulate autonomic output, offering a mechanistic link between memory consolidation, sleep, and physiological regulation.

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Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR304.09/QQ14

Topic: I.08. Learning and Memory

Title: Latent variable model with jumps reveals rich dynamics in hippocampal activity during immobility

Authors: *Z. ZHENG¹, I. Q. GARON³, I. ZUTSHI², R. HUSZAR², A. H. WILLIAMS¹, G. BUZSAKI⁴;

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Abstract: Hippocampal studies provide a rich window into the mechanisms of memory via the paradigm of spatial navigation. To interpret neural responses, it is common practice to first build encoding models using position during locomotion as the label, and then decode the position using this model. Of particular interest is the mismatch between the decoded and the actual position, for instance, replay of non-local trajectories during rest. However, it is unclear whether a priori chosen labels explain neural data the best. By imposing spatial structure to times of immobility, we may be systematically missing the influence of nonspatial factors. Furthermore, the encoding model requires an arbitrary separation of the running and immobility periods, making the transition between the two difficult to separate. Finally, many unsupervised models leverage smoothness to discover low-dimensional latents from noisy high-dimensional recordings. This assumption may prevent models from capturing sudden discontinuities in the latent space that may be present in the neural data. To solve these issues, we develop a latent variable model that is conceptually simple, flexible and computationally efficient. The model infers non-linear tuning of the latent states, and whether the state is smoothly varying or jumping. The expectation-maximization (EM) algorithm for fitting the model simply corresponds to iteratively applying the two steps in hippocampal studies: tuning curve estimation and label decoding. We apply this model to rodent CA1 datasets and show that learned latents explain pyramidal cell activities better than position labels. Furthermore, latent states exhibit abrupt but sustained transitions even within immobility epochs. Some of these transitions reflect transitioning in the behavioral state like movement initiation. Surprisingly, others do not have ostensible behavioral correlates. Overall, our model highlights the shortcomings of imposing

external spatial labels on the cognitive map and provides a path forward to an inside-out approach to understanding hippocampal function.

Disclosures: **Z. Zheng:** None. **I.Q. Garon:** None. **I. Zutshi:** None. **R. Huszar:** None. **A.H. Williams:** None. **G. Buzsaki:** None.

Poster

PSTR304: Behavioral Neurophysiology Studies in Hippocampal and Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR304.10/QQ15

Topic: I.08. Learning and Memory

Support: Marie Curie Postdoctoral Individual Fellowship

Title: Large-scale spatiotemporal coupling of the neocortex with the hippocampus

Authors: ***R. GARCIA CORTADELLA**¹, **A. SIROTA**², **D. BEAM**³, **G. BUZSAKI**⁴;
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Abstract: The hippocampus supports long-term memory processes through communication with numerous partner brain regions, yet communication between the hippocampus and distinct cortical regions is not fully understood. The hypothesis is that distinct neocortical regions interact differently with the hippocampus during memory consolidation and this distinct communication is reflected in the field potential oscillations that are generated by large neuronal populations in the cortex. To test this hypothesis, we used large scale multiplexed graphene micro-ECoG grids in combination with Neuropixel probes in the CA1 and dorsal neocortex to obtain high density LFP profiles. Our results show the importance of high density cortical mapping to capture diverse spatio-temporal sub-cycle dynamics temporally locked to hippocampal activity, revealing stereotypical travelling patterns of cortical excitability around SPW-R.

Disclosures: **R. Garcia Cortadella:** None. **A. Sirota:** None. **D. Beam:** None. **G. Buzsaki:** None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.01/QQ16

Topic: I.09. Spatial Navigation

Support: For grant support please see NeuroBatLab website

Title: Motor-hippocampal interaction during spatial navigation in freely flying bats

Authors: *X. CHEN^{1,2}, B. STYR^{3,2}, M. M. YARTSEV^{1,3,4,2,5};

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Abstract: Navigation is one of the most complex and essential behavioral functions across species, requiring precise coordination across sensory, motor, and cognitive domains. In particular, navigation requires movement whereby navigational decisions are both informed by, and must be transformed into, motor actions. To investigate the interaction between motor actions and spatial representations, we performed simultaneous large-scale (Neuropixels) neural recordings from three brain regions in freely flying Egyptian fruit bats: (i) motor cortex, (ii) dorsal hippocampus (HPC) and (iii) medial entorhinal cortex (MEC). In doing so we first discovered a robust oscillatory pattern in the bat motor cortex occurring at the theta frequency band, which emerged during flight and was strongly phase-locked to the wingbeat cycle. This oscillatory activity was NOT present in the simultaneously recorded HPC or MEC, consistent with previous reports in crawling (Yartsev et al., *Nature*, 2011) and flying (Forli et al., SfN 2025 poster) Egyptian fruit bats. Furthermore, many neurons in the motor cortex exhibited strong phase-locking to the theta rhythm. When examining the relationship in neural dynamics between motor cortex and HPC/MEC we observed clear behaviorally driven synchronization patterns emerging including (i) theta coherence (ii) and directional information flow that was modulated at specific times before, during and after flight. Together, these findings reveal a dynamic, cross-regional theta-driven routing system that could support both behavioral and computational needs during navigation. These findings can also provide a potentially unifying mechanism applicable across species, including those that exhibit hippocampal theta (e.g., rodents) and those that do not (e.g., bats, monkeys and humans).

Disclosures: X. Chen: None. B. Styr: None. M.M. Yartsev: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.02/QQ17

Topic: I.09. Spatial Navigation

Support: For grant support see NeuroBatLab website

Title: Ensemble Dynamics in the Hippocampal Formation of Freely Flying Bats

Authors: *A. FORLI¹, W. FAN¹, K. K. QI¹, M. M. YARTSEV^{1,2};

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Abstract: Cognitive functions for navigation and memory are believed to rely on emergent properties of neural ensembles in the hippocampus, such as activity replay and theta sequences. Yet, whether and how these phenomena generalize across species with distinct navigational demands and neurophysiological properties remains unclear. Here we wirelessly recorded neural activity from large populations of cells and local field potentials (LFPs) from the hippocampus of freely flying bats engaged in free, spontaneous foraging behavior. During rest, we identified time-compressed forward and reverse replay of multiple flight trajectories coinciding with sharp-wave ripples (SWRs). Yet, replay events occurred predominantly at locations that were both spatially and temporally distant from the replayed behavior, and their speed scaled with trajectory length, challenging existing models of replay mechanisms. During flight, ensembles of neurons exhibited fast representational sweeps, where the decoded location cyclically moved ahead of the bat's position. In contrast to reports in rodents, these sweeps occurred in the absence of theta oscillations and were instead phase-locked to a prominent motor behavioral rhythm - the bat's wingbeat cycle. This suggests that behaviorally-relevant sensorimotor, rather than neural, rhythms can interact with hippocampal ensemble dynamics in a highly structured manner. Combined, our findings challenge existing models of ensemble dynamics in the mammalian hippocampus and highlight the importance of comparative studies under ethologically relevant conditions for elucidating brain function. Moreover, our results raise the question of whether fast representational sequences (i.e. sweeps) may be inherited from input regions, such as the entorhinal cortex and if so, whether these share similar (or different) functional properties to those described in rodents (e.g., alternating between the left and right directions along the animal's path). Finally, the exquisite accessibility to both the motor (wing-motion) and sensory (echolocation) aspects of the bats spatial behavior at high spatial and temporal resolutions allows addressing how ensemble dynamics might relate to the navigational challenges animals face during natural spatial behaviors.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.03/QQ18

Topic: I.09. Spatial Navigation

Support: For grant support see NeuroBatLab website

Title: Periodic grid cells and toroidal dynamics in the medial entorhinal cortex of freely flying bats

Authors: *K. K. QI¹, M. M. YARTSEV^{1,2,3,4,5},

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Abstract: Grid cells in the medial entorhinal cortex (MEC) are a fundamental component of the brain's spatial navigation system. Since their discovery, significant progress has been made towards a detailed theory of how the population dynamics of grid cells give rise to the characteristic periodic firing patterns seen in two-dimensional (2D) environments. Yet, the real world is 3D and many animals, including humans, reside and navigate in three dimensional spaces. Therefore, it is important to resolve how grid cells might support 3D navigation from the single cell to the population level. In rodents, population activity of co-modular grid cells has been shown to be constrained to a 2D toroidal manifold, consistent with predictions from continuous attractor network (CAN) models. Whether grid cells are similarly confined to a 2D manifold during 3D motion, such as in the case of flying bats, remains unknown. Furthermore, it is unclear how the spatial firing patterns of grid cells in flying bats, which has been suggested in previous studies to lack global periodic structure during flight (Ginosar et al., 2021), can be compatible with established CAN models. To address these gaps, we established high density wireless electrophysiological techniques (using Neuropixels) to simultaneously record hundreds of neurons from the MEC of freely flying bats during spontaneous aerial foraging. During foraging, bats naturally developed highly stable and structured flight paths, allowing rigorous assessment of neural dynamics while controlling behavioral variability. At the single cell level, we found that many MEC neurons exhibited highly periodic spatial firing patterns along structured flights - consistent with the periodic nature of grid-cells found in rodents. We further found that these grid cells were organized into modules with similar periodic spacing, reminiscent of the canonical organization of grid cells found in rodents. Analysis of co-modular grid cell population activity revealed topological signatures consistent with that of a 2D torus, in line with experimental findings and theoretical models from rodents. Together these findings suggest that grid cells in bats provide a spatially periodic representation of self-position during flight, and that the toroidal organization of grid cell population activity may generalize across species, including those specialized for three-dimensional navigation. Importantly, these findings allow us to begin addressing how might MEC computations (from the single cell to the population level) contribute to complex 3D navigation.

Disclosures: K.K. Qi: None. M.M. Yartsev: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

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Topic: I.09. Spatial Navigation

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Title: Sequential predictive learning is a unifying theory for hippocampal representation and replay

Authors: *D. LEVENSTEIN¹, A. EFREMOV², A. HALAWA⁵, R. HENHA EYONO², J. CORNFORD⁶, B. A. RICHARDS³, A. PEYRACHE⁴;

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Abstract: The hippocampus is widely known for spatially-selective responses in individual cells. However, many of its cells do not have easily-interpretable spatial tuning curves, and can be selective for other task-relevant variables. To better understand the computations played by this structure, it may be necessary to look to population-level features of neural activity and the distributed computations they enable. Here, we focus on two such features: 1) a continuous neural manifold that maps the spatial environment and non-spatial task structure, and 2) a sparse orthogonalized representation of space that "sweeps" ahead of the animal as it moves through the environment. While recent results indicate that learning to predict sensory inputs produces spatially tuned cells in artificial networks, there are multiple approaches to predictive learning and it remains unknown which, if any, can account for a continuous manifold, tuning to non-spatial task features, or sweeping representations.

In this work, we train a recurrent neural network to predict egocentric sensory input while navigating a virtual environment. We find that predicting sequences of input produces a population-level manifold with spatially-tuned cells, and representational dynamics that sweep ahead of the agent during movement. Like the hippocampus, this network maps non-spatial task features, and when imbued with sparse-lognormal connectivity like seen in hippocampal CA3, develops a sparse-orthogonalized representation of space. Further, we find that the emergence of a continuous manifold is critical for the network's ability to produce simulated "replays" in the absence of input, which, like the hippocampus, have a range of similarity to experienced trajectories in the environment. These simulations can be directed by a fictitious action signal, suggesting a means by which upstream regions can use the hippocampal map for learning effective behavioral strategies or planning. Together, these results suggest that sequential predictive learning underlies hippocampal representation and replay, and that representational sweeps reflect a data-efficient algorithm for sequential predictive learning in the brain.

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Poster

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(<https://doi.org/10.69777/332942>)

Title: Excitatory-inhibitory coordination of the thalamic head direction signal

Authors: *H. DENNY¹, A. PEYRACHE²;

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Abstract: Neurons in the thalamic head direction (HD) system, the anterodorsal Nucleus (ADn), fire for a specific orientation in the horizontal plane, acting as the animal's compass. Remarkably, this signal is very stable; correlations between neurons are maintained during sleep despite the deprivation of sensory information. Indeed, a "virtual" HD not only persists during sleep, but rapidly rotates around the compass. The stability of the HD signal suggests the HD system is an attractor network, and models suggest the network requires 2 properties to function; recurrent connections, and global inhibition. We hypothesize these properties are provided by the Thalamic Reticular Nucleus (TRn). The TRn is the inhibitory nucleus of the Thalamus is reciprocally connected to the ADn. As such, we hypothesize the reciprocal connections between TRn and ADn are the primary source of the thalamic HD attractor. In this work, we first use an 8-shank silicon probe in the thalamus and linear probe in the hippocampus to simultaneously record multi-unit activity from the ADn, TRn, and CA1. Animals were allowed to explore a square arena for 20 minutes, before being returned to the home cage to sleep for 2 hours. We first examined whether neurons in the TRn carried any HD information and found that HD tuning was heterogenous. We then used a Poisson Generalized Linear Model to extract pairwise coupling filters. Coupling strength between TRn and ADn neurons, was positively associated with the similarity of their head direction tuning curves. This relationship was modest but significant in both directions, from TRn to ADn and ADn to TRn. To causally test whether these functional connections support ADn attractor dynamics, we expressed ArchT-GFP in the TRn of VGAT-CRE mice, then implanted an optical fiber and silicon probe to the ADn. Preliminary results suggest that during optogenetic silencing of TRn terminals in the ADn, correlations between HD neuron pairs deteriorate, suggesting the attractor is disrupted. These findings shed light on how the TRn coordinates the HD attractor in the ADn.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

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Topic: I.09. Spatial Navigation

Support: Canadian Institutes of Health Research Project Grants (190289 and 180330)
National Sciences and Engineering Research Council Discovery Grant (RGPIN-2018-04600)

Title: Ultra-fast oscillations in the head-direction system coordinate with dentate spikes

Authors: *G. VIEJO¹, S. SKROMNE CARRASCO², H. DENNY³, A. PEYRACHE²;
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Abstract: The head-direction (HD) system is a critical component of the brain's spatial navigation network and is thought to provide internally generated directional signals during sleep that support memory consolidation in parahippocampal regions. Within the Papez circuit, the HD signal is shaped by a mammillary-thalamic-subicular loop, with the lateral mammillary nucleus (LMN) receiving angular velocity input from the brainstem and hypothesized to operate as an attractor network. By recording neuronal ensembles in the LMN of freely moving mice during wakefulness and sleep, we identified a previously uncharacterized fast population event: brief (<20 ms) high-synchrony bursts that we termed Ultra Fast Oscillation (UFO). These events occurred predominantly during sleep at a rate of ~1 Hz and reliably recruited spiking in the LMN, as well as in downstream thalamic and subicular stages of the HD circuit. During non-rapid eye movement (NREM) sleep, UFOs were positively coupled with dentate spikes (DS) and negatively coupled with hippocampal sharp-wave ripples (SWRs), indicating that UFOs and SWRs/DSs are orchestrated by opposing neuromodulatory states. Notably, UFOs were also observed during wakefulness, typically preceding head rotations, suggesting a predictive role in movement initiation. During sleep, short auditory tones reliably triggered UFOs, indicating that this circuit remains sensory-responsive and selectively excitable even in offline states. These findings reveal a novel form of structured population activity in the HD system with potential implications for sleep-dependent memory processing.

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Poster

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Topic: I.09. Spatial Navigation

Support: Canadian Institutes of Health Research Project Grants 190289 and 180330
National Sciences and Engineering Research Council Discovery Grant
RGPIN-2018-04600

Title: Thalamic head-direction dynamics persist during sleep independent of upstream input

Authors: *A. PEYRACHE¹, S. SKROMNE CARRASCO², G. VIEJO³;
²Montreal Neurolog. Inst., ¹McGill Univ., Montreal, QC, Canada; ³Flatiron institute, Flatiron
institute, New York, NY

Abstract: Continuous attractor networks are thought to underlie diverse cognitive functions, from working memory to spatial navigation. However, the neuronal dynamics and circuits supporting these dynamics *in vivo* remain poorly understood. A well-characterized example is the head-direction (HD) system, which encodes the orientation of the animal's head and provides a crucial signal for navigation. This signal is relayed to the cortex by the anterodorsal nucleus (ADn) of the thalamus, where the vast majority of neurons are tuned to HD. Notably, ADn HD cells maintain their mutual coordination during sleep, when sensory inputs are largely absent, consistent with dynamics governed by an internal attractor. The rigid organization of HD cell activity in the ADn raises the question of the origin of these structured patterns. The lateral mammillary nucleus (LMN), an upstream structure, has been proposed as a key generator of the HD signal. To test this, we recorded LMN neuronal ensembles across wakefulness and sleep, and compared their organization to that of simultaneously recorded ADn cells. We found that LMN ensemble activity exhibits two distinct regimes. During REM sleep, when global brain dynamics closely resemble those of wakefulness, LMN HD cells were coordinated similarly to wake, mirroring ADn activity. In contrast, during non-REM sleep, LMN HD cell coordination was markedly reduced, while ADn neurons maintained the same degree of mutual coherence as during REM and wake. The breakdown in LMN coordination was linked to a switch toward hypersynchronous firing, where neurons co-fired regardless of their preferred direction. Optogenetic inhibition of the post-subiculum resulted in a complete disruption of LMN HD cell coherence during NREM, but not ADn HD cell coherence, suggesting that cortical feedback entirely drives pairwise coupling in the LMN during this phase of sleep. These results suggest that the HD signal in the ADn operates as a self-sustained attractor, capable of maintaining coherent dynamics independently of its LMN inputs.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

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Topic: I.09. Spatial Navigation

Support: National Science and Technology Innovation 2030 Major Program
2022ZD0205000
Strategic Priority Research Program of the Chinese Academy of Sciences
XDB1010201
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Title: Hierarchical chunking and abstraction of complex events in the primate brain

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Abstract: Humans and animals have an impressive ability to extract knowledge structures, called schemas, and specific details from complex real-world events. This skill helps them apply what they learn to different settings and make sense of situations based on the context. We investigated how two brain areas in macaque monkeys—the hippocampus (HPC) and the orbitofrontal cortex (OFC)—team up to break down events during challenging, lifelike tasks. To study this, we designed a unique task where the monkeys navigated in the combination of two kinds of spaces: a physical one in an open area and an abstract one based on the order of certain cues. Using chronically implanted microdrives, we recorded neuronal activity in HPC and OFC while monitoring the monkeys' head and gaze stances. The monkeys demonstrated robust behavioral flexibility, achieving a success rate above 75% across events. We found that neurons in both HPC and OFC hierarchically decomposed each event into subgoals and goals, and further showed abstraction across events through dimensionality reduction, aligning with reinforcement learning (RL) principles. In addition, OFC neurons tended to exhibit stronger abstraction across events, while HPC neurons were more sensitive to event-specific details. These findings reveal how the HPC-OFC in primates encode multidimensional events, offering insights for designing advanced RL algorithms.

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Poster

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Topic: I.09. Spatial Navigation

Support: National Science and Technology Innovation 2030 Major Program
2022ZD0205000
National Natural Science Foundation of China 32371076

Title: Spatial organization of primate hippocampal functions

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Abstract: The hippocampus is a medial temporal lobe structure that is critically involved in episodic memory and spatial navigation. The left and right hippocampi in humans exhibit striking lateralization in some cognitive domains, supported by molecular asymmetries and anatomical connections. Additionally, accumulating evidence indicates that the longitudinal axis of the hippocampus is organized along a gradient in spatial representations and episodic memory processing. However, the precise mechanisms mediating the integration of episodic memory components through uniform neuronal ensembles along the longitudinal axis of bilateral hippocampi in primates remain elusive. To address this question, we trained two macaque monkeys on a multi-task paradigm designed to examine neural encoding of item, spatial, and temporal aspects of episodic memory, as well as their associations. We recorded single-neuron activity along the entire long axis of both hippocampi using chronically implanted electrodes. We found striking differences in neural representations between the left and right hippocampi. The right hippocampus exhibited significantly stronger representations for association memory and spatial path encoding compared to the left, while the left tended to be more involved in outcome monitoring. We also identified important functional differences along the anterior-posterior axis: neural codes for item memory became increasingly more prevalent towards the anterior hippocampus. These findings suggest that, in non-human primates, the left and right hippocampi process qualitatively different information for remembering episodic events in space. Furthermore, spatial organization across axes optimizes hippocampal function through complementary hemispheric strategies and hierarchical integration along the longitudinal axis.

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Poster

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Support: National Science and Technology Innovation 2030 Major Program
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XDB1010201

National Natural Science Foundation of China 32371076

Title: Primate view representation arises from successive stages of reference frame transformations

Authors: Y. HUANG, A. LIU, X. XU, K. DU, *D. MAO;
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Abstract: We constantly align self-centered sensory inputs (egocentric) with world-centered spatial map (allocentric) to locate and orient ourselves while navigating. The hippocampus (HPC), retrosplenial cortex (RSC), and posterior parietal cortex (PPC) are key regions implicated in processing spatial information from different reference frames. Primates have greatly developed visual capabilities. The primate hippocampus forms a representation of visual space of the external world. What role the PPC-RSC-HPC pathway plays in the arising of such a world-centered visuospatial representation is unclear. Leveraging wireless neural recording, motion capture, and wireless eye tracking, we examined spatial coding transformations along the PPC-RSC-HPC pathway while three macaque monkeys freely explored in an open arena. We wirelessly recorded single neurons and local field potentials (LFPs) across HPC, RSC, and PPC. We recorded in total 1261 well-isolated single units (HPC, 813 neurons; RSC, 284 neurons; PPC, 164 neurons). The PPC-RSC-HPC pathway showed opposing gradients for egocentric and allocentric spatial coding, with PPC preferring the former and HPC preferring the latter, respectively. As an intermediate hub, the retrosplenial cortex showed the strongest interactions between these two reference frames, using gaze and head directions to transform parietal egocentric into allocentric view representations in the hippocampus. Moreover, RSC exhibited preferred phase-locking to distinct frequency bands of HPC and PPC LFPs: more neurons were locked to HPC theta while this was true for PPC beta. These findings suggest that the monkey PPC-RSC-HPC pathway gradually translates between egocentric and allocentric spatial representations, with information from both reference frames converging in RSC, likely mediated by directional tuning. These results may underlie the computational principles that enable the primate brain to construct a (allocentric) world model from (egocentric) first-person experience.

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Poster

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Title: Septal glutamatergic neurons contribute to speed and spatial coding in the medial entorhinal cortex

Authors: *J. ROBINSON¹, J. E. CARMICHAEL³, M. P. BRANDON⁴, M. E. HASSELMO²;
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Abstract: The medial entorhinal cortex (MEC) is integral to spatial navigation and memory processing. This region contains several cell types that together represent different spatial aspects of the environment, including: 1) grid cells, 2) boundary vector cells, and 3) head direction cells. Previous work has shown that pharmacological inactivation of the medial septum (MS) disrupts both the spatial periodicity of grid cells and speed coding in the MEC. The MS comprises three distinct populations: cholinergic, GABAergic, and glutamatergic neurons. Glutamatergic neurons in the MS have been linked to locomotor behaviors, with running speed correlating with terminal activation in the MEC. Here, we evaluate the contribution of septal glutamatergic neurons to grid cells and speed coding in the MEC. Specifically, we selectively silenced glutamatergic neurons in the MS using optogenetic activation of Archaelrhodopsin while recording from neurons in the MEC. A stimulation protocol of 30-second inactivation followed by a 30-second recovery period was applied throughout the recording session. Our results show that silencing septal glutamatergic neurons does not affect MEC theta power or frequency. However, inactivation of this population distorts central grid fields and modulates the velocity coding. Together, these findings highlight the importance of medial septal input in spatial and speed coding in the entorhinal cortex.

Disclosures: J. Robinson: None. J.E. Carmichael: None. M.P. Brandon: None. M.E. Hasselmo: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.12/RR7

Topic: I.09. Spatial Navigation

Support: NIH R01 MH120073

Title: Internal Representations Underlying Egocentric Boundary Cell Responses in Retrosplenial Cortex

Authors: *J. FANG^{1,2}, P. A. LACHANCE², M. E. HASSELMO²;
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Abstract: Egocentric boundary cells (EBCs) fire when environmental barriers occupy specific positions in egocentric coordinates relative to an animal's current position and direction. Understanding the internal representation that underlies these responses is critical for comprehending spatial navigation and cognition. In this study, we compared several simplified computational models to characterize the receptive fields of EBCs, including (but not limited to) models based on a Gaussian function of egocentric angle alone, a 2D Gaussian function of egocentric cartesian coordinates, and a 2D Gaussian function of radial distance combined with angle (angle-distance). Models were fitted to electrophysiological recordings from 129 retrosplenial cortex (RSC) neurons identified as EBCs, recorded from freely moving rats exploring an arena. For each video frame, boundary points perceived by the animal were simulated using ray-tracing. Neural firing probabilities in the model were calculated by summing receptive-field values over a subset of boundary points and then passing this sum through a sigmoid function. Model parameters were optimized via maximum-likelihood estimation. Model comparisons using Akaike Information Criterion (AIC) and Bayesian Information Criterion (BIC) indicated that the angle-distance (radial) model consistently achieved the lowest median AIC and BIC values and provided the best fit for over half of the neurons, suggesting that EBCs might encode a combined internal representation of angle and radial distance from boundaries. Interestingly, we found that even a model based solely on egocentric angle had competitive AIC and BIC values, demonstrating that egocentric angle alone is a major determinant of EBC firing patterns. All tested models—except the purely distance-based model—significantly outperformed the baseline model using each cell's mean firing rate (permutation test, $p < 0.001$). A model incorporating only the nearest boundary point captured substantial variance and extending it by summing over the closer half of additional boundary points further improved the model fit.

Disclosures: J. Fang: None. P.A. LaChance: None. M.E. Hasselmo: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.13/RR8

Topic: I.09. Spatial Navigation

Support: NIH Grant MH120073
NIH Grant MH139270

Title: Influence of the height and tilt of environmental boundaries on cortical egocentric spatial representations

Authors: *P. A. LACHANCE, E. GROSS, M. E. HASSELMO;
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Abstract: The navigational affordances of an environment are restricted by the placement of noncircumventable boundaries, such as the walls that define a room. Neurons in the rat postrhinal (POR) and retrosplenial (RSC) cortices have been demonstrated to fire in response to the bearing and/or distance of environmental boundaries in an egocentric reference frame, often referred to as egocentric boundary or egocentric bearing cells (collectively, EB cells). However, the physical properties of environmental boundaries that drive EB cell firing in these regions are not fully understood. To expand our understanding of these properties, we performed tetrode recordings from EB cells in the POR and RSC of rats during free foraging in an open field environment (120 x 120cm with 60cm high opaque walls) while parametrically manipulating the height or tilt of environmental boundaries. In one manipulation, part of the arena floor was tilted upward in increments of 30 degrees until it became a fully vertical wall (floor -> 30deg ramp -> 60deg ramp -> vertical wall). In another, the arena floor was lifted upward relative to the surrounding walls to decrease their apparent height (60cm walls -> 30cm walls -> 4cm 'lip'). In both tilt and height manipulations, EB cells in both POR and RSC maintained their egocentric bearing and distance preferences but demonstrated a graded change in firing rate along the manipulated walls, such that tall or vertical walls elicited the strongest firing while short or horizontal walls led to weak or absent firing. Computational modeling of EB cell responses based on optic flow or static visual features successfully captured these firing properties and produced important insights into potential distinctions between POR and RSC EB representations. Overall, this study constitutes a critical step toward understanding the sensory features that drive egocentric spatial representations in POR and RSC, as well as the mechanisms by which these spatial signals may be used to guide navigation in environments with different boundary conditions.

Disclosures: P.A. LaChance: None. E. Gross: None. M.E. Hasselmo: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.14/RR9

Topic: I.09. Spatial Navigation

Support: Howard Hughes Medical Institute

Title: Connectome-constrained RNNs learn modular implementations supporting attractor dynamics and angular path integration

Authors: *B. HULSE¹, A. PREMA BALAKRISHNAN², S. ROMANI², V. JAYARAMAN¹, A. M. HERMUNDSTAD¹;

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Abstract: Recurrent neural networks (RNNs) trained to perform cognitive tasks can learn dynamical solutions that resemble those found in biological neural networks, suggesting that

RNNs can be used to generate hypotheses for how specific computations are implemented in animal brains. Yet it remains unclear how these dynamics arise from the underlying network connectivity and, in the absence of ground-truth connectomic data, whether task-trained RNNs learn structural implementations that resemble those of their biological counterparts. We address these questions through the lens of attractor dynamics that support angular path integration in the *Drosophila* head direction system, a modular recurrent network where a complete connectomic wiring diagram has recently become available. By comparing three classes of RNNs, ranging from fully connectome constrained to fully unconstrained, we characterize a family of solutions with distinct implementations at both the dynamical and structural levels. While connectivity-constrained networks converge towards known solutions — including previously described hand-crafted theoretical models — unconstrained networks learn novel, non-modular solutions characterized by considerable variability in single-unit tuning curves and network connectivity. These results suggest that connectomic information provides an important constraint on network dynamics and that unconstrained RNNs are unlikely to converge towards modular implementations on their own. While the specific implementations found in modular and non-modular RNNs differ, both rely on the same core mechanism to implement their dynamics — balanced, phase-shifted connectivity. Overall, we find that unconstrained RNNs can, with an appropriate analytical framework, be used to generate hypotheses for key structural and functional features required to implement specific computations.

Disclosures: B. Hulse: None. V. Jayaraman: None. A.M. Hermundstad: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.15/RR10

Topic: I.09. Spatial Navigation

Title: Developmental maturation of ring attractor dynamics in the *Drosophila* central complex

Authors: *A. HAMID¹, A. STUART^{1,2}, C. CHRISTOFOROU¹, G. IHRKE¹, M. SYED^{3,1}, B. HULSE¹, V. JAYARAMAN¹;

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Abstract: In *Drosophila*, head direction (HD) is encoded by compass neurons that generate a bump-like population activity pattern in a highly conserved insect brain region called the central complex. This bump of neural activity is thought to be maintained through ring attractor dynamics, allowing the neurons to represent a stable and unique HD over time. In contrast to most theorized ring attractor networks, which require large numbers of neurons to ensure a continuous HD representation, the *Drosophila* compass system is impressively continuous with a small number of computational units. The process by which this compact recurrent ring attractor network develops its final structure and dynamical properties remains unclear. We adopted a

developmental approach to explore how the key neural components of the *Drosophila* compass circuit develop and how the network dynamics evolve to form a functional ring attractor system in adult flies. Morphological analysis of individual cell types within the head direction system show that certain components of the network develop later during the pupal stages. By imaging calcium activity in compass neurons during pupal stages, we discovered that flickers of population activity emerge in these neurons as early as the mid-pupal stages. This flickering activity later transitions into transient bump-like activity, which eventually matures into more consistently stable bump activity, characteristic of adults, in the late pupal stages. In addition to localized bump-like activity, we also observed bursts of large-scale compass neuron activity in the pupal stages. We are now exploring whether spontaneous activity in the pupal stages tunes the dynamics of the continuous ring attractor network in adults. Our goal is to use our understanding of the developmental maturation of the circuit to guide our thinking about the mechanistic basis of ring attractor dynamics in this small but impressively powerful navigational circuit.

Disclosures: A. Hamid: None. A. Stuart: None. C. Christoforou: None. G. Ihrke: None. M. Syed: None. B. Hulse: None. V. Jayaraman: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.16/RR11

Topic: I.09. Spatial Navigation

Title: Estimating and using self-motion for navigation

Authors: *A. STANOE¹, B. HULSE², H. J. HABERKERN³, S. ROMANI¹, V. JAYARAMAN⁴;

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Abstract: Internal representations of an animal's relationship to space, such as its head direction and traveling direction, enable flexibility in navigation across species. For example, they allow an animal to retain its bearings even in the transient absence or unreliability of localizing sensory cues, such as landmarks. A requirement for this adaptability is that these spatial representations be reliably updated by integrating estimates of self-motion. In most animals, these self-motion estimates are constructed from multiple sources of information, including motor commands (efference copies), visual optic flow, and proprioception. In *Drosophila melanogaster* and other insects these processes converge and combine in a highly conserved brain region called the central complex. Recent studies have elucidated some of the core circuits underlying the integration dynamics, however, an end-to-end characterization of these signals in relation to their functional role is still missing. We are using two-photon calcium imaging to characterize different self-motion inputs to the central complex and are analyzing their upstream networks

using the connectome. To better link self-motion estimates to the fly's behavior, we are quantifying the fly's navigational performance during two- and three-dimensional variants of local search, a stereotypical walking behavior exhibited by many species upon encountering a sugar-associated reward. This combination of quantitative navigational behavior and physiology in the fly's central complex will allow us to firmly connect the computation of self-motion to its use in behavior, a core need for flexible navigation in many animals.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.17/RR12

Topic: I.09. Spatial Navigation

Title: A rotational velocity estimate constructed through visuomotor competition updates the fly's neural compass

Authors: *C. SAUVOLA¹, B. HULSE¹, A. STANOEV¹, D. TURNER-EVANS², J. D. SEELIG³, H. J. HABERKERN⁴, A. M. HERMUNDSTAD¹, V. JAYARAMAN¹;

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Abstract: Navigating animals continuously integrate velocity signals to update internal representations of their directional heading and spatial location in the environment. How neural circuits combine sensory and motor information to construct these velocity estimates and how these self-motion signals, in turn, update internal representations that support navigational computations are not well understood. Recent work in *Drosophila* has identified a neural circuit that performs angular path integration to compute the fly's head direction, but the nature of the velocity signal is unknown. Here we identify a pair of neurons necessary for angular path integration that encode the fly's rotational velocity with high accuracy using both visual optic flow and motor information. The relative contributions of visual and motor signals to the rotational velocity estimate are not fixed. Rather, these signals are integrated in a flexible manner depending on context. In one-dimensional rotational environments with all visual features effectively at infinity, these neurons prioritize motor information over visual information when these signals align directionally but differ in magnitude. When the two signals are experimentally put in conflict by inverting the closed-loop gain, reciprocal inhibition selects either the motor or visual signal at the expense of the other. In cluttered two-dimensional virtual environments with objects at different distances from the fly, such as tufts of grass in a natural scene, translational movements can also generate locally rotational optic flow. In such confusing conditions, the fly's rotational velocity neurons continue to favor motor signals over those generated by visual optic

flow. However, in these more complex environments, we observe that the contribution of visual optic flow to the rotational velocity estimate often changes with accumulated navigational experience. Together, our results suggest that flies update their head direction representation by constructing an estimate of rotational velocity that relies primarily on motor information with optic flow signals being incorporated in a context- and experience-dependent manner.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.18/RR13

Topic: I.09. Spatial Navigation

Title: Spatiotemporal filtering of visual information for head direction estimation

Authors: *S. CHITNIS, M. NOORMAN, J.-Y. PARK, H. J. HABERKERN, A. M. HERMUNDSTAD, V. JAYARAMAN;
HHMI Janelia Res. Campus, Ashburn, VA

Abstract: Many animals, including insects, maintain internal representations of their direction to help them navigate more flexibly in their surroundings. These neural representations are estimated and continuously updated from available sensory and motor information. In *Drosophila*, compass neurons (EPGs) in a highly conserved insect brain region called the central complex carry a head direction (HD) representation, which is updated by self-motion as well as multisensory landmark information. The EPG population appears to maintain a stable global HD estimate in cluttered naturalistic virtual environments with celestial cues, like intensity gradients in the sky of the sort that might be generated by the sun (Haberkern et al., 2022). We aim to understand how visual processing upstream of the fly's HD circuits contributes to the stability of the fly's HD system. Visual information about landmarks is communicated to the HD system via multiple classes of visual ring neurons (ERs). We first focused on characterizing the spatiotemporal receptive fields of elevation-insensitive, azimuthally tuned ER4d neurons using white noise receptive field mapping. Consistent with previous reports (Sun et al., 2017), we observed biphasic temporal responses and contralateral inhibition in the dendritic calcium responses of these neurons. We are now testing specific hypotheses for how these properties might be combined with additional circuit processing to allow the HD system to reliably tether to global visual landmarks amidst local clutter. In our poster, we will report on the results of this work, which combines connectomics, compartment-specific calcium imaging, and whole-cell patch clamp electrophysiology in ring neurons with computational modeling.

Disclosures: S. Chitnis: None. M. Noorman: None. J. Park: None. H.J. Haberkern: None. A.M. Hermundstad: None. V. Jayaraman: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

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Program #/Poster #: PSTR305.19/RR14

Topic: I.09. Spatial Navigation

Support: IITP-2025-RS-2024-00436857
RS-2024-00341894

Title: Embedding of egocentric spatial representations into memory engrams in the retrosplenial cortex

Authors: *Y. YEO¹, J. KWAG²;

¹Brain and Cognitive Sci., ²Dept. of Brain and Cognitive Sci., Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: The retrosplenial cortex (RSC) is critically involved in the consolidation and retrieval of contextual memories. Recent studies have identified egocentric boundary and vertex vector cells in the RSC that encode the environmental geometry in egocentric spatial reference frames. However, how such egocentric spatial codes contribute to memory processes remains unclear. To address this, we performed longitudinal *in vivo* calcium imaging of RSC excitatory neurons in C57BL/6J mice engaged in a contextual fear memory task comprising habituation, conditioning, and retrieval sessions. We found that a subset of egocentrically tuned neurons that were activated by aversive foot shocks during conditioning were selectively reactivated during memory retrieval. These findings indicate that egocentric spatial representations are embedded in retrosplenial memory engrams and play a role in retrieving learned contextual information. Thus, egocentric coding in the RSC supports spatial navigation but also the encoding and retrieval of contextual memory.

Disclosures: Y. Yeo: None. J. Kwag: None.

Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR305.20/RR15

Topic: I.09. Spatial Navigation

Support: VolkswagenStiftung Grant CLAM 9C854

Title: Parallax and rapid visual reorientation in the head-direction system

Authors: S. KRAUß^{1,2}, E. NEFTCI^{1,2}, F. T. SOMMER³, *A. RENNER¹;

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Abstract: Spatial navigation relies on head-direction (HD) cells, which function as neural compass. Located in postsubiculum and thalamus, HD cells integrate angular head velocity (AHV) from vestibular, proprioceptive, and optic flow inputs while recalibrating through visual landmarks to prevent drift accumulation. However, fundamental questions remain about how the HD system processes visual cues, particularly whether it compensates for parallax effects, apparent positional shifts of proximal objects when viewed from different angles.

We investigated HD visual processing using postsubicular recordings from mice navigating with a single visual cue on a circular platform. We discovered systematic parallax bias in decoded HD, indicating the system misinterprets cue position based on viewing angle. Observed errors were smaller than predicted by parallax alone, explainable by AHV integration combined with simple visual anchoring at different angles to the cue. This, and integration of multiple cues, may explain why there is, to our knowledge, no prior evidence of parallax in the HD system.

Interestingly, each animal exhibited a distinct anchoring angle to the cue, which can be calculated from the parallax. This may allow investigating the stability and learning of associations between retrosplenial bearing cells and postsubicular HD cells.

Computational modeling confirmed that integration across multiple locations and with multiple cues, without explicit parallax correction, leads to similarly small parallax errors, depending on the weighting of visual vs. movement information. To estimate the weighting (i.e., speed) of visual reorientation, we examined reorientation dynamics when cues switched positions. We found that reorientation occurred rapidly (with $\tau < 3s$), but only when cues became visible to the mouse, not immediately when cues switched positions, resolving an apparent contradiction in the literature.

Both parallax error strength and reorientation speed allow us to separately estimate the weighting of visual landmark cues vs. AHV integration, yielding consistent results. These findings suggest that the HD system prioritizes computational efficiency over accuracy, combining rapid and simple visual anchoring with AHV integration to manage parallax- and integration-induced errors rather than performing complex explicit correction. Our results illuminate fundamental properties of biological navigation systems and highlight trade-offs between computational efficiency and positional accuracy, with implications for neuroscientific understanding and bio-inspired artificial navigation systems.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

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Topic: I.09. Spatial Navigation

Support: Emergence en recherche, IdEx 2019-20
FLAG-ERA JTC 2021- HBP grant "VIPattract" (ANR-21-HBPR-0002-02)

Title: Cell type dynamics of the head direction system in the presubiculum of freely behaving mice.

Authors: *E. BELHADEF¹, O. GATEAU¹, E. Z. TÓTH², D. LIM¹, L. WITTNER², D. FRICKER¹, M. GRAUPNER¹;

¹Univ. Paris Cité, CNRS, Saints-Pères Paris Inst. for the Neurosciences, Paris Cedex 06, France;

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Abstract: Navigation is essential for survival, enabling animals to locate food, find shelter and avoid threats. This process is supported by specialized circuits such as the head-direction (HD) system, which act as an internal compass through the collective activity of neurons, each tuned to a specific head orientation. While the HD signal originates from vestibular inputs, it is stabilized by sensory cues such as visual landmarks. HD cells can maintain their tuning in the absence of visual inputs and flexibly remap to environmental changes. This balance between stability and flexibility is thought to arise from an interplay between excitatory neurons and inhibitory interneurons, yet the microcircuit mechanisms underlying these interactions is unknown. To reveal circuit interactions, we focused on the presubiculum, a key region integrating both visual and vestibular signals. We used miniscope calcium imaging in freely moving C57BL/6J mice (n=12 mice), combined with post-hoc cell-type identification (n=9 mice). Mice were recorded in a circular arena with a white visual cue which position could be shifted and a controllable light. Each session consisted of multiple rotations of the visual environment and a period of darkness. Each animal was recorded for at least two weeks (n=156 sessions across animals), with an average of 53 recorded cells per animal (7 -234 cells, mean 53 ± 41 SD), of which 46% were HD cells on average (defined as $R > 0.2$, $p < 0.05$, Rayleigh's test; range :14.6-87.5%, mean 46.2 ± 26 SD). To determine cell identity, we sectioned the imaged brains along the imaging plane, performed immunostaining against GABAergic interneurons makers (parvalbumin, vasointestinal peptide and somatostatine), and reconstructed the tissue in 3D. This approach enabled us to match GCaMP8m expressing neurons imaged in vivo with their molecular identities, allowing us to assess HD tuning in defined cell types.

By combining in vivo imaging and cell type identification this study sheds light on cell type-specific contributions to HD tuning and unravels the presubicular microcircuit dynamics underlying HD signal stability and remapping. These results advance our understanding of how specific cell types contribute to the neural computations that support spatial orientation.

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Poster

PSTR305: Position, Heading, and View Modulated Cells

Location: SDCC Halls B-H

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Program #/Poster #: PSTR305.22/RR17

Topic: I.09. Spatial Navigation

Support: IITP-2025-RS-2024-00436857
RS-2024-00341894

Title: Distinct Contributions of Entorhinal Cortex and Anterior Thalamic Inputs to Egocentric Geometry Representation in the Retrosplenial Cortex

Authors: *D. OH, J. KWAG;
Dept. of Brain and Cognitive Sci., Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: The retrosplenial cortex (RSC) plays a central role in spatial navigation by encoding egocentric information about environmental geometry, such as the positions of boundaries and vertices relative to the animal. It receives converging inputs from the entorhinal cortex (EC), which primarily conveys allocentric spatial signals including grid, border, and head direction information, and from the anterior thalamic nuclei (ATN), which predominantly encode head direction. This anatomical organization positions the RSC as a potential hub for transforming allocentric spatial signals into egocentric reference frames. However, the circuit mechanisms underlying this transformation remain poorly understood. To investigate how EC and ATN inputs contribute to egocentric geometric encoding in the RSC, we performed in vivo calcium imaging of GCaMP-expressing excitatory neurons in the RSC of freely moving mice exploring an open-field arena, while selectively inhibiting either EC or ATN inputs using 590 nm optogenetic inactivation. We found that suppression of EC input preserved the preferred wall and vertex angles of individual egocentric representations but significantly reduced their tuning sharpness, suggesting that EC input enhances the precision of egocentric encoding. In contrast, inhibition of ATN input disrupted the preferred angle representations themselves, indicating that thalamic head direction input is essential for stabilizing egocentric spatial tuning. Together, these findings reveal that the transformation of allocentric spatial signals into egocentric geometric representations in the RSC relies on distinct contributions from cortical and thalamic inputs, providing circuit-level insights into how spatial reference frames are integrated to support navigation

Disclosures: D. Oh: None. J. Kwag: None.

Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.01/RR18

Topic: I.10. Human Learning and Cognition

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Title: Unpacking Bimanual Motor Skill Learning: Behavioural Components and White Matter Correlates in Acute Stroke

Authors: *C. VAN RAVESTYN^{1,2,3}, L. DRICOT¹, N. DELINTE^{1,4}, B. BIHIN⁶, B. HERMAN^{3,5}, B. DE COENE⁷, N. MULQUIN⁷, Y. VANDERMEEREN^{1,2,3};

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Abstract: Introduction: Bimanual motor skill learning (bim-MSkL) is central to most daily activities and may be impaired after stroke. Identifying damaged (sub)cortical brain regions and disrupted white matter tracts (WMT) by an acute stroke may uncover motor and/or cognitive substrates critical for bim-MSkL. **Objectives:** To (1) quantify bim-MSkL in acute stroke patients versus healthy individuals (HI); (2) identify brain lesions associated with bim-MSkL impairment; (3) assess whether disruption of microstructural integrity and/or WMT disconnection relate to impaired bim-MSkL. **Methods:** 90 (sub)acute stroke patients (age:67.7±13.4, 44W/48M) and 62 age-matched HI (age:64.7±18, 33W/28M) trained for three days on a bim-MSkL robotic cooperation task. Standardized motor and cognitive scales were administered. Patients underwent multimodal MRI (3D-T1, diffusion-weighted imaging) for voxel-based lesion symptom mapping (VLSM), microstructural integrity and tractography analyses. **Results:** Age and sex did not differ between groups ($p>0.05$). Bim-MSkL progression over three days, assessed via a speed-accuracy trade-off (biSAT), was impaired in patients compared to HI (-0.52 [a.u.], $p<0.001$), as was bimanual coordination (-0.08 [a.u.], $p<0.001$). Patients improved in error (-0.23 [cm], $p=0.006$) but not speed (-0.0022 [cm/sec], $p=0.98$). Poor learners (<25% biSAT improvement) comprised 43% of patients versus 13.3% of HI. VLSM ($n=83$) linked motor and functional impairments (Fugl-Meyer Upper Extremity (FMA-UE), Arm Motor Ability Test (AMAT)) to lesions in the internal capsule (IC), putamen, thalamus (FMA-UE), and posterior limb of the IC (PLIC) and superior corona radiata (AMAT). Bimanual coordination deficits in poor learners were associated with PLIC lesions. Tract-based analyses (Diffusion Tensor Imaging, Neurite Orientation Dispersion and Density Imaging, $n=70$) showed reduced microstructural integrity in the corticospinal tract, corpus callosum and superior longitudinal fasciculus in patients scanned ≥ 1 -week post-stroke. Ongoing analyses will examine: (1) relationship between the variation of microstructural integrity along the tracts and stroke injury; (2) whole-brain analyses to quantify the inter-hemispheric asymmetry of WMT integrity induced by an acute stroke; and (3) correlations between bim-MSkL impairment and WMT-

specific damage and asymmetry. **Conclusion:** Correlating acute bim-MSkL impairments with key brain structures and disrupted WMT refines the knowledge about the network supporting bim-MSkL networks and may help developing biomarkers identifying poor learners and leading to personalized rehabilitation.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.02/SS1

Topic: I.10. Human Learning and Cognition

Title: Premotor mu rhythm waveform shape dynamics reflect motor sequence learning

Authors: *T. SURESH¹, B. MILLS¹, S. HUSSAIN²;

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Abstract: Human motor sequence learning recruits a distributed cortical network. Within this network, the premotor cortex (PMC) encodes sequence information. Like the sensorimotor cortex, neuronal populations in PMC exhibit oscillatory activity that is spectrally dominant in the mu (8-13 Hz) range. While previous research has established the role of sensorimotor mu rhythm phase in human corticospinal output, corticospinal plasticity and motor learning, the role of PMC mu rhythm phase in motor learning remains unexplored. Here, we evaluated the effect of motor sequence learning on the non-sinusoidal characteristics of the PMC mu rhythm using cycle-by-cycle waveform shape features typically overlooked by conventional Fourier-based methods. Given that PMC encodes motor sequence information and exhibits strong mu rhythms, we predicted that motor sequence learning would alter PMC mu peak-trough asymmetry. Healthy neurotypical adults (N=18 per group) were randomly assigned to either a SEQ or NO-SEQ group. Participants in the SEQ group practiced a serial reaction time task (SRTT), which contained an embedded, repeating 12-item sequence. Participants in the NO-SEQ group practiced a control version of the SRTT that contained no such sequence. Participants in each group also performed a shorter version of the SRTT one hour after initial practice to evaluate retention. Resting eyes-open EEG was recorded at baseline, immediately after, and 30 minutes after serial reaction time task (SRTT) acquisition. Preliminary analysis (N=5 per group) of resting EEG recordings over the PMC (Hjorth-transformed FC3 electrode) revealed sequence-specific changes in mu waveform shape. Specifically, participants in the SEQ group showed greater reductions in mu peak durations, greater increases in mu trough durations, and greater increases in mu peak-trough asymmetry than participants in the NO-SEQ group immediately after SRTT acquisition. Further, changes in mu peak-trough asymmetry from baseline to immediately after SRTT acquisition positively correlated with both acquisition and retention

skill in the SEQ but not the NO-SEQ group. These results were specific to PMC mu rhythms, as similar effects were not observed in EEG recordings over posterior parietal cortex (Hjorth-transformed Pz electrode) or for beta (14-33 Hz) waveform shape. Together, these early results suggest that motor sequence learning transiently shifts PMC mu peak-trough asymmetry, resulting in sharper mu peak and broader mu trough phases. Findings suggest that non-sinusoidal features of PMC mu oscillations reflect sequence-specific plasticity processes underlying motor learning.

Disclosures: T. Suresh: None. B. Mills: None. S. Hussain: None.

Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.03/SS2

Topic: I.10. Human Learning and Cognition

Support: NIH Grant 1R01NS132926-01

Title: Neural Coding of Movement and Abstract Internal Models across the Sensorimotor Hierarchy

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Abstract: Internal models in the motor system enable effective motor control by relating movement commands to their expected sensory consequences. However, it is not well understood how these models are represented neurally, independent from lower-level features of movement. Here, we developed an fMRI reaching task where participants made the same movements to reach the same sensory goal (target), but under differently structured sensory perturbations (visuomotor rotation versus mirror reflection). This required the participants to plan their movements using distinct internal models, despite the lower-level features (e.g. target location and optimal reach direction) being identical on many trials.

Participants (N = 28) moved a joystick to control a cursor to make a center-out reach toward one of four possible targets on the screen. Before each block of trials, participants were informed about what perturbation they will experience (visuomotor rotation, where the cursor rotated clockwise away from the hand, or mirror reversal, where the cursor reflected off a vertical mirror axis), both of which they had been pre-trained on. Critically, for two of the four possible goal locations, the optimal hand reach angle and endpoint visual feedback were identical under both rotation or reflection perturbations. Before executing each reach, participants saw the target location for a jittered interval (2-4s) and were instructed not to move (planning phase). After movement execution, visual feedback on the accuracy of the reach was delayed by 1 second after completion to isolate explicit learning processes and limit the contribution of implicit sensorimotor recalibration.

We examined multivariate patterns in the BOLD data across trials, focusing on trials where the lower-level features of movement plans should be identical in order to classify abstract internal model representations (rotation vs. reflection) in the brain. Specifically, we trained classifiers on searchlights across the whole brain to explore where these distinct internal models are reliably distinguished in neural patterns. Preliminary results suggest that superior prefrontal and premotor areas encode abstract internal models for motor planning even when controlling for lower-level movement parameters. Representational similarity analyses further support these results, and point to lower-level motor representations in primary motor cortex. Our preliminary findings thus suggest that “higher” regions in the sensorimotor hierarchy represent more abstract components of motor planning, such as abstract internal models, while lower regions more reliably encode lower-level movement kinematics.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.04/SS3

Topic: I.10. Human Learning and Cognition

Support: NIH Grant R01DC022097
NIH Grant R01DC017439

Title: Auditory and somatosensory cortex causally contribute to human speech motor memory retention

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Abstract: Humans have a complex ability to process speech sounds. When we learn to produce a novel sound, how durable is the newly acquired memory? What are the sensorimotor factors that affect retention and retrieval of this memory? And which brain regions causally contribute to retention? Using adaptation to altered auditory feedback as a model task in healthy adults (18-40 years of age), our first study (n=58) showed that the newly acquired speech motor memory was extensively retained both 8- and 24-hours post learning. The retention was also unaffected by abrupt versus gradual introduction of the altered feedback and showed negligible offline gains. Notably, retrieval of the speech motor memory was observed even in absence of speech error feedback when tested immediately after learning. However, when tested 8- or 24-hours later, retrieval was only observed in presence of the speech error feedback and not when feedback was withheld, indicating a context dependency of speech motor memory. Next, we tested the causal contribution of auditory cortex, via the superior temporal gyrus (STG), primary somatosensory cortex (S1), and primary motor cortex (M1) in speech motor memory retention. In this study, we

used continuous theta burst transcranial magnetic stimulation (cTBS) to disrupt neural activity in STG, S1, or M1 in healthy adults (n=56) immediately following the speech motor learning session. We found that speech motor memory retention was substantially impaired following cTBS to either STG or S1, but not M1. This evidence underscores the causal contribution of auditory and somatosensory but not motor cortical activity to speech memory retention. Taken together, our findings established substantial retention of speech motor memory following a short 20-minute altered auditory feedback task, showed dependency of this memory retrieval on the availability of speech error feedback and determined the causal sensory cortical contribution to speech motor memory. The evidence so far suggests that perceptual changes within the sensory cortical activity are critical for motor learning and memory.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.05/SS4

Topic: I.10. Human Learning and Cognition

Support: JSPS KAKENHI Grant JP23K16535

Title: Investigating impact of resting-state tACS on motor skill learning

Authors: *L. JINYUE¹, H. HAMADA¹, T. KAWASAKI², K. KIKUCHI¹, Q. AN¹, A. YAMASHITA¹;

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Abstract: Transcranial alternating current stimulation (tACS), a non-invasive method of modulating brain oscillatory activity, has garnered increasing attention in neuromodulation. Previous studies have suggested that tACS may enhance cognitive functions and potentially benefit motor skill learning, though effective protocols remain unestablished. In particular, mechanisms underlying its effects of tACS on motor learning remain unclear. Our previous research indicated that increased alpha-band activity in the prefrontal cortex, specifically around the AFz electrode site, during the resting state is positively correlated with motor learning performance. Therefore, the present study aims to investigate whether applying tACS over the AFz region during rest can enhance the rate and retention of subsequent motor skill learning. A single-blind design and a ball-rotation task were used. 8 healthy participants were randomly assigned to a tACS group or a sham group. A high-definition electrode montage was used, with AFz as the central site and four symmetric return electrodes. Participants received 10 minutes of 10 Hz tACS at 1 mA, or sham stimulation, during the resting state. The experimental procedure consisted of a pre-test, stimulation, motor skill learning task, post-test, and a retention test. During the pre-test, participants performed baseline ball-rotation trials in both clockwise (CW) and counterclockwise (CCW) directions. After the stimulation, participants engaged in 25 CW-

direction training trials, each lasting 1 minute. A post-test assessed CW and CCW performance, followed by a retention test 30 minutes later. The task was confirmed to induce motor learning, as both CW and CCW performance significantly improved from pre-test to post-test ($p < 0.01$). To assess tACS effects on the learning process, linear slopes were derived from the 25 training trials and compared between groups. Although no significant group difference was found ($p = 0.32$), a moderate-to-large effect size was observed (Cohen's $d = 0.78$, 95% CI [-0.66, 2.22]), which may indicate a trend toward enhanced learning speed in the tACS condition. Retention performance declined significantly in both tACS and sham groups ($p < 0.05$), with no significant group difference observed. A similar decline was noted in the untrained CCW direction, suggesting the reduction may reflect general fatigue or time-dependent decay rather than a stimulation-specific modulation. These preliminary findings suggest that resting-state alpha tACS may facilitate motor skill learning speed, though further studies with larger samples are warranted to confirm its efficacy.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

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Program #/Poster #: PSTR306.06/SS5

Topic: I.10. Human Learning and Cognition

Title: Phonological networks underlying the visual recognition of playing cards in experts of a traditional Japanese card game

Authors: *M. TANAKA^{1,2}, K. YOSHINAGA², H. TOGO², T. HANAKAWA²;

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Abstract: “Competitive karuta” is a traditional Japanese card game based on the “Ogura Hyakunin Isshu”, a classical anthology of 100 Japanese poems compiled in the 13th century. Competitive karuta players are required to quickly identify the correct karuta cards distributed on the playing field as soon as they hear the beginning of a poem, which requires an expert skill for audio-visual association. To investigate the neural substrates underlying this expert skill, we examined brain activity during a visual recognition task using functional magnetic resonance imaging (fMRI). FMRI data were collected from 16 *karuta* experts and 20 non-experts during a *karuta* card recognition task involving the discrimination of *karuta* cards from “non-*karuta* cards” (traditional Japanese poetry not included in the *Hyakunin Isshu*). Each *karuta* card stimulus contained 14 characters, originally arranged in three columns of five, five, and four characters (the “three-column” original stimulus). We hypothesized that the arrangement of characters on a card would be critical for fast audio-visual recognition. We thus reconstructed *karuta* cards into two columns of seven characters each to serve as a control stimulus (the “two-

column" control stimulus). While the experts showed faster reaction times than the non-experts overall, their reaction times were longer for the two-column stimuli than for the three-column ones, to a greater degree than the non-experts showed (ANOVA, $p < 0.05$). We compared brain activity associated with *karuta* card recognition with that associated with non-*karuta* card recognition. The experts, compared with the non-experts, tended to exhibit greater activation in the left inferior frontal gyrus and the ventral precentral gyrus, and reduced activation in the left middle frontal cortex and the angular gyrus (uncorrected, $p < 0.001$). Additionally, experts exhibited greater activation in the ventral precentral gyrus during the three-column condition compared to the two-column condition (uncorrected, $p < 0.001$). The identified regions constitute the phonological network, part of the language networks. Our findings suggest that phonological processing plays a role in facilitating the rapid recognition of *karuta* cards.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

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Program #/Poster #: PSTR306.07/SS6

Topic: I.10. Human Learning and Cognition

Title: Inferring age from touchscreen movements in a game-based learning task

Authors: *C. ABESHAUS¹, G. AGARWAL², T. QUENDERA³;

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Abstract: We intuitively read others' internal states through their movement. How precisely can this be quantified? Using hexxed, a mobile app-based cognitive task developed by our lab, we examine how the dynamics of on-screen movement relate to age and game outcome. We defined nine movement metrics relating to the temporal and spatial qualities of on-screen taps and swipes. Using game outcome (passing vs. quitting level 1) as a measure of task performance, we find movement metrics to be highly predictive of a player's eventual success and find much of this correlation to be accounted for by the player's reported age. Those who quit the game exhibit movement patterns consistent with older biological age, demonstrating how their movement style may reflect an aging process distinct from chronological time. These findings show how on-screen movement can predict fundamental qualities, such as biological age and performance.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.08/SS7

Topic: I.10. Human Learning and Cognition

Title: Transcranial ultrasound stimulation (TUS) of the hippocampus affects rapid motor memory consolidation

Authors: *M. P. MONTENEGRO¹, S. BAO², Y. LEI¹;

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Abstract: Recent evidence indicates that performance improvements observed during the acquisition of a new motor sequence occur predominantly during quiet rest periods interspersed with practice sessions (micro-offline gains, MOGs). However, it remains uncertain whether these MOGs are primarily attributed to the hippocampus or other cortical regions involved in motor skill learning. This study establishes that the hippocampus plays an essential role in MOGs, and its disruption enhances initial motor performance while expediting stabilization. In this study, young adult participants utilized their non-dominant hand to perform a 5-item explicit sequence (4-1-3-2-4) tapping task designed to assess motor skill learning across acquisition and test phases. Transcranial ultrasound stimulation (TUS) was applied contralaterally to the non-dominant hemisphere hippocampus prior to the acquisition phase to transiently downregulate hippocampal activity. The practice phase of the sequence tapping task comprised 36 alternating 10-second practice and rest intervals, whereas the test phase included 9 trials conducted 24 hours later. Performance metrics included speed, defined as the time interval in seconds between correct keypress transitions, and accuracy, defined as a correct keypress. Micro-online gains were measured as the difference in speed between the last 5 correct transitions of a block and the first 5 correct transitions of that block. Micro-offline gains were defined as the difference in speed between the first 5 correct transitions of a block and the last 5 correct transitions of the previous block. Results indicated differences between the control group and the experimental group across the overall performance curve. In terms of microscale learning, the experimental group demonstrated contributions from both micro-online and micro-offline gains to total learning, whereas the control group's total learning was primarily attributed to micro-offline gains. The findings reveal that hippocampal downregulation significantly reduced MOGs during early learning while leading to higher initial performance levels and a faster acquisition plateau. These results highlight the dynamic role of the hippocampus in mediating both performance and learning stability during motor skill acquisition, providing valuable insights into memory-motor integration.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

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Program #/Poster #: PSTR306.09/SS8

Topic: I.10. Human Learning and Cognition

Support: FRRB 3438840 BOOST
Italian Ministry of Health (Ricerca Corrente 2024-2025,
Scientific Institute, IRCCS E. Medea to AF
5 x mille

Title: Investigating the effects of transcutaneous Vagus Nerve Stimulation on motor cortex excitability and inhibition through paired-pulse Transcranial Magnetic Stimulation

Authors: *S. BOSCAROL^{1,3}, L. TURCHI¹, V. OLDRATI², C. URGESI^{4,1}, A. FINISGUERRA²;

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Abstract: Transcutaneous Vagus Nerve stimulation (tVNS) has been proposed as a prospective treatment for clinical conditions with altered GABAergic transmission. While possible effects of tVNS on behavioral performance in inhibitory control tasks have been previously reported, neurophysiological evidence showing its effects on GABA-mediated inhibition in the motor cortex is limited. Concurrently, the possible influence of participant's gender and state conditions remains unexplored. Here, we applied, single- and paired-pulse TMS to the right or the left primary motor in two different groups of participants. We measured corticospinal excitability (CSE), short and long intracortical inhibition (SICI and LICI), cortical silent period (cSP) and intracortical facilitation (ICF) indexes. The measures were taken, in separated sessions of a within-subject design, at baseline prior to tVNS and after delivering active and sham tVNS in the Cymba conchae of the left ear. To exploit state dependent effects and assess the role of tVNS in motor learning, tVNS was applied, during the execution of a computerized visuomotor task. In the left TMS group, we observed better visuomotor performance during active than sham tVNS, regardless of participant's gender. Interestingly, in both groups, we found a specific increase of SICI, which is mediated by GABA_A activity, after active compared to sham-tVNS and baseline evaluations, which was specifically limited to female participants. No effects on CSE, ICF or GABA_B-mediated intracortical inhibition indexes were observed. The results show specific effects of tVNS on motor learning and GABA_A-mediated motor inhibition, providing supportive evidence for the application of tVNS as an alternative and coadjuvant treatment for disorders featured by altered inhibition mechanisms.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

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Program #/Poster #: PSTR306.10/SS9

Topic: I.10. Human Learning and Cognition

Support: NSF
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Title: Perturbation History Influences Learning in a Novel Redundant Human-Machine Interface

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Abstract: Motor interfaces have decoders that translate high-dimensional motor inputs, such as neural activity, muscle activity, or movement kinematics, into lower-dimensional device outputs. This creates challenges for users learning to control the device, as they must navigate both feedback redundancy (fewer feedback than input dimensions) and potential decoder nonstationarity from sensor drift and recalibration. Training may help users identify effective, robust input patterns for control, but little is known about how to design curricula for motor interfaces. We explored perturbation history as a possible curriculum. We developed a novel reaching task where participants control a 2D cursor via 3D hand movements, allowing us to separate user learning from device limitations while introducing perturbations. The task projects hand position onto a control plane via a transformation $C=MH$, where motions parallel to the plane move the cursor while motions normal to it do not (i.e. are redundant). This structure also allows systematic manipulation of the mapping matrix M to introduce nonstationarity. Unlike in neural interfaces, there are minimal physical constraints on the input space, all inputs and outputs are fully observable, and the task-relevant components of the inputs are clearly defined. Moving along the control plane yields a minimal-effort, maximal-accuracy solution toward which we can try to guide users.

Biomimetic or intuitive decoders are often seen as a goal for motor interfaces (Shenoy & Carmena, 2014). To test the utility of intuitive decoders as part of a curriculum, participants were assigned a projection M that was either “congruent” with the upright display or “incongruent” (flat, unaligned) during Baseline, followed by a shared Perturbation phase. The congruent group showed higher initial task performance, suggesting congruence is a user prior. However, only participants exposed to the incongruent baseline progressively refined their strategies toward the control plane across both phases. Incongruent participants also showed greater variance early and reduced task-irrelevant movement over time, suggesting active exploration and refinement. By the end of learning, their reaches were more aligned with the Perturbation plane. Consistent with recent reports from myoelectric interfaces (Schone et al., 2024), we found that unintuitive decoders misaligned with user priors may promote more generalizable adaptation. These results highlight the need to consider how decoder adaptation can be leveraged as a part of a curriculum to shape user learning in adaptive human-machine interfaces.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

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Program #/Poster #: PSTR306.11/SS10

Topic: I.10. Human Learning and Cognition

Title: Informational Constraint Facilitates Motor Learning by Enhancing Search Efficiency in the Solution Space of a Redundant Two-Joint Throwing Task

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Abstract: The human motor system inherently exhibits high redundancy, allowing multiple solutions to achieve the same task goal. Motor learning thus involves an optimization process through exploration in the high-dimensional solution space. While guidance can facilitate learning by constraining exploration, the effects of different forms of guidance during exploration remain unclear, especially when performing a motor task with redundancy. Here, we examined the learning process while healthy young adults performed a redundant computer-based throwing task that required coordination of the shoulder and elbow joints. Thirty-six healthy right-handed participants (27 males, 9 females; mean age 23.33 ± 3.16 years) were randomly assigned to one of three groups ($n = 12$ per group): physical guidance (PG), verbal instruction (VI), or no instruction (NI). Participants performed four blocks of 60 trials (240 in total) using a semi-virtual throwing task, a modified version of the Skittles task paradigm. Physical guidance was implemented using a KINARM exoskeleton. We hypothesized that the type of guidance would influence motor performance and exploration. Performance and exploration were evaluated by hit rate, performance error, kinematic variability, release parameters, and Tolerance-Noise-Covariation (TNC) cost analysis. We conducted a two-way mixed ANOVA to test group (between-subject) and block (within-subject) effects for each variable. Results showed that the VI group exhibited significant reductions in the Tolerance cost compared to the NI group at the initial stage of practice, indicating more efficient search toward the optimal region of the solution space, along with superior performance outcomes relative to NI. Importantly, no significant differences were observed between PG and VI in most measures during the initial stage, except for variability in release velocity, suggesting that verbal instruction was similarly effective to physical guidance in facilitating early learning efficiency. As expected, the PG group demonstrated more restricted exploration overall, reflected in lower variability of release velocity and reduced T-cost compared to both VI and NI. These findings demonstrate that informational constraints (VI) effectively regulate search during the initial stage of motor learning. Moreover, physical and verbal guidance differentially shape the learner's search process, with both capable of regulating exploration and enhancing task performance in redundant coordination tasks.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

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Program #/Poster #: PSTR306.12/SS11

Topic: I.10. Human Learning and Cognition

Support: NS092079
NS105839

Title: Selective engagement of the cerebellum in action and social prediction throughout learning

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Abstract: A large body of neuroimaging evidence has highlighted how the cerebellum is engaged in a broad range of task domains beyond motor control. This work has revealed the functional topography of the cerebellum along with its functional connectivity with the cerebral cortex. In general, this literature has used tasks in which performance is assumed to be stable; for example, how does the cerebellum respond during a demanding working memory task or when listening to a story? Despite the prominent role of the cerebellum in sensorimotor learning, few studies have asked how cerebellar activation patterns during non-motor tasks change with learning. In the present study, we trained young adults (n=25) on five tasks: semantic prediction, visual search, action prediction, N-back, and social prediction. Participants completed 5 training sessions over 3 weeks. Sessions 1, 3, 5 were conducted in the MRI scanner while sessions 2 and 4 provided additional training outside the scanner. We leveraged a pre-trained cortico-cerebellar connectivity model to predict cerebellar activity at each scanning time point, with the main analysis focused on identifying task-specific deviations between observed and predicted cerebellar activity patterns. Importantly, while cerebellar BOLD signal decreased with learning across domains, the only tasks that showed greater cerebellar activity than predicted were the action and social prediction tasks. These findings suggest that cerebellar contributions are greater when learning tasks that involve predicting dynamic information, with this amplification observed across the sessions. The cerebellar activations were not greater than predicted by the cortico-cerebellar connectivity model for the perception, semantic, and working memory tasks.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

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Program #/Poster #: PSTR306.13/SS12

Topic: I.10. Human Learning and Cognition

Title: A practice-dependent working memory contribution to skill performance

Authors: *F. IWANE¹, E. R. BUCH², L. G. COHEN³;

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Abstract: Working memory capacity influences how well skills are executed in daily life. Learning new skills is typically characterized by an initial phase of rapid improvement (early learning where 95% of performance improvement occur), followed by a later period during which performance levels off or plateaus. Practice trials during this plateau period are characterized by high initial performance (HIP) segments followed by skill performance drops. These performance dynamics may reflect (a) speed-related fatigue induced by repetitive typing—a common explanation in the literature—or (b) an effective working memory capacity that limits further performance improvements.

Since skills may be relatively easier or harder to perform depending on constraints imposed by musculoskeletal factors, central nervous system organization or task complexity, we tested these competing hypotheses through evaluation of learning dynamics associated with skills performed at different speeds. We first used independent keypress transition time data acquired for all possible keypress pairs (n=16) performed with four digits (index, middle, ring and little finger) to develop a difficulty index for all possible 6-item keypress sequences. We tested three groups of healthy subjects (n=407), each of which learned a different Easy, Medium or Hard skill sequence.

We report that expression of early learning transitioned from offline (i.e. - skill improvements following periods of rest) to online (i.e. - skill improvements during practice) with decreasing naturalistic skill difficulty. After performance plateaued, the number of correct keypresses performed within the HIP period was greater for easier skills (Pearson's $r = -0.38$, $p < 0.001$) and continued to increase with additional practice for all three skills tested (Easy, $r = 0.64$, $p = 0.002$; Medium, $r = 0.71$, $p < 0.001$; Hard, $r = 0.67$, $p = 0.001$). Importantly, HIP keypresses temporally clustered within 5-7 chunks across all skills, with the average chunk content of more difficult skills (i.e. - keypresses/chunk) increasing with practice (Easy, $p = 0.18$, Medium, $p < 0.001$; Hard, $p = 0.001$). These results are consistent with slot- and resource-based models in which chunks compress information to improve effective working memory capacity. Furthermore, stable HIP dynamics were not observed in control groups that typed randomized sets of speed-matched sequences within each practice trial.

In conclusion, contrary to previous assumptions, performance dynamics during the plateau phase immediately following early skill learning are more likely due to practice-related changes in effective working memory capacity than to speed-related fatigue.

Disclosures: F. Iwane: None. E.R. Buch: None. L.G. Cohen: None.

Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.14/TT1

Topic: I.10. Human Learning and Cognition

Title: Micro-offline contributions to early learning transition from offline to online with decreasing skill difficulty

Authors: F. IWANE¹, E. R. BUCH², *L. G. COHEN³;

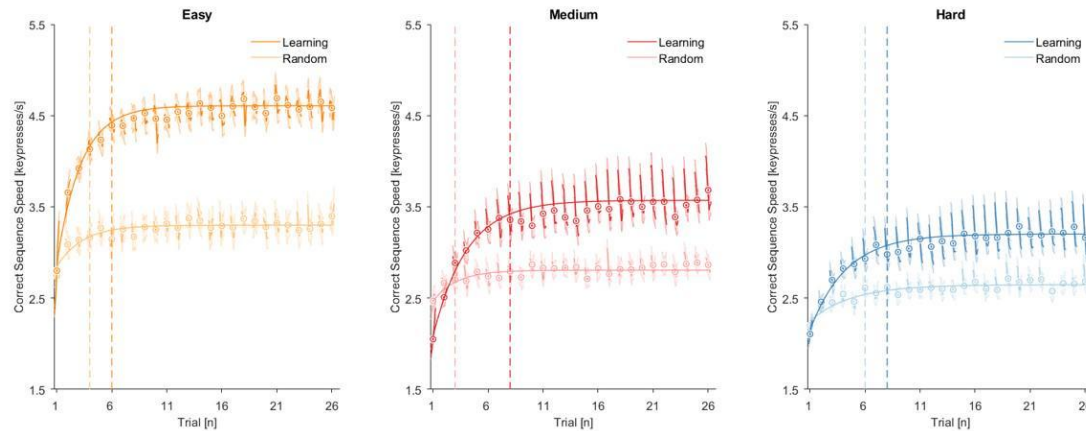
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Abstract: Naturalistic sequential skills require precise and coordinated finger movements. Skills are easier or harder to perform due to constraints imposed by musculoskeletal factors, central nervous system organization and task complexity. Thus, skill learning experiments could be enriched by comparing outcomes with those obtained from practicing random sequences that closely match the learned sequence in terms of naturalistic difficulty and biomechanical characteristics. Here, we compared performance dynamics with practice of repetitive sequence skills of different difficulty and matched random control sequences.

We developed a difficulty index for 6-item keypress sequences based upon independent keypress transition time data acquired for all possible keypress pairs (n=16) performed with four fingers. We then tested performance dynamics with practice of three skill sequences (n=407; Easy, Medium and Hard) and practice of random control sequences of matching difficulty indexes (n=256).

We found that relative to practice of random sequences of matching difficulty, practice of sequential skills resulted in significantly larger (a) total learning, comprising performance improvements across 26 practice trials (two-sample t-test; $p < 0.001$ for the three skills); (b) early learning, comprising skill improvements until performance reached 95% of the maximal for the practice session ($p < 0.001$ for the three skills); and (c) micro-offline gains across rest periods of early learning of harder skills (Medium: $p = 0.009$, and Hard: $p < 0.001$). Overall, early learning transitioned from offline to online with decreasing skill difficulty. Modeling the impact of random practice of matching difficulty on learning curves resulting from practice of repetitive skill sequences confirmed the specificity of micro-offline contributions to early skill learning of challenging skills.

We conclude that micro-offline contributions to early learning are skill-specific, transitioning from offline to online with decreasing task difficulty.



Disclosures: F. Iwane: None. E.R. Buch: None. L.G. Cohen: None.

Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.15/TT2

Topic: E.08. Multisensory Integration

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 SCIRB- DOH-C37714GG
 SCIRB- DOH-C38338GG
 Stratton VA Medical Center
 ICTS 5TL1TR001415-09

Title: Electroencephalogram correlates of proprioceptive processing and plasticity following training in chronic stroke survivors

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Abstract: Purpose: Proprioception—our sense of movement that contributes to motor learning and rehabilitation response after stroke—is impaired in over 50% of stroke survivors but is poorly understood. We measured brain signals with electroencephalography (EEG) during a proprioceptive “Crisscross” task with the Finger Individuating Grasp Exercise Robot (FINGER)

before (1 week) and after (1 month) 3 weeks of robotic hand therapy (NCT04818073), to identify neural markers of proprioceptive processing, stroke related impairment and therapy induced change.

Method: 46 chronic (>6 mos.) stroke survivors (18-85 y.o., 33 male), with unilateral upper extremity weakness participated with informed consent. In Crisscross, FINGER passively crossed the affected index and middle fingers in symmetric, alternating flexion/extension trajectories (0-36 deg) at 10 random speeds (8-18 deg/s) with inter-trial intervals of 2-3.5 s for 100 trials (5 runs, 20 trials/run) with vision of the hand occluded. Participants pushed a button with the unaffected hand when they perceived their finger's crossing; proprioception was quantified by mean detection error. 19 channels of EEG was acquired (DSI-24, Wearable Sensing). EEG data was filtered (0.1-30 Hz), denoised, baseline corrected (-0.2 to 0 s) and epoched (-0.2 to 4 s) with respect to movement onset; noisy epochs were removed ($\pm 100 \mu V$, <10% trials). Event related potentials (ERP) were calculated as the mean across trials.

Results: Proprioception improved with training (Wilcoxon t-test, $p < 0.005$). ERP showed a Contingent Negative Variation (CNV) over sensorimotor regions of the propriocepting hand that began at movement onset (initial stimulus) and slowly decreased to a negative peak at perceived finger crossing (imperative stimulus). Pre-training, better proprioception was associated ($r^2 = 0.30$, $p < 0.0001$) with larger CNV deflections (0.5-1s). Post training, the association strengthened ($r^2 = 0.53$) and indicated different responses between initially unimpaired (UP) and impaired (IP) participants. For UP, the CNV decreased more in the affected hemisphere compared to IP (Wilcoxon t-test, $p < 0.03$). For IP, the CNV decreased more in the unaffected than affected hemisphere (Wilcoxon signed rank test, $p < 0.03$). Post hoc analysis showed that finger velocity was correlated with EEG measured in similar sensorimotor regions as the CNV.

Conclusion: The CNV response elicited during proprioceptive estimation tasks is reflective of proprioceptive ability, shows plasticity with training, and may reflect velocity estimation processes. Initial proprioceptive ability elicited differential patterns of neural plasticity with training.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.16/TT3

Topic: I.10. Human Learning and Cognition

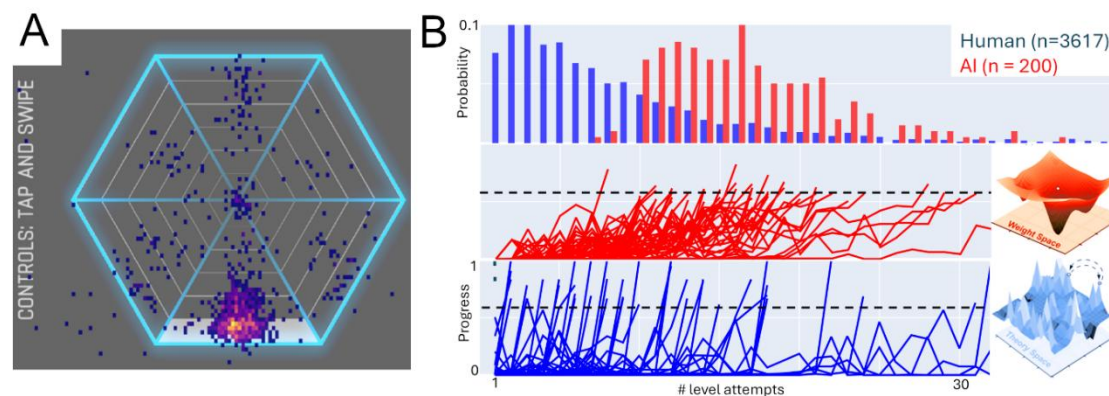
Support: Bial Foundation Grant #360/18
Department of Natural Sciences Startup Grant

Title: Humans learning a complex task are picky, sticky, and leapy

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Abstract: While neural networks approach human levels of performance in many complex tasks, they require much more training than humans. This may be because only humans can infer and apply generalizable principles from prior experience. However, the statistics that underlie the human learning process are poorly understood and hard to investigate in the large state spaces found in most complex tasks. We thus designed a cognitive task whose potential solutions are few enough for subjects to densely sample policy space, but complex enough to compel intelligent search. We launched the game as a smartphone-based app (hexxed.io) to collect data from 10k human participants. We find that unlike reinforcement learning agents (Deep-Q Networks ; DQNs), humans (1) search a highly restricted subset of the policy space (i.e., are 'picky'); (2) attempt even poor solutions many times before discarding them (are 'sticky'); (3) arrive at the optimal policy suddenly and unpredictably with a “leap of insight” (are 'leapy'). Our data suggest a “top-down” learning process by which humans propose explanatory solutions which they replace only upon collecting sufficient evidence to the contrary, in contrast to the “bottom-up” learning of DQNs that associates states with rewarding actions.



A) Humans are picky. Heat map of subjects' first action shows that certain locations are consistently targeted across the population. b) Humans are sticky and leapy. (top) Histogram of number of level attempts needed to pass the first level shows that humans (blue) are generally faster but more variable than DQNs (red). (middle) Each line shows the learning curve of a randomly selected DQN, showing a gradual increase in reward earned per trial as the agent gradually improves its policy through gradient descent, due to "bottom-up" learning (schematized in the right panel, adapted from Ullman et al. 2012). (bottom) Each line shows a learning curve of a randomly selected human, showing sudden jumps in performance after long periods of very low reward, suggestive of "top-down" learning over theory space (schematized in the right panel).

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.17/TT4

Topic: I.10. Human Learning and Cognition

Support: CIHR

Title: Sleep Spindle Dynamics Gate Motor Memory Consolidation: Evidence from Closed-Loop Targeted Memory Reactivation

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Abstract: Newly acquired motor memory traces undergo repeated reactivation, reorganization, and consolidation across cortical and subcortical networks, processes thought to occur during specific sleep EEG events known as sleep spindles. While spindles correlate with motor memory consolidation, their causal role remains unclear. We investigated whether targeted memory reactivation (TMR) during spindles could enhance motor memory consolidation using a closed-loop TMR paradigm. Thirty healthy right-handed adults learned two distinct 8-element motor sequences, one with each hand, paired with unique auditory cues. During subsequent non-REM 2 and 3 sleep, one sequence (TMR) was reactivated using auditory cues time locked to spindles, while the other sequence (non-TMR) served as a within subject control. TMR significantly enhanced overnight speed gains for correct sequence execution compared to non-TMR sequence. This behavioral enhancement correlated with the percentage of cues delivered during spindles and was specifically associated with temporally clustered spindles (≥ 2 within 6 seconds) rather than isolated spindles (> 6 seconds apart), suggesting spindle dynamics critically influence consolidation efficacy.

Pre-sleep fMRI data revealed differential neural engagement during initial learning: non-dominant hand sequences recruited additional bilateral frontal, cerebellar, and sensorimotor regions compared to dominant hand sequences, which showed focused activation in motor planning areas (SMA, caudate), and primary motor areas. This differential recruitment indicates compensatory neural strategies for non-dominant hand learning. Post-sleep fMRI revealed distinct TMR effects: non-dominant sequences showed TMR-specific increases in precuneus, hippocampus, dorsolateral prefrontal cortex, and angular gyrus, while dominant sequences

showed no neural changes, likely reflecting pre-existing neural efficiency that limited further optimization through reactivation. Crucially, across both hands, the percentage of TMR cues delivered during spindle clusters predicted post-sleep precuneus activation, suggesting this region may be particularly responsive to reactivation during periods of clustered sleep spindles. These findings establish that spindle-locked TMR causally enhances motor memory consolidation for both dominant and non-dominant hand sequences, though the underlying neural mechanisms differ. The temporal dynamics of spindle clusters, rather than isolated spindles, appear critical for effective memory reactivation. This highlights the therapeutic potential of closed-loop TMR for motor rehabilitation.

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Poster

PSTR306: Motor Skill Learning, Memory Consolidation, and Sensorimotor Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR306.18/TT5

Topic: I.08. Learning and Memory

Support: NINDS IRP

Title: Behavioral characterization of different error types and correction strategies in naturalistic skill learning

Authors: *I. KARUNATHILAKE¹, F. IWANE³, L. G. COHEN², E. R. BUCH⁴;
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Abstract: Many real-world motor skills rely on the generation of action sequences from memories acquired through practice. For example, pianists reciting a well-known piece of music demonstrate expertise by playing keypress (KP) sequences at high speeds with few errors. When errors do occasionally occur, they can be classified as: 1) substitutions (e.g. - playing the wrong note), 2) insertions (e.g. - playing an extra note) or 3) deletions (e.g. - omission of a note that should have been played). Importantly, in real-life settings these errors must often be intrinsically inferred, and different error types may be related to different underlying neural processes. Thus far, no unsupervised method for identifying different types of generative KP sequence errors has been reported. Furthermore, it is not known if (a) the predominant error type changes as a function of skill learning, and (b) if individual error types are characterized by unique patterns of performance speed changes relative to correct sequences. We developed a novel probabilistic approach for labeling generative KP sequence errors and error types with 100% accuracy in a simulation study, and then evaluated how error composition evolved over an initial naturalistic skill learning session (n = 400 participants). Participants were instructed to

type a fixed sequence as fast and as accurately as possible over 26 trials with their left non-dominant hand. Participants were not provided with any explicit feedback when errors occurred, nor instructed on how they should be corrected. Under these conditions, we found that substitutions were more prevalent across the session than other error types ($68.1 \pm 36.3\%$ SD; $p < 0.001$) followed by equally distributed insertions and deletions ($16.8 \pm 24.9\%$ vs $15.4 \pm 22.1\%$; $p = 0.42$). Substitution errors occurred most frequently in the context of middle-to-little finger transitions. Furthermore, quantitative decomposition of keypress transition speed patterns surrounding individual errors was used to infer whether they were (a) undetected, (b) detected but not corrected, or (c) detected and corrected. For example, relative patterns of slowing around insertion errors could inform on mistakes without conscious awareness, thus followed by no slowing, and those consciously detected and corrected, as shown by subsequent slowing. Characterization of unique error features and adaptive responses in unconstrained naturalistic learning paradigms will facilitate the investigation of neural mechanisms underlying generative skill behavior in health and disease.

Disclosures: I. Karunathilake: None. F. Iwane: None. L.G. Cohen: None. E.R. Buch: None.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR307.01/TT6

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: RS-2004-00487298
RS-2024-00348451

Title: Insightminer™-driven discovery of novel aav capsids for targeted gene therapy

Authors: *B. LEE, M. KIM, H. LEE, J. LEE, J. LEE, J. KANG, H. SO, S. KIM, S. CHOE, M. KWON, B. YOO, J.-W. KIM;
Genixcure, Suwon-si, Korea, Republic of

Abstract: Efficient and neuron-specific gene delivery across the central nervous system (CNS) is essential for advancing gene therapies for a broad spectrum of neurological disorders. Although adeno-associated virus serotype 9 (AAV9) is widely used for CNS-targeted delivery, it exhibits limited neuronal specificity and suboptimal transduction efficiency. To address these limitations, we developed GenC2, a novel AAV capsid variant incorporating a rationally designed 7-mer peptide insertion, identified in part using InsightMiner™, an AI-guided peptide sequence recommendation platform for capsid engineering. GenC2 exhibited significantly improved transduction efficiency over AAV9 in both SH-SY5Y human neuroblastoma cells and primary mouse motor neurons. Following intravenous administration in mice, GenC2 mediated robust, neuron-specific transduction throughout the brain, with minimal off-target expression in non-neuronal tissues. In the CNS of the common marmoset, a non-human primate model, GenC2

maintained strong neuronal tropism, supporting its translational potential for clinical application. To further improve GenC2, we performed saturation mutagenesis at the 7-mer insertion site and adjacent capsid residues. Concurrently, we constructed a diverse AAV capsid library, comprising both mutagenesis variants and additional GenC2-derived sequences recommended by InsightMiner™. This library is undergoing systematic evaluation to identify novel capsids with enhanced tissue specificity and transduction efficiency. These efforts are expected to yield next-generation AAV vectors with improved blood-brain barrier (BBB) penetration, enhanced neuronal specificity, or novel tropisms beyond those of GenC2. Collectively, our findings establish GenC2 as a promising neuron-targeted AAV capsid with strong translational relevance for CNS gene therapy. More broadly, this work demonstrates the utility of AI-assisted rational design in accelerating the development of advanced gene delivery platforms for diverse therapeutic applications.

Disclosures: **B. Lee:** A. Employment/Salary (full or part-time): Genixcure. **M. Kim:** A. Employment/Salary (full or part-time): Genixcure. **H. Lee:** A. Employment/Salary (full or part-time): Genixcure. **J. Lee:** A. Employment/Salary (full or part-time): Genixcure. **J. Lee:** A. Employment/Salary (full or part-time): Genixcure. **J. Kang:** A. Employment/Salary (full or part-time): Genixcure. **H. so:** A. Employment/Salary (full or part-time): Genixcure. **S. kim:** A. Employment/Salary (full or part-time): Genixcure. **S. choe:** A. Employment/Salary (full or part-time): Genixcure. **M. Kwon:** A. Employment/Salary (full or part-time): Genixcure. **B. Yoo:** A. Employment/Salary (full or part-time): Genixcure. **J. Kim:** A. Employment/Salary (full or part-time): Genixcure.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR307.02/TT7

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH/NINDS 1F99NS141400-01
NIH/NCATS TL1TR003169
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Presidents Research Excellence Award (University of Texas MD Anderson UTHealth Houston Graduate School of Biomedical Sciences)
Dr. John J Kopchick Fellowship (University of Texas MD Anderson UTHealth Houston Graduate School of Biomedical Sciences)
NIH/NIGMS 1T32GM152796-01
NIH/NIDDK R01DK131446

Title: Rab4 spatially and functionally converges with Rab7 in the degradative endolysosomal network across species

Authors: *H. YANG¹, S. M. FARMER, Jr.², S. XU², Y. YU², X. YE², J. CAI³, B. RIOS², A. SOLBACH², D. COVARRUBIAS⁴, V. CHUONG², T. MOORE², E. FURR STIMMING², Q. TONG², S. ZHANG²;

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Abstract: Endolysosomal trafficking is critical for maintaining cellular homeostasis, especially in neurons, which must maintain their tidiness throughout an organism's lifespan. Rab GTPases are key regulators of this pathway, yet most functional insights come from cell-based studies, with limited validation in a physiological system. In mammalian cells, Rab4 and Rab7, associated with endosomal recycling and degradation, respectively, are thought to localize to spatially distinct early and late endosomes. However, their localization and functional relationship in a physiological system remains unclear. Using *Drosophila* (fruit flies), we carried out super-resolution imaging and observed widespread colocalization of endogenous Rab4 and Rab7 across tissues and developmental stages, including in the larval neuromuscular junction and adult brain. Through genetic analysis using the UAS-Gal4 binary system and Rab4 knockout (KO) mutants, we found that Rab4 and Rab7 are recruited to endosomal membranes via distinct mechanisms but have opposing effects on endolysosomal size: Rab4 overexpression or Rab7 impairment caused enlargement, while Rab4 loss or constitutively active Rab7 reduced them. Further, Rab4 deficiency suppressed Rab7 impairment-induced enlargement, and vice versa. Moreover, genetic interaction assays revealed that loss of Rab4 compromised the viability of heterozygous Rab7-deficient flies but not Rab5 or Rab11 mutants, suggesting functional interplay between these two proteins. To assess downstream consequences of Rab4's overlap with degradative Rab7, we used western blotting and immunostaining to analyze the levels of β PS-Integrin (fly β 1-integrin ortholog), a Rab4-recycled cargo critical for synaptic integrity. We found that its levels increased in Rab4-KO and decreased in Rab4-overexpressing flies. Moreover, to verify our results in a mammalian system, we used immunostaining and found extensive overlap between Rab4 and Rab7 in mouse hippocampal neurons and human SH-SY5Y neuronal cells. Finally, live super-resolution SIM imaging of SH-SY5Y cells revealed dynamic trafficking of β 1-integrin between Rab4- and Rab7-positive endosomes, suggesting that Rab4 promotes cargo influx towards Rab7 degradative compartments. Taken together, our data uncover a novel, conserved role for Rab4 in cargo degradation, and future work will explore how this pathway becomes disrupted in neurodegenerative diseases and whether it can be therapeutically targeted.

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Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR307.03/TT8

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: China Brain Initiative Grant (STI2030-Major Projects 2021ZD0202803)

Title: An AAV Toolset for Microglia-Specific Transgene Targeting across Species

Authors: *F. YU;

Chinese Inst. for Brain Research, Beijing(CIBR), beijing, China

Abstract: An AAV Toolset for Microglia-Specific Transgene Targeting across Species

Authors *F. YU¹, M. LUO¹; ¹Chinese Institute Chinese Institute for Brain Research, Beijing (CIBR), China **Disclosures** F. YU: None. **M. Luo:** None. **Abstract** Microglia, the resident immune cells of the central nervous system, are critical to brain health and disease. Despite advances in AAV capsid and promoter engineering, achieving precise and efficient viral targeting of microglia across brain regions and species remains challenging. Here, we introduce an AAV toolset that enables stable, specific, and robust transgene expression in microglia across diverse brain areas and species. Using single-cell chromatin accessibility sequencing, we identified a 446-bp microglia-specific regulatory element (MSRE). We also developed the AAV variant 2/9.m, which provides higher production yields and improved transduction efficiency. To address suboptimal expression of certain genes, we devised a microglia-specific codon optimization strategy. Therefore, this system enables sensor expression and gene knockout in microglia, facilitating functional studies. The AAV2.9m-MSRE system supports robust microglia-specific transgene expression in healthy and diseased mice, rats, non-human primates, and human ex vivo brain slices. This versatile platform advances microglial gene delivery for basic research and enhances the therapeutic potential of AAVs for neurological disorders.

Disclosures: F. Yu: None.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR307.04/TT9

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: NRF-2019R1A5A2027340
RS-2024-00399237
RS-2024-00345742
NRF-2022R1A6A3A01087307

Title: Engineered exosomes enable CRISPR-Cas-based epigenome editing in Alzheimer's disease models

Authors: *J. LIM¹, J. HAN^{2,1}, D.-G. JO¹;

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Abstract: Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by synaptic dysfunction, protein aggregation, and cognitive decline. Despite advances in therapeutic development, current strategies remain largely ineffective at halting or reversing disease progression. One promising avenue involves the intracellular delivery of therapeutic proteins, which can modulate disease-related pathways directly. However, the efficient transport of large, functional proteins across the cell membrane remains a significant technical challenge. In this context, exosomes—naturally secreted nanoscale vesicles—have gained attention as biocompatible and efficient carriers for delivering bioactive molecules into target cells. To address this unmet need, we developed a photoinducible exosome system termed MAPLEX (mMaple3-mediated protein loading into and release from exosome). In this platform, cargo proteins are tagged with the photocleavable protein mMaple3, which binds exosomal proteins such as CD9. This interaction allows stable cargo loading into exosomes, and upon blue light illumination, photocleavage of mMaple3 subsequently release intact proteins from the exosomal membrane. We applied this system to epigenetic modulation in the brain, using MAPLEXs loaded with dCas9 ribonucleoprotein complexes targeting β -site amyloid precursor protein cleaving enzyme 1 (*Bace1*). These complexes were fused to the catalytic domain of DNA methyltransferase 3A (DNMT3A) and guided by a *Bace1*-specific single guide RNA. Following intranasal delivery in 5xFAD and 3xTg-AD mice, two widely used Alzheimer's disease models, we achieved successful methylation of CpG sites within the *Bace1* promoter region. This resulted in sustained downregulation of *Bace1* expression, reduced amyloid plaque deposition, and improvements in recognition memory. These findings demonstrate that MAPLEX enables targeted, on-demand epigenetic editing in the brain and represents a flexible and minimally invasive approach to regulate gene expression in Alzheimer's disease and other neurodegenerative disorders.

Disclosures: J. Lim: None. J. Han: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ExoStemTech. D. Jo: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ExoStemTech.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR307.05/TT10

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: Taiwan NSTC grant 111-2320-B-001-008-MY3
Taiwan NSTC grant 112-2321- B-002-024

Taiwan NSTC grant 111-2314-B-001-004
Taiwan NHRI grant NHRI-EX113-11336SI

Title: Development of an inducible SF1-FlpeERT2 mouse line for precise genetic manipulation in the ventromedial hypothalamic neurons

Authors: *S. LIN^{1,3}, S.-B. YANG^{1,2};

¹Inst. of Biomed. Sci., ²Neurosci. Program of Academia Sinica, Academia Sinica, Taipei, Taiwan; ³Taiwan Intl. Grad. Program in Interdisciplinary Neurosci., Natl. Taiwan Univ., Taipei, Taiwan

Abstract: Cell-type-specific genetic tools are essential for the functional dissection of neural circuits. Although Cre-loxP systems are widely used, they are often limited by ectopic and transient developmental expression, which can confound the interpretation of the experimental outcomes. To enhance spatial and temporal specificity and enable intersectional genetic targeting, we generated an inducible *SF1-FlpeERT2* knock-in mouse line via CRISPR-Cas9 genome editing. Steroidogenic factor 1 (SF1, *NR5A1*) is a transcription factor selectively expressed in neurons of the ventromedial hypothalamus (VMH), a region critical for regulating energy homeostasis and innate behaviors. Existing SF1-Cre BAC transgenic lines suffer from off-target expression in non-VMH regions—including the cortex, amygdala, and peripheral tissues—limiting their utility in adult functional studies. In our new *SF1-FlpeERT2* line, Flp recombinase activity is tightly regulated upon tamoxifen induction, providing accurate temporal control of transgene expression. Using the Ai213 triple-recombinase reporter line, we verified that Flp activity is undetectable without tamoxifen and, upon induction, is specifically restricted to the dorsomedial VMH. To further validate cell-type specificity, we crossed *SF1-FlpeERT2* mice with *vGlut2-Cre*, *GAD2-Cre*, and *Esr1-Cre* lines. As expected, the majority of SF1-Flpe neurons were glutamatergic, with minimal overlap with GABAergic neurons. A distinct subpopulation co-expressing *Esr1* was identified in the middle VMH. In conclusion, this inducible *SF1-FlpeERT2* mouse line provides precise spatial and temporal control of transgene expression in SF1-expressing VMH neurons and is compatible with existing Cre-driver lines, making it an excellent tool for intersectional studies of hypothalamic circuits.

Disclosures: S. Lin: None. S. Yang: None.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR307.06/TT11

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: Bayerische Forschungsförderung AZ-1597-23

Title: Characterization of novel adeno-associated virus (AAV) - capsid variants in the mouse brain for future use in seizure therapy

Authors: *E. ARSLANTAS, V. MEHLFELD, M. BIEL;
Ludwig-Maximilians-Universität Munich, Munich, Germany

Abstract: AAV vectors have emerged as a powerful tool for gene therapy of brain disorders, providing targeted, long-lasting therapeutic effects. New capsid variants with improved cell specificity and reduced immunogenicity are of great interest for neuropathological therapeutic approaches. In this context, the Hyperpolarization-activated cyclic nucleotide-gated channel 2 (HCN2) knock-in mice with a point mutation in the nucleotide-binding site (HCN2EA) serve as a model for generalized epilepsy with absence seizures (AS). Since HCN mutations also cause epilepsy in humans, the HCN2EA mouse is an interesting animal model for testing new gene therapy approaches for HCN channelopathies. We tested novel variants of the AAV9 capsid (GL and NN) encoding eGFP under the control of different promoters in the mouse brain and compared them to the AAV9 WT variant. In transduced primary cells, generated from mice aged P0 (primary neurons) or P5-7 (primary astrocytes / microglia; sex not considered), we observed that all capsid variants using the CMV promoter resulted in eGFP expression in neurons and astrocytes, but not in microglia. Using the neuron-specific human synapsin promoter (hSyn), eGFP expression was restricted to neurons. Based on these results, we injected the viral capsids into the thalami of WT mice using stereotaxic surgery. Three weeks later, eGFP expression was quantified in different brain regions using RT-qPCR and Western blotting (n=3 per capsid). All capsid variants exhibited the strongest expression in the thalamus. AAV9.GL was also able to express eGFP in thalamo-cortical neurons, which are the main target for AS therapy. Therefore, we are currently investigating a gene supplementation strategy using the AAV9.GL capsid for functional studies in the HCN2EA mouse model. Stereotaxic injection of AAV9.GL hSyn-HCN2-HA into the thalami of HCN2EA mice resulted in 5-fold increase in HCN2 expression, as determined by RT-qPCR (n=3, p<0.005, unpaired t-test) and Western blotting. Preliminary EEG measurements of treated HCN2EA mice (male only) suggest a reduction in the seizures per day compared to untreated mice. Together, these findings provide an important basis for the development of a novel gene therapy for AS.

Disclosures: E. Arslantas: None. V. Mehlfeld: None. M. Biel: None.

Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.07/TT12

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: 5R00MH121534-05

Title: Circuit and cell type-specific modulation of gene sequence and expression with CRISPR-rabies

Authors: *Z. ZHANG, D. VELMESHEV;
Neurobio., Duke Univ., Durham, NC

Abstract: CRISPR gene editing has revolutionized our ability to study and manipulate specific genes, enabling novel insights into gene function and potential therapies for brain disorders. Recent advances in cell type-specific regulatory elements and viral delivery systems have made precise in vivo gene editing possible. However, neurons with similar molecular profiles can belong to distinct neural circuits, complicating efforts to manipulate circuit function and behavior. To address this challenge, we developed CRISPR-rabies (CRV), a novel approach that enables gene editing within anatomically defined neural circuits. By combining CRV with cell type-specific Cas9 expression, we achieved circuit-specific and cell type-restricted gene modifications in vivo. Compared to conventional viral vectors such as AAV and lentivirus, CRV offers a unique advantage in targeting functionally connected neurons, regardless of shared molecular identity. *In vitro* validation using T7E1 mismatch assays and targeted deep sequencing confirmed that the gene editing efficiency of CRV is comparable to AAV or lenti-delivered CRISPR systems. We further optimized viral production protocols to maximize sgRNA packaging fidelity, ensuring consistent and efficient delivery across experiments. *In vivo*, CRV effectively knocked down endogenous ion channels such as KCNA1 and SCN1A in parvalbumin-positive interneurons, resulting in measurable changes in downstream CA3 pyramidal neuron excitability. Immunofluorescence staining validated robust and specific protein-level regulation in targeted neurons, confirming the functional impact of CRV-mediated regulations. Together, CRISPR-rabies enables precise gene modulation within defined neural circuits, bridging anatomical connectivity with genetic control. This platform offers a powerful tool for dissecting the causal relationship between gene function and circuit dynamics, and lays the groundwork for next-generation gene therapies targeting specific neural pathways in brain disorders.

Disclosures: Z. Zhang: None. D. Velmeshev: None.

Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.08/TT13

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: ZIA MH002928

Title: A comprehensive surgical protocol for high-throughput stereotaxic injections in nonhuman primates

Authors: *A. C. TANGEN¹, P. HARMON², B. B. AVERBECK³, R. AZADI⁴;

¹Carnegie Mellon Univ., Pittsburgh, PA; ²NIH, Bethesda, MD; ³NIMH/NIH, Kensington, MD;

⁴NIMH, Bethesda, MD

Abstract: Precise and efficient delivery of substances such as viral vectors or tracers into specific brain regions is essential for systems neuroscience, particularly in nonhuman primate models. These models are crucial for bridging the gap between basic research and clinical applications. Here, we present a detailed step-by-step surgical protocol in macaque monkeys for high-throughput microinjections that enables accurate targeting across a wide range of cortical and subcortical regions, including amygdala, striatum, hippocampus, area 25, area 24, ventral pallidum, basal forebrain and auditory areas. We also provide specific injection parameters such as number of sites and their spacing, injection volumes, and delivery rates. Moreover, we introduce a novel injection circuit capable of reliably delivering substances into targeted brain regions. The system consists of air tight connectors and tubing, made from commercially available components, along with a customized 3D-printed hub designed to hold the connectors and interface with stereotaxic manipulators. This setup enables multiple parallel injections, significantly reducing overall procedure time. We also present postoperative brain MRI to evaluate substance delivery both quantitatively and qualitatively. In addition, we provide electrophysiological data as supporting evidence of successful viral delivery, demonstrating the effectiveness of our methods for optogenetic experiments, as well as anatomical studies.

Disclosures: A.C. Tangen: None. P. Harmon: None. B.B. Averbeck: None. R. Azadi: None.

Poster

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Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: NSF-2525840
NIH-RF1MH120005
NIH-RF1MH123402
NIH-RF1MH124611
NIH-RF1MH133764

Title: A Novel Multicolor Trans-Synaptic System for High-Throughput Circuit Mapping in *Drosophila*

Authors: *Y. LI¹, J. TANG¹, N. SBISA¹, H. CHEN¹, B. DUAN¹, T.-Y. CHENG¹, K. ATHUKORALA¹, R. POLLOCK¹, L. A. WALKER¹, H. JIANG², M. CUI², Y. YAN³, D. CAI¹;
¹Univ. of Michigan, Ann Arbor, MI; ²Purdue Univ., West Lafayette, IN; ³Univ. of Illinois Chicago, Chicago, IL

Abstract: Understanding neural circuit function requires mapping connections between individual neurons at scale. While EM connectomics provides synaptic resolution, it is limited by throughput, animal-to-animal variability assessment, and it lacks molecular information. Existing trans-synaptic labeling tools like transTango can identify connected neurons, but they use single-

color reporters that prevent reconstruction of individual neuron morphologies, limiting circuit analysis.

We developed a breakthrough solution by creating novel transgenic *Drosophila* lines that combine sparse labeling mechanisms with Bitbow multicolor reporters and transTango trans-synaptic systems. This enables sparse or single-cell labeling of presynaptic neurons alongside multicolor labeling of their postsynaptic partners. Using expansion microscopy and custom high-throughput line-scanning confocal microscopy, we can image entire fly brains at sub-hundred-nanometer resolution within hours.

Our approach allows rapid reconstruction of individual neuron morphologies within connected circuits, enabling direct correlation with EM connectome datasets such as FlyWire and Hemibrain. Computer-aided tracing generates circuit consensus maps while preserving molecular and anatomical profiling capabilities unavailable in EM. This bridges the critical gap between high-resolution EM structure and scalable functional analysis.

These tools represent a paradigm shift for connectomics, enabling quantification of circuit variability across individuals, integration of molecular identity with connectivity, and high-throughput discovery of circuit motifs. We demonstrate the system's power to rapidly map and molecularly profile neural circuits, dramatically accelerating our ability to understand brain function from molecules to behavior.

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Poster

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Program #/Poster #: PSTR307.10/TT15

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Title: Aav-shds: liver-detargeted aav capsids with high bbb activity in non-human primate and in vitro human models

Authors: *B. WANG¹, C. XU¹, W. HU², Q. WANG¹, D. YU¹;

¹AAVnerGene Inc., Rockville, MD; ²Dept. of Anat. and Neurobio., Virginia Commonwealth Univ. Sch. of Med., Richmond, VA

Abstract: The therapeutic potential of gene therapy hinges on precise transgene delivery and expression in target tissues—especially the central nervous system (CNS)—while minimizing off-target effects. Adeno-associated virus serotype 9 (AAV9) is widely used for its broad tropism and robust expression, but its limited ability to cross the blood-brain barrier (BBB) remains a major barrier to CNS gene delivery. Leveraging our ATHENA capsid engineering platform, we developed a novel family of capsids, termed AAV-ShDs, specifically optimized for CNS targeting in non-human primates (NHPs). In macaques, the lead capsid, AAV-ShD, achieved a

385-fold increase in DNA and an 8-fold increase in RNA expression in the brain compared to AAV9, as determined by barcoded next-generation sequencing. In the spinal cord, AAV-ShD exhibited 25-fold higher DNA and 28-fold higher RNA levels. In an in vitro transwell BBB model, AAV-ShDs demonstrated enhanced transcytosis and efficiently transduced human neurons, astrocytes, and microglia. They also effectively delivered transgenes to primary human brain microvascular endothelial cells, pericytes, hippocampal astrocytes, and cortical microglia. Notably, in human iPSC-derived cerebral organoids containing microglia, AAV-ShDs showed superior transduction compared to AAV9 across various neural cell types. Importantly, AAV-ShDs also exhibited liver detargeting in NHPs, reducing peripheral exposure. The elevated DNA levels in transduced CNS tissues and cells suggest potential for long-term expression at lower doses. Conclusion: AAV-ShDs represent a promising cross-species gene delivery platform capable of efficiently crossing the BBB, with reduced off-target transduction. Their high CNS specificity and expression profile support future development for neurological gene therapy applications.

Disclosures: B. Wang: None. C. Xu: None. W. Hu: None. Q. Wang: None. D. Yu: None.

Poster

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Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: NIA grants R01AG058854 (ENIGMA World Aging Center)
U01AG068057-05 (AI4AD)
Alzheimer's Association grant (to P.R.)

Title: Mapping Focal and Generalized Effects of Common Genetic Variants on Human Brain Structure

Authors: *E. J. GLEAVE^{1,2}, L. GARCÍA-MARÍN³, Z. CEJA³, M. E. RENTERÍA³, P. M. THOMPSON², P. RAJAGOPALAN^{2,4};

¹USC, Los Angeles, CA; ²Imaging Genet. Center, Mark and Mary Stevens Neuroimaging & Informatics Institute, Keck Sch. of Medicine, Univ. of Southern California, Marina Del Rey, CA; ³Brain and Mental Hlth. Program, QIMR Berghofer Med. Res. Inst., Brisbane, Australia; ⁴Dept. of Radiology, Los Angeles Gen. Med. Ctr., Los Angeles, CA

Abstract: Genome-wide association studies (GWAS) have greatly advanced the quest to understand how specific genetic variants affect human brain structure. Recent GWAS in over 75,000 individuals scanned with MRI (Garcia-Marin et al., Nature Genetics, 2024) identified more than 500 common variants associated with subcortical brain volumes, sparking interest in how strongly these genetic markers overlap across different brain systems. While this can be estimated by hierarchical clustering of the genetic correlation matrix to identify 'genetic

modules' (Chen et al., 2012; Fjell et al., 2015), no brain-wide maps of these effects are available. Here, we computed polygenic scores (PGS) from loci associated with ten brain volume regions of interest (ROIs): nine major subcortical structures and intracranial volume (listed in Fig 1), where each locus was weighted by its strength of association with regional volume. SBayesR was implemented to derive the PGS (Lloyd-Jones et al. 2019) implemented within the Genome-wide Complex Trait Bayesian analysis (GCTB v2.0) software tool (Chung et al., 2021). In an independent sample not included in the discovery GWAS, we ran voxel-based morphometry (VBM) to map the 3D profile of brain regions where gray matter volume (GMV) was associated with each PGS. The ENIGMA Computational Anatomy Toolbox (CAT12: Gaser et al., 2024) was used to perform the large-scale VBM segmentation of 3D T1W brain MRI data from the UK Biobank (UKB). The statistical relationship between voxel-wise brain GMV and each ROI PRS was assessed using a linear mixed model (see Fig 1 for formula, p-values and maps) on the independent subset of 2,830 UKB participants (51.0% female; 68.8 (8.1SD) years old). We found highly significant, localized effects for PGS defined for the amygdala, thalamus, and components of the basal ganglia, but, intriguingly, PGS for brain stem volume was associated with widespread differences throughout the brain. These brain-wide maps distinguish both modular and distributed genetic influences on the brain, offering a novel lens on the genomic architecture of brain structure.

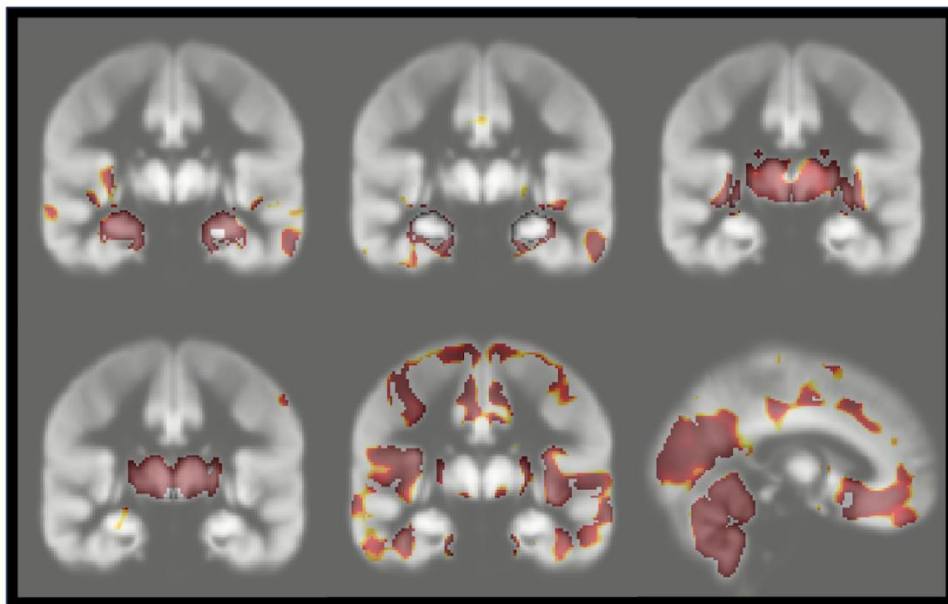


Figure 1. Linear mixed model analyses show associations between polygenic scores for ten brain ROIs and GMV(y) from 2,830 UKB participants— adjusting for age, sex, genetic ancestry (first four principal components), total intracranial volume (TIV; estimated via CAT12 VBM segmentation), and a random effect (Γ) accounting for the different scan sites in the UKB: $y = \beta_{ROI,PGS} + \beta_{age} + \beta_{sex} + \beta_{PC,1-4} + \beta_{TIV} + \Gamma_y$.

Voxel-wise whole-brain significance maps are shown after standard-FDR correction ($q=0.05$) for: Amygdala (top left; standard-FDR critical P-value=0.0032), Hippocampus (top center; standard-FDR critical P-value=0.0028), Putamen (top right; standard-FDR critical P-value=0.0031), Thalamus (bottom left; standard-FDR critical P-value=0.0011), and the Brainstem (bottom center, bottom right; standard-FDR critical P-value=0.0239).

Additionally, significant results were found for: Caudate (standard-FDR critical P-value=0.0012), Nucleus Accumbens (standard-FDR critical P-value=0.0014), Pallidum (standard-FDR critical P-value=0.0013), Ventral Diencephalon (standard-FDR critical P-value=0.0128), and ICV (run without TIV to avoid overfitting; standard-FDR critical P-value=0.0498).

Disclosures: E.J. Gleave: None. L. García-Marín: None. Z. Ceja: None. M.E. Rentería: None. P.M. Thompson: None. P. Rajagopalan: None.

Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.12/TT17

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R01EB032854

Title: Development of a novel photostable yellow fluorescent protein through directed evolution

Authors: *A. GREY¹, G. LAMBERT², N. C. SHANER²;

¹Univ. of California - San Diego, San Diego, CA; ²Neurosciences, Univ. of California San Diego, La Jolla, CA

Abstract: Fluorescent proteins are genetically encodable labels used broadly for probing biological systems, enabling researchers to observe dynamic processes in living cells. While most existing fluorescent proteins have been extensively optimized for brightness and fast maturation, many remain vulnerable to photobleaching, limiting the number of images that can be collected for a given sample. Fluorescent proteins in the “yellow” wavelength class (YFPs) have historically been among the least photostable, even though variants such as mVenus are used very commonly as imaging tags. Here, we report the directed evolution of a monomeric yellow variant of the bright green fluorescent protein AausFP1 from *Aequorea australis*. Using error prone mutagenesis with screening for improved photostability, we identified key residues that we then targeted in site-directed mutagenesis libraries. After several rounds of alternating random and directed library selection, the top clones were further vetted using OSER and FLUOPPI assays for monomeric behavior and fused to a variety of target proteins to evaluate performance in cells. The final YFP clone, mAbbY, was benchmarked in cells against the previously published *Aequorea victoria* GFP-derived variants mEYFP, mVenus, mGold, mGold2s, and mGold2t, demonstrating its higher photostability under both widefield and laser scanning confocal illumination. The development of mAbbY fills a long-standing gap in available photostable fluorescent proteins, bringing YFPs into parity with other wavelength classes.

Disclosures: A. Grey: None. G. Lambert: None. N.C. Shaner: None.

Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.13/TT18

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Title: An orthogonal TRAP for intersectional labeling of activated neurons in the mouse brain

Authors: *N. CHATZIRIS¹, B. JARVIE^{1,4}, C. LIU², Z. KNIGHT^{1,2,3,4},

¹Physiol., ²Neurosci. Grad. Program, ³Kavli Ctr. for Fundamental Neurosci., Univ. of California San Francisco, San Francisco, CA; ⁴Howard Hughes Med. Inst., San Francisco, CA

Abstract: The study of neural circuits has been greatly enabled by methods for genetic access to activated neurons. For example, the widely-used TRAP system uses tamoxifen to induce Cre recombination in neurons that are transiently activated. However, these approaches tag neurons based on their response to only a single stimulus, which limits the ability to define precise subpopulations of cells. Here we describe an orthogonal system (X-TRAP) in which the activity-dependent expression of Flp recombinase is gated by branaplam, a small molecule that triggers

splicing of the X-ON switch. We show that X-TRAP knock-in mice exhibit undetectable Flp recombination in the absence of drug and that branaplam treatment results in robust induction of recombination selectively in neurons that express *Fos*. We demonstrate that this approach can be used to label neurons that are activated by hormones, drugs, and dehydration. By combining the X-TRAP and TRAP2 alleles in the same mouse, it is possible to label cells with Cre and Flp recombination triggered by two different stimuli, and we apply this strategy to map neural circuits that control food intake. This approach for intersectional, activity-dependent labeling should enhance our ability to identify the neural correlates of behavior.

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Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.14/UU1

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Title: Bitbow transgenic for hyper-spectral neuron labeling in the mouse brain

Authors: *X. NIU¹, D. H. ROOSSEN, Jr.², L. A. WALKER³, Y. LI³, H. SU⁴, K. SHENG⁵, P. YIN⁶, D. CAI⁷;

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Inspired Engin., harvard university, Boston, MA; ⁵Wyss Inst. for Biologically Inspired Engin.,

⁶harvard university, boston, MA; ⁷Cell and Developmental Biol., Michigan Neurosci. Institute, Univ. of Michigan, Ann Arbor, MI

Abstract: Mapping the cellular origins and structural complexity of neuronal networks has been a fundamental challenge in neuroscience, complicated by the remarkable heterogeneity among brain cells. While Brainbow-based transgenic systems enable multispectral labeling to distinguish neighboring cells, their utility is constrained by limited color capacity. To address this limitation, we developed Bitbow—a digital variant that exponentially expands the labeling palette to 63 spectrally resolvable unique labels by a single transgene allele. Bitbow operates through a Flp-dependent, TRE-driven system utilizing inverted FRT sites flanking a reversely oriented fluorescent protein that switches between stable ON/OFF expression states upon Flp-mediated recombination. This system offers flexible implementation through crossing with Flp or tTA transgenic mice, viral vector delivery, or Cre-activated controls for cell-type-specific labeling. To enhance detection sensitivity and multiplexing capacity, we integrated Bitbow with Amplification by Cyclic Extension (ACE), a signal amplification strategy employing DNA barcodes conjugated to antibodies. ACE achieves over 100-fold signal amplification and enables simultaneous detection of many distinct proteins, facilitating quantitative analysis of rare proteins and complex molecular interactions within tissues. The combination of Bitbow transgenic mice with multi-round ACE staining provides unprecedented visualization of neuronal

connections, long-range projections, and spatial distribution of synaptic markers, creating a powerful platform for studying neuronal morphology and neural network architecture with exceptional resolution, scale, and throughput. This integrated approach opens new research avenues for investigating neurodevelopmental disorders, neurodegenerative diseases, and circuit-level plasticity by simultaneously tracking anatomical changes and molecular perturbations. The methodology extends to functional mapping applications, enabling correlation of labeled circuit motifs with immediate early gene expression and activity markers in response to behavioral paradigms. Collectively, Bitbow and ACE provide a comprehensive, multimodal framework for understanding brain structure and function, effectively bridging connectomics, molecular neuroscience, and systems biology to advance our understanding of neural circuit organization and dynamics.

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Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.15/UU2

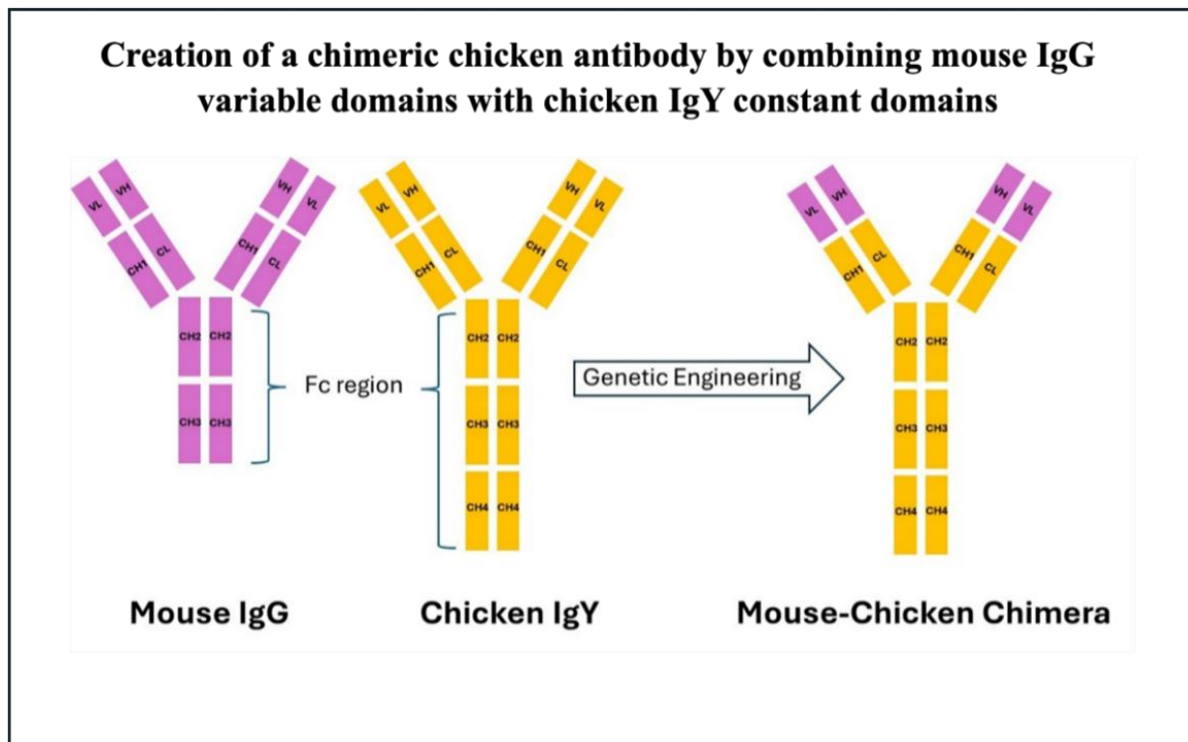
Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Title: Reformatting NeuroMab Mouse Monoclonal Antibodies into Novel Recombinant Chicken Chimeric Antibodies Enables Enhanced Reproducibility, Multiplexing, and Mouse Tissue Staining

Authors: *S. MOHIBI, M. HANS, V. PERNG, A. KRSTIC, J. CHOWDHARY, W. FRY; Antibodies Inc., Davis, CA

Abstract: The NeuroMab collection of mouse monoclonal antibodies is widely cited for its specificity, extensive validation, and performance in neuroscience research. To expand their utility and enable more flexible experimental designs, we have successfully converted 21 (out of 23 attempted) NeuroMab clones into recombinant chimeric chicken IgY antibodies. These novel reagents retain the validated antigen-binding regions of their NeuroMab predecessors, fused to the constant regions of chicken IgY, allowing researchers to combine the proven performance of NeuroMab antibodies with the unique advantages of chicken immunoglobulins, and offering distinct experimental advantages for use in mammalian systems. To generate these antibodies, we cloned the heavy and light chain variable regions from each NeuroMab mouse monoclonal clone into custom expression vectors containing chicken IgY constant domains. These constructs were co-expressed in mammalian cells cultured under serum- and protein-free conditions, enabling animal-free production and ensuring lot-to-lot consistency. The chicken IgY Fc region does not bind to mammalian Fc receptors, rheumatoid factors, or activate the complement system - features that reduce off-target binding and background signal in immunoassays. Importantly, these antibodies avoid the “mouse-on-mouse” problem, making them especially well-suited for

staining mouse tissue samples. Moreover, their use alongside mouse and rabbit antibodies enables expanded multiplexing options in immunofluorescence without signal overlap or cross-reactivity. In addition to rigorous in-house validation, these recombinant chicken antibodies have been independently validated. We demonstrate their effectiveness in immunostaining of mouse brain tissues and in multiplexed fluorescence labeling with mouse and rabbit antibodies. These results, including staining patterns and multiplexing performance demonstrate the superiority of these engineered antibodies over traditional mouse monoclonal antibodies.



Disclosures: **S. Mohibi:** A. Employment/Salary (full or part-time): Antibodies Incorporated. **M. Hans:** A. Employment/Salary (full or part-time): Antibodies Incorporated. **V. Perng:** A. Employment/Salary (full or part-time): Antibodies Incorporated. **A. Krstic:** A. Employment/Salary (full or part-time): Antibodies Incorporated. **J. Chowdhary:** A. Employment/Salary (full or part-time): Antibodies Incorporated. **W. Fry:** A. Employment/Salary (full or part-time): Antibodies Incorporated.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

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Program #/Poster #: PSTR307.16/UU3

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

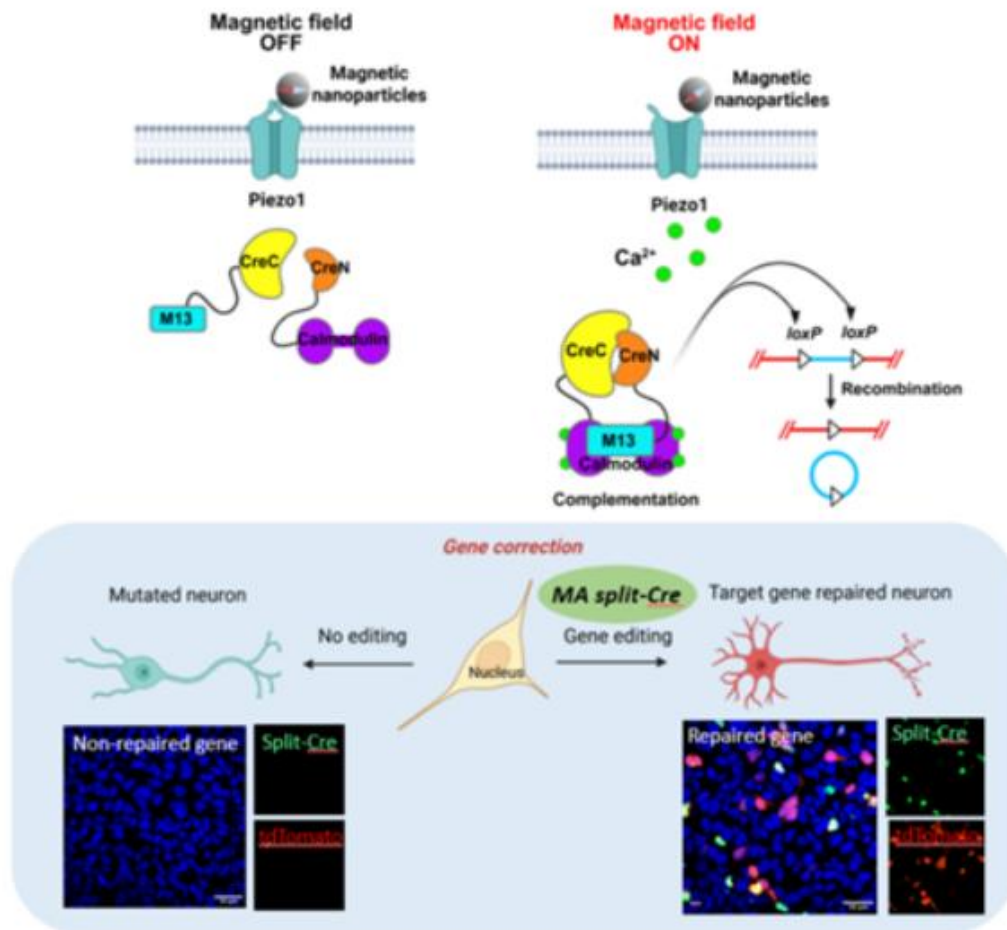
Support: IBS-R026-D1

Title: Magnetogenetically guided split Cre-loxP recombination system with spatiotemporal control of genomic engineering

Authors: *S.-H. CHOI¹, L. JAE HYUN³, J. CHEON²;

¹Yonsei Univ., Seoul, Korea, Republic of; ²Yonsei Univ., Seoul, ; ³YONSEI UNIVERSITY, SEOUL, Korea, Republic of

Abstract: The versatile Cre-loxP recombination system has been widely used as a site-specific genetic manipulation tool to precisely manipulate genomes of mammalian cells and transgenic animals. However, challenges with chemically inducible Cre-loxP systems include cytotoxicity, leakiness, off-target recombination, as well as limited ability to control systems with high spatiotemporal resolution. In particular, controlling gene expression in a timely and spatiotemporal manner in neurological and developmental disorders remains a major challenge. The magneto-activatable (MA) split Cre system functions as a single-chain, fast-responding, Ca²⁺-inducible Cre recombinase, providing a novel switch for spatiotemporal control of gene expression. We demonstrated that the Ca²⁺-sensitive mechanosensitive ion channel Piezo1 is activated upon application of an external magnetic field, leading to calcium influx and assembly of split Cre recombinase that enables gene editing. The MA split Cre system enables the targeted editing of genes associated with neurodevelopmental disorders *in vivo*. Genome engineering was successfully manipulated by the magneto-activatable split Cre system in the lateral hypothalamus region of the mouse brain, resulting in tdTomato expression. Our study provides a powerful tool to magnetogenetically manipulate genome engineering via the magneto-activatable split Cre system, enabling a previously unattainable level of noninvasive control that facilitates studies and therapies targeting a broad range of biological processes.



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Poster

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Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: The Brain and Behavior Research Foundation NARSAD Young Investigator Award
Mary E. Groff Foundation

McNair Scholars Program
Biology, Murchison, Semmes, and Voelcker SURF programs

Title: Label and destroy: identifying synaptically coupled neurons and removing their synapses

Authors: G. Z. SUN¹, S. BABBAR¹, A. JAUREGUI², A. FLANAGAN¹, A. DANG², J. CLARK³, E. FARRELL², C. WILLIAMS⁴, J. VALADEZ⁵, *G. M. BEAUDOIN^{1,2};

¹Biol., ²Neurosci. Program, ³Trinity Univ., San Antonio, TX; ⁴The Univ. of Texas at Austin, Austin, TX; ⁵Sch. of Med., Vanderbilt Univ., Nashville, TN

Abstract: Mapping synaptic connectivity is essential for understanding how information is transformed as it travels through the brain. Knowledge of how brain regions interact is required to understand the logic behind the modification of information between them. To address this, we are developing two synthetic biology tools for labeling synaptically-connected neurons and selectively eliminating synapses based on direct intercellular contact.

For trans-synaptic labeling, we repurposed *C. elegans* Notch-Delta orthologs Glp-1 and Lag-2 to use as contact dependent signalling pairs. A chimeric Glp1-Cre fusion protein was engineered by replacing the intracellular domain of Glp-1 with Cre recombinase. In theory, upon contact between a Glp1-Cre-expressing cell and a Lag-2-expressing partner, Cre is released and drives recombination at loxP sites, resulting in fluorescent labeling of synaptically connected neurons. This system successfully induced Cre-dependent reporter expression in HEK293 cells and is being validated in primary hippocampal neurons, where robust construct expression was confirmed via fluorescence microscopy. Notably, transfection experiments revealed that Glp1-Cre signaling is subject to lateral inhibition, as co-expression blocks activation.

For synapse-specific modification, we created CadPlexin, a chimeric receptor comprising the extracellular domain of *D. melanogaster* DE-cadherin and the transmembrane/intracellular domains of *M. musculus* Plexin-B2. Cadherins engage in homophilic binding across synapses, causing activation of Plexin-B2 signaling that induces repulsion via actin remodeling. We hypothesize that CadPlexin activation across synaptically linked neurons will mimic Semaphorin-induced clustering and lead to synapse destabilization. In non-neuronal cells, CadPlexin expression led to cell isolation and antibody-induced clustering triggered actin polymerization consistent with Plexin signaling activation.

The aim of these tools is to provide a platform to selectively label or eliminate synapses with cell-type and connection specificity. Ongoing work aims to continue to apply these constructs in more neuronal systems and to quantify the functional consequences of targeted synapse destruction or labelling.

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Poster

PSTR307: Genetic Techniques

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NIH Grant UF1MH128339
NIH Grant U19MH114830
Allen Institute for Brain Science

Title: Enhancer AAV Genetic Tools for Cell Types in the Mouse Brain

Authors: *B. TASIC;
Mol. Genet., Allen Inst. For Brain Sci., Seattle, WA

Abstract: The mouse brain comprises over 5,000 transcriptomically defined cell types. As part of the BRAIN Initiative Armamentarium Consortium, we have generated many tools across the mouse brain including cortex, basal ganglia and thalamus. We have characterized these tools through imaging and single cell transcriptomics approaches and provide all tools through public repositories. We also present all data through a public website the Genetic Tools Atlas (<https://portal.brain-map.org/genetic-tools/genetic-tools-atlas>).

Disclosures: B. Tasic: None.

Poster

PSTR307: Genetic Techniques

Location: SDCC Halls B-H

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Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant RF1MH121274
NIH Grant U19MH114830
Allen Institute for Brain Science

Title: Transgenic Tools for Cell Type Specific Intersectional Targeting of Cell Types

Authors: *S. NARAYAN, M. HOOPER, Y. BEN-SIMON, D. DWIVEDI, T. L. DAIGLE, B. WYNALDA, N. LUSK, S. W. WAY, A. OSTER, N. DEE, K. A. SMITH, B. THYAGARAJAN, J. WATERS, J. T. TING, M. REDING, K. RONELLENFITCH, L. NG, S. MUFTI, H. ZENG, J. ROTH, M. TAORMINA, S. D. HASTINGS, B. TASIC;
Allen Inst. for Brain Sci., Seattle, WA

Abstract: Single cell genomics has enabled the discovery of many cell-type-congruent marker genes. In addition, it has reinforced the notion that cell types are rarely defined by unique marker genes. Therefore, tools based on intersectional expression of two or more genes are critical for cell-type specific access. We recently published a toolkit comprised of driver lines, reporter

lines, and >1,000 different enhancer AAV (EnhAAV) vectors covering most subclasses of cortical cells (Ben-Simon et al., Cell, 2025). When coupled with suitable reporters, these driver lines and EnhAAVs give us unprecedented access to specific cell populations. Depending on the target cell type, cell-type specific access can be achieved with a combination of one or more driver lines and/or EnhAAVs. Some cell types can be labeled with a single driver line whereas others require careful selection of driver lines and/or EnhAAVs to achieve labeling at a single-cell-type level. These driver tools can be used in combination with old and new intersectional reporters. To enable easy creation of intersectional reporters, we have developed a transgenic platform with a new landing pad for Bxb1-integrase-mediated cassette exchange at the TIGRE locus. Dual (or triple) recombinase reporters can be Cre “AND/OR” FLP reporter lines with separate transcription units that independently report the presence of Cre “OR” FLP. This design can be used to drive different fluorescent proteins (FP), or different Opsins. The dual recombinase strategy can also be used for intersectional targeting, where the cargo is expressed in cells that express BOTH Cre and FLP. These driver and conditional reporter mouse transgenic lines and viral tools can be used to label, monitor, and manipulate distinct cell types in the brain thus enabling studies ranging from optical physiology to neuroanatomy and complex behavioral studies. All tools are or will be available from Jax, and their characterization is publicly available through the Genetic Tools Atlas (GTA) (<https://portal.brain-map.org/genetic-tools/genetic-tools-atlas>).

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Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.20/UU7

Topic: J.03. Anatomical Methods

Support: NIH R21 NS135479

Title: Small RNA barcodes for sensitive labeling of AAV transcripts

Authors: *Y. MAGAÑA¹, S. WANG², B. CHAVEZ¹, A. TORREBLANCA-ZANCA¹, M. J. CASTLE¹;

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Abstract: Adeno-associated virus (AAV) vectors are widely used in neuroscience research for recombinant gene expression and endogenous gene silencing. Labeling vector mRNA by *in situ* hybridization can track AAV expression with high sensitivity and without the need for a reporter

gene, but it can be challenging to detect similar transcripts when multiple AAVs are injected in the same animal, or when a single AAV can produce multiple RNA species (e.g. after Cre-Lox recombination). To address these challenges, we validated three small RNA barcodes (less than 50 bp) and developed methods for sensitive detection at single-transcript resolution in rodent and non-human primate nervous tissues using the BaseScope *in situ* hybridization platform. Beyond more sensitive and flexible labeling of AAV transcripts, we present two specific applications of these RNA barcodes for modern neuroscience research. First, intersectional experiments use multiple vectors to target gene expression to anatomically defined subsets of neurons. For example, when a cre-dependent “flip-excision” AAV is injected in one brain region, and AAV2retro-Cre is injected in a second connected brain region, cre-dependent gene expression is activated only in projection neurons that innervate the second region. This powerful method enables targeted manipulation and tracing of neural circuits, but results can be biased by variable injection targeting or spontaneous recombination of flip-excision vectors, and standard immunolabeling methods typically cannot detect these confounding variables. To address this problem, we placed two small RNA barcodes on both the sense (unrecombined) and antisense (recombined) strands of the flip-excision vector, and a third RNA barcode on the AAV-Cre vector. Labeling of multiple barcodes can then track the spatial distribution of both vectors and identify cells that exhibit nonspecific spontaneous recombination of the flip-excision vector without AAV-Cre. Second, multiplexed *in vivo* screening of pooled AAV libraries often requires that transduction be restricted to only one AAV genome per cell. Immunolabeling of reporter proteins may underestimate the number of cells that receive multiple AAV genomes due to insufficient sensitivity. By adding one RNA barcode to half of the AAVs in a pooled library and a second RNA barcode to the other half, *in situ* hybridization can identify cells that receive multiple AAV genomes (multiple barcodes) with maximum sensitivity and without the need for reporter proteins. These new methods for sensitive tracking of multiple AAV vectors and transcript species will improve the rigor and reliability of modern neuroscience research.

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Poster

PSTR307: Genetic Techniques

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Program #/Poster #: PSTR307.21/UU8

Topic: J.01. Molecular, Biochemical, and Genetic Techniques

Title: Platform for large-scale analysis of brain cell types at single cell resolution spanning longitudinal multi-modal measurements

Authors: *O. F. VILA¹, N. H. ELDER¹, S. FARAHVASHI¹, I. THOMAS¹, J. LAMSTEIN¹, S. DESHMUKH¹, T. AI², P. GHERARDINI¹, G. SCHROTH¹;

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Abstract: Single-cell technologies have revolutionized our understanding of cellular functions and heterogeneity. However, current high-throughput methods face limitations due to reliance on limited modalities, cell dissociation, and flow-based microfluidics. These constraints hinder the study of cell types relevant for neuro-immunological and neurodegenerative diseases.

Specifically, challenges arise with 1) cells inadequately identified by protein markers, 2) large or fragile cells that lose viability under harsh microfluidic conditions, and 3) cells requiring disruption from functional units for transcriptional analysis. Many brain cell types fall into one or more of these categories.

To overcome these limitations, we developed a high-throughput, multi-modal platform facilitating the longitudinal (days to weeks) study of individual cells, co-cultures, or neuronal networks without enzymatic digestion. Using this platform, we capture tens of thousands of cells in CellCage™ Enclosures (CCEs) and generate linked imaging and transcriptome data. CCE formation is highly flexible, enabling strategies to study diverse cellular functions, including isolation of single or multiple cells, somas within neuronal networks, or custom CCEs with openings for axonal outgrowth. Cells can be cultured and imaged for days to weeks in the CCEs to evaluate morphological changes due to differentiation, maturation, or proliferation before being processed for downstream sequencing (mRNA, CRISPRseq).

We demonstrate the platform's utility through multi-modal analyses of neurons, microglia, and astrocytes at single-cell resolution. It also enables culturing, imaging, and transcriptome analysis of small neurospheres. We established assays integrating phenotypic and transcriptomic data from the same cells, enabling study of molecular pathways underlying heterogeneous cellular functions at scale. This includes phagocytosis by microglia and calcium imaging in neurons and astrocytes. After longitudinal live-cell analysis of these assays, cells are processed for single-cell mRNAseq within CCEs. Notably, somas from neurons are sequenced intact within networks. Unlike current approaches that require dissociation—disrupting function—our method captures transcriptomes from neurons that remain functionally intact. This enables the generation of rich datasets integrating imaging, transcriptomics, and CRISPR perturbations for pathway analysis and target discovery.

By enabling transcriptomic study of clinically relevant functions while preserving cell integrity, this platform provides a powerful tool for advancing neuro-immunological research.

Disclosures: **O.F. Vila:** A. Employment/Salary (full or part-time): Cellanome. **N.H. Elder:** A. Employment/Salary (full or part-time): Cellanome. **S. Farahvashi:** A. Employment/Salary (full or part-time): Cellanome. **I. Thomas:** A. Employment/Salary (full or part-time): Cellanome. **J. Lamstein:** A. Employment/Salary (full or part-time): Cellanome. **S. Deshmukh:** A. Employment/Salary (full or part-time): Cellanome. **T. Ai:** A. Employment/Salary (full or part-time): Cellanome. **P. Gherardini:** A. Employment/Salary (full or part-time): Cellanome. **G. Schroth:** A. Employment/Salary (full or part-time): Cellanome.

Poster

PSTR307: Genetic Techniques

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Topic: J.01. Molecular, Biochemical, and Genetic Techniques

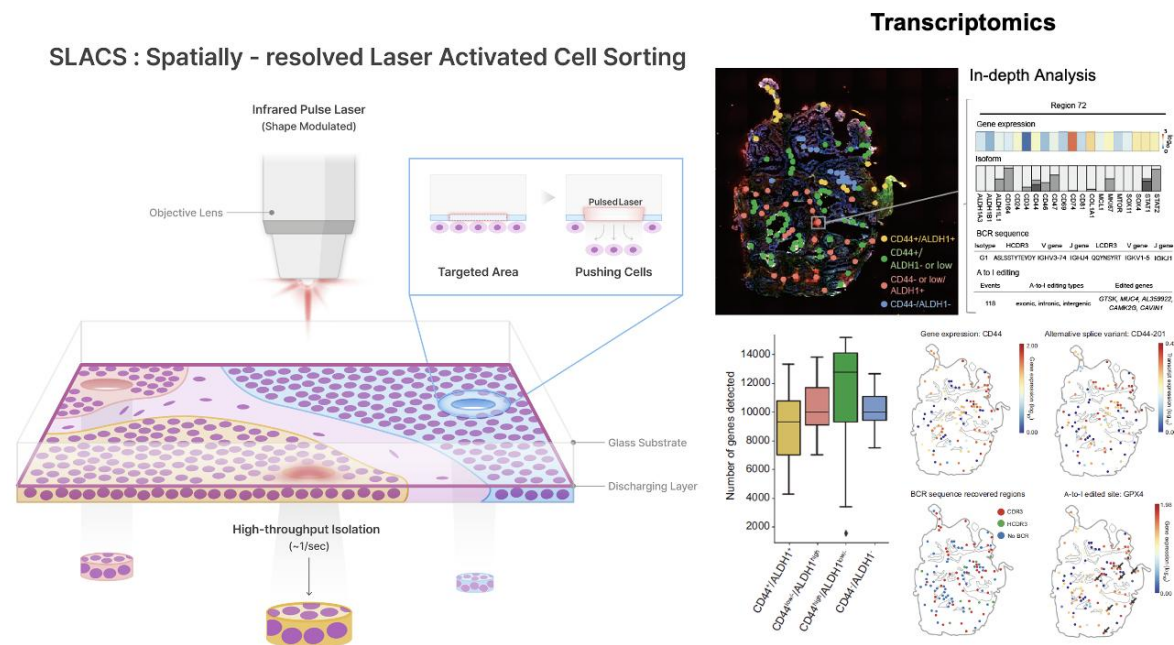
Support: RS-2024-00508416
RS-2024-00454407
RS-2024-00468338
RS-2024-00451981
RS-2024-00508416

Title: Spatially Resolved Full-Length Transcriptome and Epitranscriptome Profiling with Spatially Resolved Laser Activated Cell Sorting

Authors: *S. LEE¹, A. LEE²;

¹Meteor Biotech, Seoul, Korea, Republic of; ²Meteor Biotech, Co. Ltd., Seoul, Korea, Republic of

Abstract: Understanding tissue function and disease pathology requires spatially resolved insights into transcript diversity, regulatory variation, and cell-cell communication. We present an integrative platform combining Spatially-resolved Laser-Activated Cell Sorting (SLACS) with full-length RNA sequencing for region-specific, base-resolution analysis of coding and regulatory RNA features in tissue. SLACS uses image-guided infrared laser ablation to isolate defined regions from stained tissue sections while preserving spatial context. These cells are then subjected to full-length RNA sequencing, allowing detailed analysis of isoform usage, RNA editing, gene fusions, and cell-state markers. We applied this method to immunostained triple-negative breast cancer (TNBC) tissues, isolating epithelial, immune, and stromal compartments. Full-length RNA-seq revealed region-specific expression of immune checkpoint ligands and interferon-stimulated genes. We detected spatially distinct alternative splicing and A-to-I RNA editing events, particularly in ADAR targets. Receptor-ligand co-expression was inferred between tumor and immune cells, and full BCR/TCR sequences were reconstructed to map local immune repertoires. Expressed SNVs and inferred CNVs highlighted somatic heterogeneity across tumor zones. RNA velocity revealed trajectories of epithelial-mesenchymal transition at invasive fronts. By combining spatial targeting with full-length sequencing, SLACS enables multi-layered molecular analysis, such as spanning isoforms, RNA modifications, immune clonotypes, and differentiation states, within histological architecture. This approach offers a powerful tool for investigating spatial heterogeneity and cellular interactions in cancer and other complex tissues.



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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

Location: SDCC Halls B-H

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- NSF Grant 1955981-CIF
- NSF Graduate Research Fellowship 2021324226
- Boston University Federal Work-Study Award

Title: Cellular voltage and calcium imaging reveals real-time changes induced by sustained deep brain stimulation in the awake behaving mouse brain

Authors: *C. RAVASIO, Y. WANG, X. HAN;
Biomed. Engin., Boston Univ., Boston, MA

Abstract: Deep brain stimulation (DBS) is effective for managing various neurological and psychiatric disorders. However, the therapeutic mechanisms of DBS remain unknown. While clinical DBS is traditionally delivered continuously, pre-clinical studies thus far have been limited to brain slice analysis, brief stimulation bouts, and anesthetized animal preparations. To explore the real-time effects of sustained DBS, we first performed single-cell voltage imaging using ElectraOFF over repeated 1 min bouts of DBS. We found that 1-minute-long 40 Hz DBS evoked a brief, 1-second-long membrane voltage (V_m) hyperpolarization, which transitioned to V_m depolarization within 15 s and was sustained throughout the stimulation period. Interestingly, these V_m changes were not accompanied by firing changes, highlighting the pre-synaptic effects of DBS. Of the 49 neurons tested, 27 (55%) displayed significant DBS-evoked V_m entrainment which declined in strength over time within and across successive stimulation trials. To further explore DBS-evoked changes over even greater stimulation durations, we performed cellular calcium imaging using GCaMP7f while delivering local DBS continuously for 39 minutes. We discovered that DBS evokes consistent inhibitory responses that last for a couple seconds at onset. The transient onset response was often followed by a return to pre-stimulation activity levels in many neurons. However, a considerable fraction of neurons exhibited long-lasting inhibition throughout the entire DBS period, suggesting a general inhibitory effect of DBS around the stimulation site. Together, these results reveal that DBS drives diverse responses across single neurons without producing detrimental uniform changes across neuron populations at the stimulation site, and that the evoked population-level effect quickly settles into a new state within seconds of DBS onset. This DBS-mediated response inhibition is consistent with the informational lesion mechanism of DBS.

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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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Title: Transcranial ultrasound stimulation modulates neuronal membrane potentials across broad timescales in the awake mammalian brain

Authors: *E. SAN ANTONIO¹, E. BORTZ¹, J. SHERMAN², H.-A. TSENG¹, X. HAN¹;

¹Biomed. Engin., ²Pharmacology, Physiol. & Biophysics, Boston Univ., Boston, MA

Abstract: Transcranial ultrasound stimulation (TUS) enables noninvasive neuromodulation with submillimeter spatial and millisecond temporal resolution, but its direct effects on single-neuron membrane potential (Vm) in the awake brain remain poorly defined. Here, we used kilohertz-resolution voltage imaging with the genetically encoded voltage indicator SomArchon in awake, head-fixed mice to record cortical Vm responses to 0.35 MHz TUS delivered at pulse repetition frequencies (PRFs) of 10 or 40 Hz. Across 161 neurons, 42.8% showed significant Vm modulation. Among these, the majority exhibited rapid depolarizations within 10 ms of TUS onset, typically without accompanying spiking, indicating synaptic modulation. To assess entrainment, we computed the phase-locking values between TUS pulses and Vm traces. At 10 Hz PRF, 20.8% of neurons showed significant entrainment; at 40 Hz PRF, 12.7% were entrained. We assessed adaptation across trials by computing linear trends in z-scored Vm responses. During the first 250 ms of stimulation, 11.8% of neurons exhibited significant response changes across trials, with most showing progressive reduction in Vm amplitude, suggesting that TUS engages adaptive processes such as synaptic depression, mechanosensitive channel desensitization, or homeostatic plasticity. Simultaneous multi-neuron recordings revealed that TUS modulates population-level synchrony. Both Vm and spiking cross-correlations increased significantly in the post-TUS window compared to pre- or during-stimulation, indicating a network-level reset. TUS evoked diverse Vm response profiles, including rapid depolarization, delayed depolarization, hyperpolarization, and frequency-specific entrainment. Despite heterogeneous responses, the majority of effects were observed within 250 ms of TUS onset, suggesting a temporally restricted window of modulation. Differences in latency and temporal profile between 10 and 40 Hz PRFs highlight how pulse frequency shapes membrane responses, possibly through intrinsic ion channel kinetics. Thus, low-intensity TUS directly modulates cortical neuron Vm with millisecond precision, induces frequency-specific entrainment, and recruits short-term plasticity mechanisms. Post-stimulation increases in synchrony further reveal that TUS can reset network activity. Together, these findings establish a cellular framework for interpreting and optimizing TUS neuromodulation, enabling precise control of single-neuron and circuit-level dynamics in the awake brain.

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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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1RF1NS129520

Title: Voltage imaging analysis of intracranial electrical stimulation effect on inhibitory and excitatory cells in a cortical stroke mouse model

Authors: *M. ABUMUAILEQ¹, P. FIER¹, D. SHAW², L. DANG¹, W. SHI⁵, M. A. KRAMER³, C. J. CHU⁶, X. HAN⁴;

²Grad. Program for Neurosci., ³Dept. of Mathematics and Statistics, ⁴Biomed. Engin., ¹Boston Univ., Boston, MA; ⁵Neurol., Massachusetts Gen. Hospital; Harvard Med. Sch., Boston, MA; ⁶Massachusetts Gen. Hosp. / Harvard Med. S, Cambridge, MA

Abstract: Neuromodulation through intracranial electrical stimulation reduces seizures in patients with epilepsy. However, the neurophysiological mechanisms through which electrical stimulation leads to clinical benefit is unknown. Further, the stimulation parameters that achieve optimal neuronal responses are not known. Efforts to probe these questions using conventional electrophysiological techniques are limited by electrical stimulation induced artifact on electrodes. Accordingly, we performed cellular optical voltage imaging to evaluate the impact of electrical stimulation on cortical excitatory versus inhibitory neurons in a cortical stroke mouse model expressing epileptic biomarkers. Briefly, we induced cortical stroke using a recently established photothrombosis procedure, and expressed Voltron2 in excitatory neurons in the C57BL6 mice or parvalbumin positive (PV+) interneurons in PV-cre mice. Voltage imaging was performed at 0.8kHz using a custom confocal microscope, while mice were head-fixed on a treadmill that permitted voluntary locomotion. During each recording session, 36 electrical stimulation trials were performed, with each trial lasting for 6 seconds, containing 1s baseline and 5s of stimulation. Four stimulation frequencies, 40, 140, 200, or 1000 Hz, in random orders, were tested. The stimulation current amplitudes were manually determined and kept constant across frequencies in each mouse to avoid any noticeable behavioral effects. We found that intracranial electrical stimulation at most frequencies produced robust membrane voltage changes across individual neurons. While 40Hz stimulation robustly entrained neurons, higher frequencies (140, 200, 1000-Hz) resulted in a brief excitatory effect (membrane depolarization and increased firing) followed by profound and long-lasting inhibition (hyperpolarization and firing suppression). Higher stimulation frequencies led to faster excitation at stimulation onset, which also transitioned faster towards inhibition. These results provide a principled understanding of the effect of stimulation frequency on neuronal dynamics that is relevant to epilepsy.

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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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NIH 1R01MH122971
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NSF 2002971-DIOS
NSF 1955981-CIF

Title: Neuronal slow membrane voltage dynamics and spiking differentially engage cytosolic Ca^{2+} activities in the awake mammalian brain

Authors: *Y. WANG¹, H.-A. TSENG¹, S. XIAO¹, E. BORTZ¹, Y. ZHOU¹, A. MARTIN¹, C. PORTER¹, H. MAN², J. C. MERTZ¹, X. HAN¹;
¹Biomed. Engin., ²Biol., Boston Univ., Boston, MA

Abstract: Membrane voltage (V_m) provides a summed measure of a neuron's internal dynamics and its inputs. V_m fluctuates extensively during behavior, which regulates spike timing and cellular learning. However, it is unclear how V_m engages intracellular signaling in the awake mammalian brain. We developed a bicistronic genetically encoded dual indicator SomArchon-P2A-GCaMP7/8, using which we simultaneously recorded V_m and the dynamics of cytosolic Ca^{2+} , a key regulator of cellular plasticity, in individual neurons in rat primary neuron cultures, human iPSC-derived neuron, and awake mice. We found V_m and Ca^{2+} exhibited complex relationship across neurons in CA1, striatum, and visual cortex in awake mice. We found that even though Ca^{2+} events failed to capture most spikes *in vivo*, in sharp contrast to that observed *in vitro*, Ca^{2+} events were accompanied by elevated firing rate. Furthermore, slow V_m depolarization was closely accompanied by Ca^{2+} elevation, indicating that neuronal excitability, but not spike timing, is faithfully translated into cellular changes via Ca^{2+} -mediated pathways. Additionally, individual spikes differentially modulate Ca^{2+} dynamics depending on post-spiking V_m depolarization, demonstrating the distinct impact of different spiking outputs on intracellular environment. Finally, we found that intracranial electrical stimulation, broadly used in clinical neuromodulation, evokes heterogeneous V_m and cytosolic Ca^{2+} changes in CA1 neurons in awake mice, highlighting the translational potential to differentially regulate neuronal output versus cellular learning. Together, these results demonstrate that slow V_m dynamics and spiking differentially engage cytosolic Ca^{2+} activities through cellular mechanisms sensitive to clinical neuromodulation.

Disclosures: Y. Wang: None. H. Tseng: None. S. Xiao: None. E. Bortz: None. Y. Zhou: None. A. Martin: None. C. Porter: None. H. Man: None. J.C. Mertz: None. X. Han: None.

Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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Support: NIH (1RF1NS129520, 1R01MH122971, and 1R01NS115797)
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NSF Graduate Research Fellowship 2021324226

Title: Prolonged single neuron voltage imaging in behaving mammals using a novel green-fluorescent voltage indicator ElectraOFF

Authors: Y. WANG¹, C. RAVASIO¹, Y. ZHOU¹, *X. HAN²;

¹Biomed. Engin., ²Boston Univ., Boston, MA

Abstract: Long-term recording of single-neuron membrane voltage (Vm) dynamics during behavior is highly desirable. We tested ElectraOFF, a fully genetically encoded, photostable, high-performance, green-fluorescent voltage indicator in various cell types and brain regions in behaving mice, including hippocampal pyramidal neurons, striatal cholinergic interneurons, and visual cortical parvalbumin interneurons. We achieved routine cellular-resolution imaging over tens of minutes, occasionally up to eighty minutes, in behaving mice with minimal signal loss across neuron types and brain regions. Using ElectraOFF, we found that visual cortex parvalbumin neurons exhibited a transient Vm depolarization followed by a sustained hyperpolarization upon visual flicker presentation. In striatal cholinergic interneurons, ElectraOFF captured subthreshold delta-rhythmicity in a subset of neurons. In CA1 pyramidal cells, ElectraOFF detected a gradual reduction in Vm entrainment strength during 40Hz intracranial electrical stimulation over each minute long stimulation trial and across repeated stimulation trials. These results demonstrate the broad application of ElectraOFF in capturing single spikes and subthreshold dynamics over prolonged periods during behavior, and highlight the novel insights enabled by prolonged cellular voltage imaging.

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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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Topic: J.04. Physiological Methods

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NIH grant 1RM1NS132981
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Stanford Department of Bioengineering

Title: Large-scale action potential imaging in vivo with a fast and responsive voltage indicator

Authors: *S. LEE¹, G. ZHANG⁵, L. C. GOMEZ⁶, C. DONG², G. TESTA-SILVA⁸, A. Y. HAO³, Y. SUN⁴, R. H. ROTH⁴, D. JIANG⁹, J. B. DING⁴, T. R. CLANDININ², B. ROSKA¹⁰, L. M. GIOCOMO², D. FELDMAN⁷, N. JI¹¹, M. Z. LIN¹²;

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Abstract: Fluorescent genetically encoded voltage indicators (GEVIs) directly report changes in neuronal transmembrane potential with changes in fluorescence intensity. Recently, we reported a positively tuned GEVI, ASAP4e, that exhibits exceptional photostability and responsivity while detecting spikes with similar signal-to-noise ratios (SNRs) as ASAP3. To further enhance optical spike detection, we performed multiparametric structure-guided mutagenesis of ASAP4. The resulting ASAP6 is faster and more responsive than ASAP4 while maintaining high photostability and consistent performance across multiple 1- and 2-photon imaging methods. With ASAP6, we demonstrate kilohertz-rate 1-photon imaging of spiking activity in awake behaving mice at densities exceeding 400 neurons/mm². Directly comparing ASAP6 and the calcium indicator GCaMP8f using frame rates matched to their activation kinetics, we find that ASAP6 detects single action potentials with similar SNR but higher temporal precision than GCaMP8f. Thus, ASAP6 enables large-scale action potential imaging in genetically defined neurons in vivo with improved spike timing accuracy.

Disclosures: S. Lee: None. G. Zhang: None. L.C. Gomez: None. C. Dong: None. G. Testa-Silva: None. A.Y. Hao: None. Y. Sun: None. R.H. Roth: None. D. Jiang: None. J.B. Ding: None. T.R. Clandinin: None. B. Roska: None. L.M. Giocomo: None. D. Feldman: None. N. Ji: None. M.Z. Lin: None.

Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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Program #/Poster #: PSTR308.07/UU16

Topic: J.04. Physiological Methods

Support: 23H05450

Title: Development of a fluorescent imaging device for red fluorescent protein that enables fluorescence imaging in multiple regions

Authors: *Y. OHTA¹, V. G. CASTILLO¹, Y. SUNAGA¹, J. OLOROCISIMO¹, M. OHSAWA², J. OHTA¹;

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Abstract: The development of brain fluorescence imaging technology has been remarkable, and its contributions to the fields of neuroscience, medicine, and pharmacology have been significant. We have been developing a brain-implanted ultra-compact CMOS imaging device as an optical measurement and control device for elucidating brain function. Optical methods, enabled by advances in genetic engineering, allow for the specific observation of different types of neurons, a feature that distinguishes them. Additionally, this device is ultra-compact, allowing for the implantation of multiple devices into a single brain or the simultaneous implantation of multiple modalities, such as microdialysis and optical stimulation, for concurrent measurements. However, when performing fluorescent imaging of multiple individual neurons or combining optical stimulation, it is desirable to use LEDs with different wavelengths for excitation or stimulation. In this study, we have developed an RFP sensor in addition to the existing GFP sensor and demonstrated an example of simultaneous fluorescence imaging. Furthermore, we present a combination of fluorescent imaging and optical stimulation. First, we describe the method for creating the RFP sensor. Here, a red absorption filter was applied as an absorption filter for the imaging area, and a green LED was used as the excitation LED for the red fluorescent protein. Validation experiments were conducted to verify the sensor's functionality for simultaneous fluorescent imaging. The key point here is that the blue LED light used for excitation or light stimulation does not pass through the absorption filter of the RFP sensor. Next, we describe two methods for simultaneous fluorescence imaging. We expressed Flex-GCaMP6s in the VTA of DATcre mice and RGECO in the NAcShell, which releases DA, in the same mice. In vivo experiments involved the administration of alcohol, and we observed dopamine (DA) neural activity in the VTA and NAcShell under free-moving conditions. This method is useful because it allows for the neural separation of GCaMP6 observations across multiple regions. Finally, as an application of optogenetics, we expressed ChR2 in the VTA and GRAB_DAr or RGECO in the NAcShell of mice. We performed fluorescence imaging in the NAcShell after light stimulation in the VTA under free-moving conditions. The combination of this RFP sensor is expected to expand its applications as a probe for observing neural networks, including optogenetics, in the future.

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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR308.08/UU17

Topic: J.04. Physiological Methods

Support: Brown OVPR
Carney Institute

Title: Development of a Bioluminescent Ca²⁺ Indicator to Detect Depolarization-Induced Intracellular Ca²⁺ Signals

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Abstract: Ca²⁺ signaling is essential for myriad cell functions and is used as a proxy for neuronal activity. Fluorescent-based Ca²⁺ indicators are highly sensitive and have little or no background but have certain limitations, including photobleaching, phototoxicity, and requiring activation via an external light source. Bioluminescent (BL) Ca²⁺ indicators are used to detect intracellular Ca²⁺ signals, but relatively low sensitivity and high baseline signals have limited their use in cells. The development of CaBLAM - a genetically encoded BL Ca²⁺ sensor derived from *O. gracilirostris* luciferase - creates new possibilities for using BL-Ca²⁺ indicators in living cells. CaBLAM has the highest contrast (signal relative to baseline) among BL Ca²⁺ indicators, and CaBLAM's contrast is comparable to GCaMP6s and GCaMP8f. To determine the most optimal conditions for using CaBLAM in cells, we evaluated four luciferins: cephalofurimazine (CFz) and fluorofurimazine (FFz) (used *in vivo*), and furimazine (Fz) and h-coelenterazine (h-CTZ) (used *in vitro*). We generated luciferin dose-response curves in mouse N2A cells using a microplate reader and validated with high-sensitivity imaging (EMCCD camera). We found that luciferin concentration, as well as the type of luciferin, influenced signal contrast. In general, the luciferin-induced CaBLAM BL signal increased with luciferin concentration but, after a certain concentration, was accompanied by a decrease in the relative size of the Ca²⁺-dependent BL signal. Our data suggest that CaBLAM signals are higher in contrast and brightness (peak BL signal relative to protein concentration) with luciferins FFz and Fz, as compared to CFz and hCTZ. We are now applying CaBLAM to detect depolarization-induced intracellular Ca²⁺ signals in the proximity of voltage-gated Ca²⁺ (Ca_v) channels. We generated four CaBLAM constructs: i) Myristolated-CaBLAM to anchor to the plasma membrane; ii) CaBLAM-Ca_v3.3 and Ca_v2.2-CaBLAM placing CaBLAM at the N or C-termini of these channels, respectively; and iii) β₃-CaBLAM, which localizes CaBLAM to Ca_v1 and Ca_v2 channels which form complexes with cytoplasmic β₃ subunits. Using electrophysiological recording, we also showed that tethering these moieties and proteins to CaBLAM had little effect on Ca_v2.2, Ca_v3.3, and β₃ function. All constructs generate BL in response to ionomycin-induced Ca²⁺ influx when expressed in N2a cells and β₃-CaBLAM also responds to KCl-driven depolarization. CaBLAM can be tethered to Ca_v channels without impacting their basic biophysical function in N2a cells and it can detect intracellular Ca²⁺ signals in response to membrane depolarization.

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Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

Location: SDCC Halls B-H

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Program #/Poster #: PSTR308.09/UU18

Topic: J.04. Physiological Methods

Title: Lighting Up Inflammation: Challenges in Translating Luminol IVIS Imaging from Mice to Rats

Authors: *C. PRIDDY¹, B. L. ADAMS², M. CASTANARES³, S. DUBOIS⁴;
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Abstract: In vivo imaging systems (IVIS) provide a non-invasive and highly sensitive platform for monitoring biological processes in real time, enabling longitudinal studies without the need for terminal procedures. These systems can utilize tagged fluorophores along a diverse spectrum to visualize molecular and cellular dynamics associated with inflammation in live animal models. Fluorophore tracers have been developed to monitor many aspects of the inflammatory process, from neutrophil infiltration, macrophage activity, to vascular leakage, and oedema. Luminol is widely utilized as a bioluminescence marker for in vivo imaging due to its ability to emit light upon oxidation, typically in the presence of reactive oxygen species (ROS). This property makes luminol particularly valuable for imaging inflammatory processes, as elevated ROS production is a hallmark of neutrophil infiltration. When administered systemically, luminol can highlight sites of acute inflammation, enabling real-time, non-invasive detection of immune responses in live animal models. Its rapid signal generation, compatibility with standard in vivo imaging systems, and ability to reflect dynamic changes in oxidative stress make luminol a practical and informative tool for studying inflammation, infection, and related processes. Although luminol-based IVIS imaging is well established in mouse models of inflammation, reports of its application in rats are limited. In this study, we optimized luminol IVIS imaging parameters using the complete Freund's adjuvant (CFA) model of inflammation. In mice, intraperitoneal (IP) administration of luminol up to 150 mg/kg was well tolerated and produced strong, localized bioluminescent signals at sites of inflammation with minimal background. In contrast, Sprague Dawley rats showed no detectable signal in the inflamed paw following IP or intravenous (IV) administration, though diffuse signal was observed in the body. Intraplantar delivery of luminol successfully generated localized signal in the CFA-injected rat paw, with no signal in the untreated control. This signal was attenuated by the anti-inflammatory drug naproxen, mirroring findings in mice. These results suggest that systemic distribution of luminol differs significantly between rats and mice, presenting challenges for in vivo bioluminescence

imaging (BLI) in rat models. To address this, we evaluated IV administration of a near-infrared (NIR) tracer in the rat CFA model and observed strong inflammatory signal with reduced background compared to luminol. While more costly, NIR tracers may offer a more effective alternative for inflammatory imaging in rat models.

Disclosures: **C. Priddy:** A. Employment/Salary (full or part-time): Lilly Research Laboratories. **B.L. Adams:** A. Employment/Salary (full or part-time): Lilly Research Laboratories. **M. Castanares:** A. Employment/Salary (full or part-time): Lilly Research Laboratories. **S. Dubois:** A. Employment/Salary (full or part-time): Lilly Research Laboratories.

Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR308.10/UU19

Topic: J.04. Physiological Methods

Title: Recent advances in random access projection microscopy

Authors: ***M. HE**¹, M. E. XIE^{1,2}, G. JAINDL³, R. GIBISER³, M. SEYEDOLMOHADESIN¹, K. PODGORSKI¹;

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Abstract: Emerging high-performance voltage and neurotransmitter indicators can directly report how signals flow through neural circuits with submicron spatial and millisecond temporal resolution in the living brain. However, recording rapid signals from these indicators in many targets at once is limited by existing in vivo microscopes. To address the challenge, we developed an optimized two-photon microscope (SLAP2) based on a flexible scan system that efficiently scans arbitrary regions of interest with no per-target access time cost, addressing up to >1000 dispersed targets at >1000 Hz frame rate with >99% duty cycle, and can integrate multiple pixels into individual measurements to increase imaging speed. SLAP2 was developed to be widely disseminated, and includes high-performance optical hardware, stable alignment, full-featured control software, online 3D motion correction with <150 nm error, real-time excitation power monitoring, and support for closed loop experiments capable of inducing reinforcement learning on the single-neuron level. The latest iteration is powered by a first-of-its-kind 150 W, 100 fs ultrafast laser. The system's diffraction and shot-noise limited imaging performance can be further enhanced with plug-and-play adaptive optics. SLAP2 also includes a one-photon imaging path for targeted illumination which enables simultaneous imaging of neurotransmitters and somatic voltage. To demonstrate SLAP2's unique capabilities, we recorded simultaneous activity from 200-400 excitatory synapses per neuron at >100 Hz with high resolution scanning, up to 1000 synapses per neuron at >100 Hz with volumetric integration scanning, and subthreshold membrane potentials in dendrites at >2kHz using integration

scanning. These results demonstrate the utility of SLAP2 for direct observation of rapid subthreshold dynamics involved in neural computations.

Disclosures: **M. He:** None. **M.E. Xie:** None. **G. Jaindl:** A. Employment/Salary (full or part-time): MBF Bioscience LLC. **R. Gibiser:** A. Employment/Salary (full or part-time): MBF Bioscience LLC. **M. Seyedolmohadesin:** None. **K. Podgorski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MBF Bioscience LLC.

Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

Location: SDCC Halls B-H

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Topic: J.04. Physiological Methods

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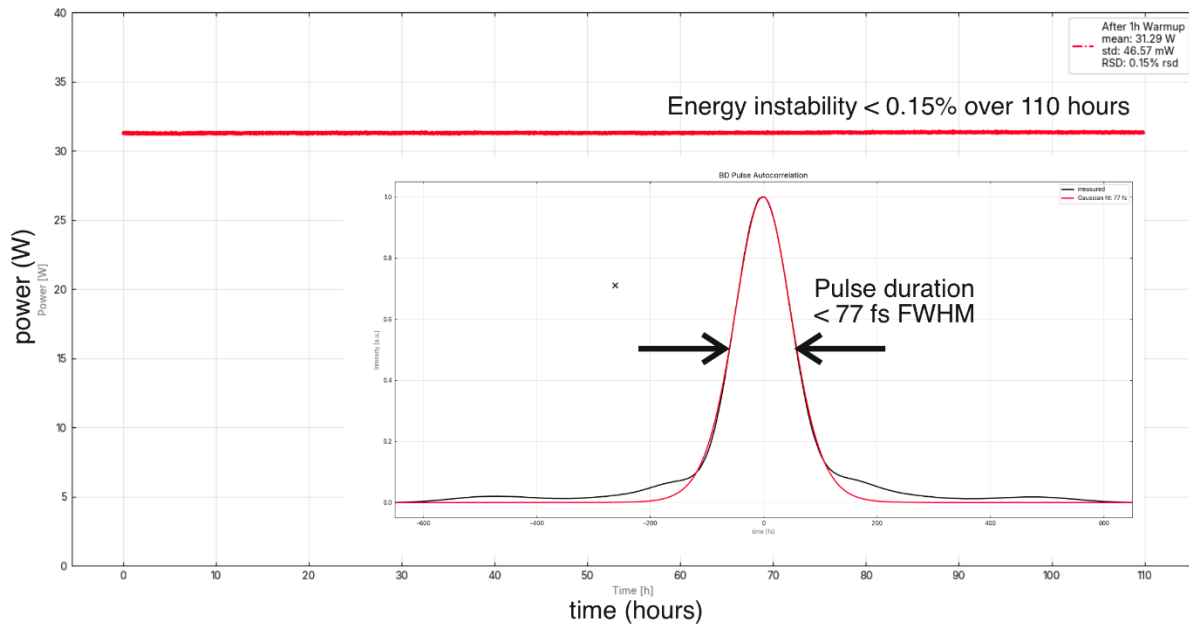
Title: Two-photon voltage imaging with sub-100 fs, 40 μ J, 1 MHz laser system

Authors: I. ABDALLAH¹, E. PAPAGIAKOUMOU², R. SIMS³, M. SCHULZ¹, I. BENDIFALLAH⁴, K. KOLATA¹, ***R. RIEDEL**¹, V. EMILIANI⁵;

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Abstract: Two-photon (2P) voltage imaging uniquely combines the cellular resolution of optical microscopy with the millisecond precision of electrophysiology, enabling direct visualization of action-potentials and sub-threshold membrane events that are inaccessible to calcium reporters, such as GCaMP. However, the limited number of membrane bound indicators and necessary kilohertz imaging rates impose demanding photon budget requirements, 10-50 $\times$ higher than calcium imaging, and even steeper barriers when moving to volumetric recordings. We present a turnkey ultrafast laser source that delivers 40 μ J pulses at 1 MHz (40 W average power) and 1030 nm after integrated nonlinear compression, retaining < 100 fs duration and $M^2 < 1.1$. Relative to an 80-MHz Ti:Sapphire oscillator, the 40-fold pulse-energy gain increases 2P excitation efficiency for red-shifted GEVIs by ≈ 15 -fold while remaining below ANSI tissue-heating limits. The laser system shows < 0.15 % RMS power drift over 110 hours, supporting day-long imaging sessions. We will demonstrate the capabilities of this system by performing scanless two-photon voltage imaging in cortical layer 2/3. By concentrating the photon budget into fewer, high-energy pulses, the presented laser architecture enables SNR gains and increased

volumetric imaging rates, paving the way for cortical-column-scale voltage recordings in behaving animals.



Disclosures: **I. Abdallah:** A. Employment/Salary (full or part-time): Class 5 Photonics GmbH. **E. Papagiakoumou:** None. **R. Sims:** None. **M. Schulz:** A. Employment/Salary (full or part-time): Class 5 Photonics GmbH. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); lass 5 Photonics GmbH. **I. Bendifallah:** None. **K. Kolata:** A. Employment/Salary (full or part-time): Class 5 Photonics GmbH. **R. Riedel:** A. Employment/Salary (full or part-time): Class 5 Photonics GmbH. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Class 5 Photonics GmbH. **V. Emiliani:** None.

Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

Location: SDCC Halls B-H

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Program #/Poster #: PSTR308.12/UU21

Topic: J.08. Methods to Modulate Neural Activity

Support: GINOP_PLUSZ-2.1.1-21-2022-00143
2021-1.1.4-GYORSÍTÓSÁV-2022-00064

Title: Fast 3D real time motion correction for in vivo two-photon imaging of motile biological samples

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Abstract: In-vivo measurements are inherently hampered by the motion of behaving animals, for example using voltage sensors when the signal is acquired from small regions of interest to get the maximal temporal precision or extreme and/or unpredictable motion range such as zebrafish or beating heart. Here we present an FPGA-based, acousto-optical solution for 3D online motion correction that enables signal extraction at a high temporal resolution of 100 kHz per point on a volume of 900x900x400 μm (1000x1000x444 px), for compensation of 1-100 Hz of motion with a residual motion under 1 μm at proper signal-to-noise ratio. The correction can be combined with all scanning modes, including point-scan, squared and ribbon-based scanning types for population and dendritic scanning, as well as raster and volume scan types (i.e. Z-Stack). The use of the closed-loop online motion compensation was validated in vivo with ~ 3 kHz JEDI-2P sensor voltage imaging of Parvalbumin neurons in cortex of awake mice, and via acquisition of multi wavelength excitation 6D datasets (2 color + 3 space + time) in cortex of awake mice. Here we further demonstrate that a huge z motions (80 to 120 μm) can be eliminated in amphibian heart. This can be open new horizons in a comprehensive analysis of the heart's physiological behavior in real-time.

Disclosures: D. Palfi: None. V. Kiss: None. P.L. Feyen: None. J.W. Herms: None. A. Losonczy: None. B. Rozsa: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Femtonics Ltd., CEO.

Poster

PSTR308: Optical Methodology: Application: Optical Functional Imaging: New Probes and Illumination Methods

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Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR308.13/UU22

Topic: J.08. Methods to Modulate Neural Activity

Support: NSF 2123830

Title: In Vivo Optical Control of Oxytocin Using Photoswitchable Azosomes

Authors: *J. TAURA¹, H. TAJARENEJAD², L. NAHAR¹, X. YU³, Z. QIN², P. A. SLESINGER¹;

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Abstract: Precise spatiotemporal control over release of neuroactive compounds remains a significant challenge in neuroscience. Many neurons express more than one neurotransmitter or neuromodulator, making electrical stimulation of release difficult to interpret. We developed and validated a novel class of light-sensitive liposomes, or "azosomes," using a photoswitchable lipid. Azosomes are constructed with azobenzene-phosphatidylcholine (Azo-PC), which can release cargo through UV (365 nm) photostimulation of Azo-PC (*Xiong et al., Nano Research, 2023*). *In vitro*, we demonstrated that azosomes loaded with a D1 receptor agonist (SKF-81297) could repeatedly activate primary striatal neurons following photostimulation. Here, we extend this technology to *in vivo* applications by combining azosome infusion through an implanted optofluidic cannula targeting the dorsomedial striatum (DMS) and triggering its photorelease in freely behaving mice. Using calcein-loaded azosomes, we achieved light-controlled, localized release after more than one hour of infusion, demonstrating the stability of azosomes in native tissue. Varying the power and duration of the light pulse allowed us to titrate the release, demonstrating the temporal control of the technology. Importantly, the calcein fluorescence (λ_{ex} , 495 nm) was spectrally compatible with jRGECO1a (λ_{ex} , 570 nm), a red genetically encoded calcium indicator, enabling simultaneous monitoring of GAD2+ medium spiny neuron (MSN) activity and azosome release *in vivo*. We further adapted this system to deliver neuropeptides by encapsulating oxytocin (OT) in azosomes. Using MTRIA-OT (*Ino et al., Nature Methods, 2022*), a genetically encoded fluorescent sensor for oxytocin, we validated the specificity of OT-azosome photorelease in the DMS, showing that release occurred only upon UV illumination and not during infusion. Coinfusion with CalceinRED enabled visualization during infusion. Moreover, we successfully combined photorelease of OT with simultaneous monitoring of MSN activity using jRGECO1a expressed in neurons of freely behaving animals. We are currently applying this approach to behavioral paradigms that involve oxytocin, such as social recognition tests. We are also exploring release in other oxytocin-receptor-rich brain regions (e.g., AON, medial amygdala) and targeting the lateral ventricles to study widespread neuromodulatory effects of photo-released oxytocin. Together, these results demonstrate the versatility and precision of azosomes as a neuromodulation tool, enabling temporally resolved molecular interventions during behavior.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.01/VV1

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: A novel mouse model for *in vivo* evaluation of anti-human TFR1 antibody penetration across blood-brain barrier

Authors: M. ZHANG, C. LI, F. GU, J. GUO, *J. HSU, X. ZHOU;
Biocytogen, Waltham, MA

Abstract: Delivering large-molecule drugs into the brain and muscles is highly challenging due to several barriers and structural limitations. In the brain, the blood-brain barrier (BBB)—formed by endothelial cells with tight junctions—significantly restricts the passage of large molecules from the bloodstream. In muscles, the structure of muscle cells themselves hinders large-molecule drug entry due to low permeability and tight intercellular junctions.

Biocytogen has successfully developed a humanized TFR1 mouse model via genomic knock-in, named B-hTFR1 mice, which provides a promising solution to overcome these barriers and facilitate the delivery of large-molecule drugs into the brain, muscles, and eyes. Humanized TFR1 expression was exclusively detected in erythroid cells, spleen, heart, liver, lung, kidney, colon, stomach, skeletal muscle, and brain microvascular endothelium in B-hTFR1 mice. The proportions of various immune cells in the spleen, blood, and lymph nodes—as well as blood biochemistry and hematology parameters—were comparable to those of wild-type C57BL/6 mice.

In vivo pharmacokinetic evaluation of anti-human TFR1 bispecific antibodies (BsAbs) showed that the BsAbs exhibited higher serum clearance and enhanced brain exposure post-dosing, confirming that B-hTFR1 mice enable uptake of intravenously administered anti-human TFR1 BsAbs into the brain. By mating B-hTFR1 mice with B-hTAU mice, the resulting B-hTFR1/hTAU mice enabled brain delivery of therapeutics, and hTAU mRNA levels in the treatment group showed a significant decrease. Similarly, mating B-hTFR1 mice with B-hDMPK mice generated B-hTFR1/hDMPK mice, which allowed the delivery of intravenously administered antibody-oligonucleotide conjugate (AOC) drugs into muscle tissue, with a significant reduction in hDMPK mRNA levels observed in the treatment group.

In conclusion, B-hTFR1 mice provide a powerful preclinical model for in vivo evaluation of the effective delivery of protein therapeutics to the brain and muscle.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.02/VV2

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: JPSP KAKENHI (22H02786)
AMED (23ak0101192h0001)

Title: Development of brain delivery technology for drugs of CNS diseases using BBB-permeable cyclic peptide fused Fc protein

Authors: ***K. KURASHIKI**¹, M. OHTSUKA², S. KAMEOKA², S. OHTSUKI³, S. ITO⁴;
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Life Sci., ³Fac. of Life Sciences, Kumamoto Univ., Kumamoto, Japan

Abstract: The blood-brain barrier (BBB) severely restricts the delivery of therapeutics to the central nervous system (CNS), which remains a major challenge for the treatment of CNS disorders. We previously identified a BBB-permeable cyclic peptide (C-SLSHSPQ-C, with a 1C-9C disulfide bond, referred to as SLS-peptide) that facilitates the brain distribution of macromolecules across the BBB, and demonstrated that the fusion of this peptide to a monoclonal antibody (mAb-SLS) significantly increased brain delivery. However, achieving efficient brain delivery for various CNS therapeutics still requires platforms beyond conventional mAb. The Fc protein offers a smaller molecular size compared to mAbs, while maintaining a similar plasma half-life and the flexibility for modification with various therapeutic molecules, making it ideal for drug delivery applications to the brain. In this study, we aimed to investigate the effect of fusing the SLS-peptide to the Fc protein on its brain distribution in mice. To modify the Fc protein, the SLS peptide was fused to its C-terminus via a GS3 linker (Fc-GS3-SLS) or inserted into a specific internal site within the Fc protein (Fc-SLS). Production of Fc-GS3-SLS, Fc-SLS, and unmodified Fc protein in CHO cells resulted in comparable yields and purity, and SDS-PAGE analysis confirmed the expected structures, indicating successful production of each Fc protein variants. These Fc protein variants were subsequently administered intravenously (2 mg/kg), and their concentrations in the brain and serum were quantified by ELISA for up to 24 hours. The brain concentration of Fc-SLS peaked at 1 hour, showing a significant 4.8-fold increase compared to that of mAb-SLS, and remaining 4.1-fold higher at 24 hours post-administration. Fc-GS3-SLS also exhibited a significant 3.4-fold increase in brain delivery compared to mAb-SLS at 1 hour, this effect decreased to 1.7-fold at 24 hours. Importantly, serum concentrations of Fc-GS3-SLS, Fc-SLS and unmodified Fc protein comparable throughout the 24-hour period, indicating that SLS fusion does not compromise serum stability or systemic exposure. These findings indicate that the insertion of SLS into the Fc protein enables efficient and sustained brain delivery, surpassing that of SLS-fused mAb, without affecting Fc protein serum stability. Our results suggest that SLS fusion to the Fc protein represents a promising platform technology for CNS drug delivery.

Disclosures: **K. Kurashiki:** None. **M. Ohtsuka:** None. **S. Kameoka:** None. **S. Ohtsuki:** None. **S. Ito:** None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

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Program #/Poster #: PSTR309.03/VV3

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: JPSP KAKENHI (22H02786)
AMED (23ak0101192h0001)

Title: Enhanced blood-brain barrier penetration of anionic liposomes by SLS peptide modification in an in vitro human BBB model

Authors: *N. WATANABE¹, S. CHIKAMATSU², K. SASAKI², S. OHTSUKI³, S. ITO⁴;
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Abstract: Liposomes are promising nanocarriers for delivering therapeutic agents to the brain; however, their transport across the blood-brain barrier (BBB) remains a major challenge for central nervous system (CNS) drug delivery. We previously identified a BBB-permeable cyclic peptide (C-SLSHSPQ-C, 1C-9C disulfide bond), termed the SLS peptide, which facilitates the transport of macromolecules across the BBB. Therefore, surface modification of liposomes with the SLS peptide is expected to enhance their BBB permeability. In this study, we aimed to optimize the surface modification of anionic liposomes with the SLS peptide through physicochemical characterization and in vitro evaluation of BBB permeability. Anionic liposomes were prepared by the thin-film hydration method with egg phosphatidylcholine (EPC), cholesterol (CHOL), dihexadecyl phosphate (DCP), and the fluorescent dye DiO (EPC/CHOL/DCP = 80/10/10, molar ratio; total lipid concentration: 20 mM). The resulting liposomes exhibited an average particle size of 148 nm, a zeta potential of -39 mV, and a polydispersity index (PDI) of 0.23. SLS peptide-modified liposomes were obtained by incubating the anionic liposomes with stearic acid-conjugated SLS peptide (SLS-STR) at 37°C for 1 hour. Increasing the concentration of SLS-STR led to a dose-dependent increase in zeta potential, an effect not observed with SLS peptide or stearic acid alone, indicating that SLS-STR was successfully incorporated onto the surface of the liposomes. The positive shift in zeta potential may be attributable to an increased distance between the particle surface and the slipping plane caused by SLS-STR. Among the tested conditions, liposomes modified with 0.250 mM SLS-STR maintained anionic properties (zeta potential: -12 mV) while exhibiting the largest increase in zeta potential. The BBB permeability of these formulations was evaluated using a human multicellular spheroidal BBB (hiMCS-BBB) model. Compared to unmodified liposomes, SLS-STR-modified liposomes showed a time-dependent increase in permeation across the hiMCS-BBB model. These results demonstrate that SLS-STR-modified anionic liposomes are promising BBB-permeable nanocarriers for brain-targeted drug delivery and represent a potential platform for CNS therapeutics.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.04/VV4

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NIH DP1 Pioneer Award
K. Lisa Yang and Hock E. Tan Center for Molecular Therapeutics at MIT
NIH R01 (R01MH085802)

Title: AAV-guided tissue-specific delivery platform for synthetic nanoparticles and large genes

Authors: *K. NAGAO¹, T. OSAKI², M. SUR³, R. MACFARLANE⁴, P. ANIKEEVA⁴;
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Abstract: Molecular therapeutics including CRISPR-Cas9 systems and magnetic nanotransducers for remote neuromodulation show significant potential, yet their clinical translation remains hindered by inadequate delivery technologies. Current approaches cannot effectively deliver large genes (e.g. full gene-editing cassettes for SpCas9, ~5.4 kbp) and functional nanomaterials (e.g. 24 nm magneto-thermal iron oxide nanoparticles) to target tissues; a challenge that must be addressed to unlock their therapeutic potential. Here, we introduce AAV-To-Therapeutics Adaptive Conjugation Homing (ATTACH), a versatile delivery platform where molecular and nanoscale therapeutic entities (e.g. gene-carrying vectors, nanomaterials, molecules) are conjugated to adeno-associated viruses (AAVs) to exploit their natural tissue tropism. ATTACH employs fast click chemistry for conjugation, with precise control achieved through quencher molecules introduced directly into the reaction to ensure controlled conjugation. We demonstrate ATTACH's versatility through two complementary strategies. First, AAV-nanoparticle chimeras precisely recapitulate the tropism of intact AAVs in vitro and in vivo. Brain-targeting chimeras achieve brain delivery in mice with the efficiency of ~10% of injected dose per brain mass, a 10-fold improvement over previous strategies (*bioRxiv*: doi.org/10.1101/2025.04.04.647267). The organ-specificity of the delivery is enhanced by increasing AAV to nanoparticle conjugation ratios. Second, AAV-AAV conjugates address fundamental limitations of AAV vectors, the size limit on their transgenes and the low reliability in co-transduction, by enabling robust co-delivery of multiple transgenes. AAV-AAV conjugates demonstrate enhanced co-transduction efficiency in vitro. When combined with split-intein systems for SpCas9 delivery, these conjugates exhibit significantly higher gene editing efficiency compared to conventional dual AAV approaches. ATTACH therefore establishes a new paradigm for targeted delivery of otherwise undeliverable therapeutics, with broad implications for translational medicine.

Disclosures: K. Nagao: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MIT. T. Osaki: None. M. Sur: None. R. Macfarlane: None. P. Anikeeva: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MIT.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.05/VV5

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: A novel AAV capsid targeting the human transferrin receptor 1 permits efficient blood-brain barrier (BBB) crossing to achieve widespread neuronal transduction throughout the CNS

Authors: ***B. P. HEITHOFF**, E. FIRNBERG, S. TEJADA, J. B. SMITH, O. DANOS;
Gene Therapy Res., REGENXBIO, Inc., Rockville, MD

Abstract: The adeno-associated viral vector AAV9 has been used to deliver therapeutic transgenes to the CNS to treat neurological diseases via direct injection into the brain's parenchyma, administration to the cerebrospinal fluid, or by penetration of the blood-brain barrier (BBB) following intravenous (IV) delivery. While less invasive, the IV delivery route has very low efficiency and requires high doses of AAV9 to achieve therapeutic levels of transgene expression in the CNS, increasing the risk of undesirable immune response and liver toxicity. To improve CNS transduction following IV delivery, capsid engineering efforts have recently focused on targeting known BBB-trafficking receptors to shuttle AAVs across the barrier, resulting in broader brain-wide transduction. Here, we utilized REGENXBIO's proprietary *NAVIGATE* directed evolution platform to identify a novel AAV9 variant (AAV9.P1) with a VR-VIII peptide insertion that binds the human transferrin receptor 1 (hTfR1), a clinically validated target known to shuttle cargo across the BBB.

Following IV delivery of an AAV9.P1 vector into humanized hTfR1 knock-in mice, we observed broad and robust transduction of the brain and spinal cord, resulting in more than 28-fold higher vector genome copies (vg) per cell and more than 44-fold higher mRNA transcripts in whole brain homogenates compared to AAV9. Histological analysis showed that the AAV9.P1 vector efficiently transduced regions throughout the entire brain, notably in thalamus, striatum, and cortex. Importantly, this vector primarily transduced neurons in these regions, compared to AAV9, which mainly transduced astrocytes. We further confirmed transgene expression in several disease-relevant neuronal subtypes, including parvalbumin interneurons in the cortex and dopaminergic neurons in the substantia nigra. The spinal cord was also strongly transduced, including cholinergic motor neurons. In addition to increased CNS tropism, we observed a 5-fold reduction in liver biodistribution of AAV9.P1 compared to AAV9, lessening the off-target profile. The AAV9.P1 BBB penetrant capsid offers a viable tool for non-invasive gene therapy in the CNS for a wide array of neurological diseases.

Disclosures: **B.P. Heithoff:** A. Employment/Salary (full or part-time): REGENXBIO, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); REGENXBIO, Inc. **E. Firnberg:** A. Employment/Salary (full or part-time): REGENXBIO, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); REGENXBIO, Inc. **S. Tejada:** A. Employment/Salary (full or part-time): REGENXBIO, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); REGENXBIO, Inc. **J.B. Smith:** A.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.06/VV6

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: New Frontiers in Research Fund
Canadian Institutes of Health Research
Toronto Dementia Research Alliance
BranchOut Neurological Foundation
University of Toronto

Title: Gene delivery and regulation in the brain of mixed-strain mice: impact of the LY6A receptor on AAV.PHP.eB transduction and focused ultrasound on GFAP-driven transgene expression.

Authors: *C. L. DIBIA¹, N. VACARESSE², D. YURTSEVER², L. RUBIO ATONAL², B. LAURETTE³, K. HYNENEN⁴, I. AUBERT⁵;

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Abstract: Our long-term goal is to develop non-invasive and regulatable gene therapy for neurodegenerative disorders. Recombinant adeno-associated viruses (AAVs), such as AAV.PHP.eB, are gaining attention as non-invasive gene delivery vehicles to the brain. Their ability to cross the blood-brain barrier hinges on the Ly6A receptor, whose expression levels differ among mouse strains. For example, the transduction efficiency of the AAV.PHP.eB is higher in C57BL/6J compared to C3H mice, presenting with the Ly6A^{b/b} and Ly6A^{a/a} haplotypes, respectively. Firstly, we sought to evaluate the transduction efficiency of the AAV.PHP.eB in a mouse model derived from mixed C57/C3H strains. Secondly, we aimed to regulate transgene expression under the control of the glial fibrillary acidic protein (GFAP) using focused ultrasound (FUS). In one cohort, 2-month-old mice of different strains and Ly6A haplotypes (C57BL/6J Ly6A^{b/b}, C57/C3H Ly6A^{b/b}, C57/C3H Ly6A^{a/b}) were injected intravenously (*i.v.*) with 1.67E13 GC/Kg AAV.PHP.eB.GFAP.GFP. One month later, brains and peripheral organs (liver, kidney, lungs) were isolated and analyzed by immunohistochemistry (IHC) for GFP and LY6A protein expression and digital droplet PCR for GFP delivery at the DNA level. In another

cohort, mice received AAV.PHP.eB.GFAP.GFP *i.v.*, and one month later treated with FUS unilaterally to the cortex and hippocampus. The corresponding contralateral regions served as controls. Brains were harvested one-week post-FUS, when GFAP immunoreactivity and astrocytic reactivity are known to peak, and analyzed by IHC to quantify the expression of the transgene, GFP, and GFAP. Our results confirmed that, while LY6A receptor expression was lower in mice of Ly6A^{a/b} in the first cohort, gene delivery of the AAV.PHP.eB to the brain was efficient in the mice of both the Ly6A^{b/b} and Ly6A^{a/b} haplotype, displaying strong GFP expression. In the brain GFP expression in astrocytes was robust, while limited GFP was detected in peripheral organs. The ability of the AAV.PHP.eB to transduce astrocytes is critical to render the regulation of the GFAP promoter by FUS possible. In conclusion, the AAV.PHP.eB is a vector of choice for brain penetrance in mice of both Ly6A^{b/b} and Ly6A^{a/b} haplotypes. This offers the opportunity to use such a vector in a mouse model of Alzheimer's disease, commonly known as TgCRND8 mice. The potential of the GFAP promoter to regulate transgene expression in response to astrocytic reactivity induced either by amyloidosis or FUS offers a promising approach for regulatable gene therapies for Alzheimer's disease.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.07/Web Only

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NeuroOne

Title: Translational sEEG-based Platform Devices for Neural Recordings, Ablation and Drug Delivery into the Brain - from Small to Large Animals

Authors: G. ZEPEDA¹, M. PORTO CRUZ², M. VOMERO³, S.-Y. CHANG⁵, J. KIM⁶, F. MIVALT⁶, A. CHAVEZ¹, M. MCNEIL¹, N. KALIA¹, G. A. WORRELL⁷, C. DIAZ-BOTIA¹, *A. KULLMANN⁴;

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Abstract: Stereoelectroencephalography (sEEG) evaluation is part of the diagnosis for patients with drug-resistant epilepsy. The sEEG electrodes are FDA-cleared for temporary (<30 days) neural activity monitoring, recording and stimulation. In 2023, an FDA-cleared radiofrequency ablation (RFA) feature was added to the sEEG platform, enabling real-time, temperature-controlled focal ablations. We are advancing this platform by developing an sEEG-based drug delivery system designed for convection-enhanced delivery (CED). Three device sizes have been developed to accommodate brain sizes across species, from rodents to large animals and humans.

Methods. Device characterization was performed using electrochemistry. CED was tested in 0.6% agarose gel. In vivo testing was conducted in rodents and swine. sEEG electrodes were stereotactically implanted into the hippocampus (HC) and putamen (PUT). Penicillin (5000 units/ μ L) was infused to modulate neural activity. Diffusion volume (Vd) was evaluated using MRI with gadolinium (Gd) contrast in swine and histological analysis in rodents. **Results.** Bench testing confirmed impedance ranges suitable for in vivo applications. CED was demonstrated at infusion rates of 0.5-15 μ L/min and infused volumes (Vi) of 60-300 μ L, with Vd/Vi ratio of 2.7-3.4. Penicillin infusion reliably induced seizure-like activity in both rodent and swine hippocampi. MRI demonstrated localized Gd diffusion in swine PUT and HC, while histological analysis confirmed confined Vd in rodent brain tissue. **Conclusions.** This integrated sEEG-based platform, capable of neural recording, stimulation, ablation, and targeted drug delivery, enables cross-species translational research using a single device. Its scalable design supports consistency in evaluating drug efficacy, dosing, toxicity, and mechanisms of action from preclinical to clinical stages. This approach may reduce late-stage drug development failures and improve therapeutic outcomes. Moreover, combining multiple functionalities in one sEEG-based device platform has the potential to increase the accuracy of diagnosis and provide treatment within one single surgical procedure.

Disclosures: **G. Zepeda:** A. Employment/Salary (full or part-time): NeuroOne. **M. Porto Cruz:** A. Employment/Salary (full or part-time): NeuroOne. **M. Vomero:** A. Employment/Salary (full or part-time): NeuroOne. **S. Chang:** None. **J. Kim:** None. **F. Mivalt:** None. **A. Chavez:** A. Employment/Salary (full or part-time): NeuroOne. **M. McNeil:** A. Employment/Salary (full or part-time): NeuroOne. **N. Kalia:** A. Employment/Salary (full or part-time): NeuroOne. **G.A. Worrell:** None. **C. Diaz-Botia:** A. Employment/Salary (full or part-time): NeuroOne. **A. Kullmann:** A. Employment/Salary (full or part-time): NeuroOne.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.08/VV7

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Enhancing the Efficacy of Low-Dose Doxorubicin Using Focused Ultrasound: A Strategy to Reduce Toxicity

Authors: *C. SONG¹, J. PARK², W. CHANG³;

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Abstract: Introduction Doxorubicin is a first-generation cytotoxic anticancer agent that exhibits strong therapeutic efficacy but also induces significant toxicity by damaging normal cells in addition to tumor cells. Therefore, this study aims to reduce the dose of Doxorubicin below the standard clinical level (5.67 mg/kg) and combine it with Focused Ultrasound (FUS), with the

goal of maintaining the anticancer efficacy of the standard dose while minimizing adverse effects such as systemic toxicity.

Materials and Methods Before initiating in vivo experiments, the cytotoxicity of Doxorubicin was first evaluated in vitro. Cell viability at various concentrations was assessed using the Cell Counting Kit-8 (CCK-8). Subsequently, a glioblastoma model was established by implanting C6 cells into the frontal cortex of rats. The animals were divided into three groups: control, Doxorubicin-only, and Doxorubicin combined with Focused Ultrasound (FUS). In vivo drug delivery and distribution of Doxorubicin were monitored using magnetic resonance imaging (MRI).

Results Based on the IC₅₀, the concentration range of Doxorubicin was further refined in vitro. Apoptosis level shifts were used to determine the timing of repeated ultrasound application in the in vivo experiments. In vivo, co-administration of Focused Ultrasound (FUS) with a reduced dose of Doxorubicin resulted in lower toxicity to normal cells compared to Doxorubicin monotherapy, while effectively reducing tumor volume.

Conclusions This study demonstrated that using Focused Ultrasound (FUS) allows for a reduced dose of Doxorubicin while maintaining anticancer efficacy comparable to the standard clinical dose and minimizing toxicity to normal cells. These findings suggest that FUS-assisted combination therapy has the potential to maximize therapeutic outcomes while minimizing side effects. Furthermore, this approach may be applicable to a wider range of treatment scenarios beyond conventional protocols.

Disclosures: C. Song: None. J. Park: None. W. Chang: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.09/VV8

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Focused ultrasound enabled biomarker discovery

Authors: *S. KOSTENSE¹, S. COTTON², M. BSIBSI³, M. SAMBA MONDONGA², S. AUTHIER⁴, Z. NOROOZIAN⁵;

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Abstract: Treatment of neurological diseases is hampered by the blood brain barrier (BBB) which limits the bioavailability of large biotherapeutics in brain tissue. Focused Ultrasound (FUS) is a technology able to transiently open the blood brain barrier and is currently being developed to enable delivery of biotherapeutics to the brain. In addition, opening the BBB allows CNS derived biomarkers to enter the circulation, facilitating the detection in blood samples. Here we report on a NHP pilot study in which FUS is utilized to deliver AAV to the brain, and to

simultaneously allow biomarkers to diffuse from the CSF into circulation. Serum samples were taken just before and just after FUS, and at days 2, 5, 15, 25 and 57. Samples were analysed for Neurofilament light (NfL) as a model biomarker to test the hypothesis that FUS would increase serum levels shortly after FUS. Additional aliquots were reserved for proteomics as a biomarker discovery tool. Directly after FUS, NfL serum concentrations increased two-fold, and by day two NfL increased 10-fold over baseline. Serum levels remained stable for another 2 weeks, until the AAV toxic effect induced a further rise in NfL serum levels. Proteomics analysis is planned for further exploration of biomarker discovery after FUS. We successfully demonstrated the feasibility to apply FUS for gene therapy delivery into the brain, and a 10-fold enhancement of CNS originated biomarker levels in circulation. FUS could enable the discovery of novel biomarkers that were previously undetectable in blood samples, thus offering a “non invasive” brain sampling method.

Disclosures: **S. Kostense:** A. Employment/Salary (full or part-time): Charles River. **S. Cotton:** A. Employment/Salary (full or part-time): Charles River. **M. Bsibsi:** A. Employment/Salary (full or part-time): Charles River. **M. Samba Mondonga:** A. Employment/Salary (full or part-time): Charles River. **S. Authier:** A. Employment/Salary (full or part-time): Charles River. **Z. Noroozian:** A. Employment/Salary (full or part-time): Charles River.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.10/VV9

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NRF/RS-2022-NR072290
KHIDI/RS-2024-00404555
NRF/RS-2024-00347107

Title: Focused ultrasound-mediated delivery of temozolomide enhances therapeutic efficacy in infiltrative glioblastoma

Authors: ***J. SHIN**¹, C. KONG², Y. SEO², S. HAN³, W. CHANG³;

¹K-MEDI Hub (Daegu-Gyeongbuk Med. Innovation Foundation), Daegu, Korea, Republic of;

²Dept. of Neurosurg., ³Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Introduction: Glioblastoma (GBM) is the most prevalent and aggressive primary brain tumor in adults, characterized by rapid proliferation and invasive infiltration into normal brain tissue. Despite maximal resection and temozolomide (TMZ) chemotherapy, over 80% of GBM cases recur near the resection margin, highlighting the need for improved therapeutic strategies. The blood-brain barrier (BBB) remains a major obstacle to effective drug delivery, limiting TMZ penetration into infiltrative tumor regions. This study explores the potential of focused ultrasound (FUS) to transiently open the BBB, optimizing TMZ delivery to GBM-

infiltrated areas of normal brain tissue before tumor neovascularization, and investigates its impact on tumor progression and survival in an orthotopic xenograft mouse model. **Methods:** Human primary GBM tumorspheres (TS15-88) were implanted into the striatum of 4- to 8-week-old male athymic nude mice to establish an orthotopic xenograft model. FUS was applied one week post-implantation, followed by intraperitoneal TMZ administration. BBB permeability was assessed using Evans blue extravasation, gadolinium-enhanced T1-weighted magnetic resonance imaging (MRI), and ZO-1 tight junction protein expression. GBM infiltration into the brain was confirmed using ZEB-1 and hematoxylin and eosin staining. Bioluminescence imaging and Kaplan-Meier survival analysis were used to evaluate the therapeutic effects of combined FUS and TMZ treatment. **Results:** MRI and Evans blue staining confirmed that BBB integrity was preserved in the tumor-only group, suggesting that tumor-induced neovascularization had not yet developed at the time of treatment. However, FUS-mediated BBB opening significantly enhanced Evans blue extravasation and reduced ZO-1 expression, indicating transient and localized BBB disruption. FUS-TMZ combination therapy significantly suppressed tumor growth, as evidenced by bioluminescence imaging, and prolonged survival compared to that with TMZ alone. Additionally, applying FUS in the early treatment phase (1-day group) showed a trend toward better tumor suppression and survival outcomes compared to that at later time points. **Conclusions:** Our findings suggest that integrating FUS with standard TMZ chemotherapy in the early treatment phase could enhance drug penetration into infiltrative tumor regions, improving tumor control and survival outcomes. These results highlight the clinical potential of FUS as an adjunct therapy to optimize TMZ efficacy, particularly in patients with early stage GBM.

Disclosures: J. Shin: None. C. Kong: None. Y. Seo: None. S. Han: None. W. Chang: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

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Program #/Poster #: PSTR309.11/VV10

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NIA/NIH R01AG085724
NIA/NIH R01AG062840

Title: The role of neonatal fc receptor in transferrin receptor-antibody fusion protein pharmacokinetics

Authors: *A. OYEBESAN;
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Abstract: The blood-brain barrier (BBB) presents a major bottleneck to delivering therapeutics into the brain. Transferrin receptor-targeting monoclonal antibodies (TfRMABs) offer a promising strategy for BBB transcytosis, and TfRMABs have demonstrated efficacy in delivering

therapeutics to the brain in preclinical models. However, the pharmacokinetics (PK), particularly under chronic dosing, are not fully understood. Emerging data suggest chronic administration can reduce plasma exposure in a dose- and fusion partner-dependent manner. The neonatal Fc receptor (FcRn) extends plasma half-life of IgG antibodies via recycling, but its saturation after repeated dosing may impact the PK profile of therapeutic IgG-fusion proteins. This study evaluated FcRn's influence on the plasma PK and biodistribution of TfRMAb-fusion proteins after repeated dosing, which remains unknown. We studied TfRMAb alone (without a fusion partner), and TfRMAb fused to erythropoietin (TfRMAb-EPO) and TfRMAb fused to the TNF receptor (TfRMAb-TNFR), two model fusion proteins, in wild-type (WT) and FcRn knockout (KO) mice. Mice received subcutaneous doses (3 mg/kg) either once (acute) or three times weekly for four weeks (chronic). Plasma levels, tissue biodistribution, and FcRn binding were measured using ELISA or electrochemiluminescence immunoassay (ECLIA). FcRn KO mice showed a trend toward lowered systemic exposure (plasma concentration and AUC) than WT mice after acute dosing for TfRMAb-EPO and TfRMAb, with a significant reduction for TfRMAb-TNFR. In WT mice, chronic dosing significantly reduced plasma exposure for all TfRMAb-fusion proteins. Despite the absence of FcRn, chronic dosing reduced plasma exposure in FcRn KO mice for TfRMAb and TfRMAb-EPO compared to their acute counterparts. For TfRMAb-TNFR, chronic dosing did not further reduce plasma exposure in FcRn KO mice. TfRMAb-TNFR also showed the weakest FcRn affinity, suggesting greater reliance on FcRn recycling and possible FcRn saturation in WT mice with chronic dosing. The impact of FcRn on biodistribution after chronic dosing varied by TfRMAb fusion protein. In conclusion, FcRn-mediated IgG protection varies with TfRMAb fusion partner and should be evaluated on a case-by-case basis. Our findings highlight the role of FcRn in sustaining systemic and tissue levels of TfRMAb fusion proteins after single dosing, with reduced impact under chronic administration, suggesting alternative mechanisms may drive plasma exposure during prolonged TfRMAb-fusion protein dosing. These insights are especially relevant amid ongoing advances in TfRMAb Fab-fusion protein development.

Disclosures: A. oyegbesan: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.12/VV11

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Development of fully human CD98hc antibodies to enhance delivery of therapeutic drugs across the blood-brain barrier

Authors: Z. WU¹, X. SUN¹, M. ZHANG¹, *K. D. LOMINAC², Z. JING³;

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Abstract: Background/Purpose:

CD98hc, also known as SLC3A2, CD98hc is a transmembrane glycoprotein belonging to type II membrane proteins. It plays a crucial role in a variety of biological processes, especially in amino acid (AA) transporters by interacting with light chains. CD98hc is highly expressed in blood-brain barrier, can enhance delivery of antibodies across the blood-brain barrier via transcytosis-enabling modules (TEMs) with inherent receptor-mediated transcytosis. Here, we developed fully human CD98hc antibodies to enhance delivery of therapeutic drugs across the blood-brain barrier.

Methods:

Fully human anti-CD98hc antibodies were generated from two transgenic mice via hybridoma screening. Mice serum titer and *in vitro* binding were evaluated using flow cytometry with CHO-K1-hCD98hc cells and CHO-K1-cynoCD98hc cells. CD98hc expression differences were analyzed by flow cytometry. In the amino acid transport assay, leucine transport mediated by CD98hc in hCMEC/D3 cells was quantified. For the endocytosis assay, the internalization of pHrodo-labeled CD98hc antibodies into hCMEC/D3 cells was measured by detecting labeled antibodies quantity. In the transcytosis assay, the CD98hc antibodies were incubated in the upper layer of the hCMECD3 cells barrier for 24 hours, and antibody concentrations in the lower chamber supernatant were determined by ELISA. The *in vivo* experiment is conducted by collecting brain tissues from humanized mice for ELISA detection after a single intravenous (I.V.) injection of CD98hc antibodies.

Results:

The recombinant CD98hc-specific antibodies exhibit excellent cross-reactivity with both human and cynomolgus. *In vitro* data demonstrates that lead molecules can induce potent endocytosis and transcytosis activity. Meanwhile, CD98hc antibodies did not impact on the expression of CD98hc or the functionality of amino acid transport. Additionally, *in vivo* pharmacokinetic studies in CD98hc-humanized mice reveal robust brain uptake of the antibodies.

Conclusion:

In summary, our fully human anti-CD98hc antibodies have demonstrated excellent properties in both *in vivo* and *in vitro* experiments, highlighting the potential of delivering therapeutic drugs across the blood-brain barrier.

Disclosures: **Z. Wu:** A. Employment/Salary (full or part-time): NeoMab Biotechnology. **X. Sun:** A. Employment/Salary (full or part-time): GemPharmatech Ltd. **M. Zhang:** A. Employment/Salary (full or part-time): NeoMab Biotechnology. **K.D. Lominac:** A. Employment/Salary (full or part-time): GemPharmatech, NeoMab Biotechnology. **Z. Jing:** A. Employment/Salary (full or part-time): GemPharmatech Ltd., NeoMab Biotechnology.

Poster**PSTR309: Techniques for Drug Delivery to the Brain**

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.13/VV12

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Longitudinal sample collection from multiple compartments utilising microdialysis for biodistribution evaluation and predictive modelling

Authors: *M. S. HEINS¹, H. DARWINKEL¹, E. DE LANGE²;

¹Charles River Labs., Groningen, Netherlands; ²Leiden Academic Ctr. for Drug Res., Leiden, Netherlands

Abstract: Biodistribution of compounds of interest into the CNS and their relative presence in the periphery is an important evaluation during drug development. As a clear understanding of these pharmacokinetics allows for a better correlation of the pharmacodynamics and supports translational pharmacokinetics modelling and predictions. *In vivo* compound level measurements over time in the CNS can be made by the use of microdialysis: a minimally invasive sampling technique that allows for accurate determination of compound levels in the extracellular fluid of the brain. Combining the quantitative MetaQuant microdialysis technology with blood and CSF collection in rodents provides samples to support biodistribution evaluation and prediction. Here we demonstrate the benefits of this experimental design following the compound levels after oral or intranasal administration.

We determined the biodistribution of diazepam and its metabolite nordazepam in freely-moving awake rats. To this end, the rats underwent surgery to position probes in the prefrontal cortex and the lateral ventricle for microdialysate collection. In the same surgical procedure, a jugular vein cannula for serial blood sample collection and a cisterna magna cannula for serial CSF sample collection were positioned. Following recovery, the probes were connected to microperfusion pumps and sample collection was initiated. Once flow speed was confirmed and the system equilibrated, the test compound was dosed and microdialysates samples were collected in 30-minute intervals. In addition, blood samples were collected at specified timepoints and processed to plasma. Finally, CSF samples were collected at specified timepoints in a crossover design to maintain the brain fluid flow balance. At the end of the experiments brain tissues were dissected and snap frozen. All collected samples were analysed for diazepam and nordazepam concentrations.

Levels of diazepam and nordazepam in rat MetaQuant microdialysate, plasma and CSF samples over time demonstrate the added value of the microdialysis technology itself. The levels of both diazepam and nordazepam in the different matrices illustrate the biodistribution within the CNS and representation in the blood and CSF. Feeding these data to relevant prediction models, add great additional value during the preclinical research phase. In addition to allowing for better predictions to the larger species these data are also relevant to support translation to human biodistribution and brain penetration in the clinical phase.

Disclosures: M.S. Heins: A. Employment/Salary (full or part-time): CRL. H. Darwinkel: A. Employment/Salary (full or part-time): CRL. E. de Lange: 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.; Elizabeth de Lange's Consultancy fees for CNS drug distribution prediction, using the LeiCNS PBPK model.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.14/VV13

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: CBF2023-2024-1982

Title: Evaluation of the reactive oxygen species production by photodynamic therapy applied to glioma cells using TiO₂-folic acid-ZnPc nanoparticles

Authors: *S. RODRIGUEZ¹, E. ORTIZ², C. RODRÍGUEZ-PÉREZ³, M.-R. MANRIQUEZ⁴, J. GUSTAVO⁵;

¹Facultad de Psicología, UNAM, CDMX, Mexico; ²Natl. Inst. of Neurol. and Neurosurg., Mexico City, Mexico; ³Mol. Neuropharm. and Nanotechnology, Natl. Inst. of Neurol. and Neurosurg., MEXICO CITY, Mexico; ⁴ESIQIE-IPN, Mexico City, Mexico; ⁵Univ. Autonoma Metropolitana, Mexico City, Mexico

Abstract: Glioblastoma is a highly aggressive brain tumor with a poor prognosis and limited therapeutic options. Photodynamic therapy (PDT) has emerged as a promising strategy to selectively eliminate tumor cells using light-activated photosensitizers that induce localized oxidative damage with minimal systemic toxicity. In this study, titanium dioxide (TiO₂) nanoparticles functionalized with folic acid (FA) and zinc phthalocyanine (ZnPc) were synthesized to enhance cellular uptake and light-mediated cytotoxicity in glioma cells. Structural and morphological characterization by Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM), and X-ray diffraction (XRD) confirmed a partial crystalline anatase phase and the successful incorporation of ZnPc. UV-Vis spectroscopy revealed absorption at ~700 nm associated with π - π^* transitions of ZnPc, supporting its potential as a photosensitizer. Interplanar distances calculated by Bragg's law and Fourier transforms from TEM were consistent (~0.35 nm for TiO₂ and ~1.5 nm for ZnPc), indicating structural stability after functionalization. Cell viability was assessed in LN18 glioma cells using the MTT assay at 24, 48, and 72 h, showing high viability at concentrations ≤ 100 μ g/mL and significant, dose-dependent cytotoxicity at higher concentrations, particularly for the TiO₂-FA-ZnPc sample. The nanocomposite thus maintained its structural and optical integrity while demonstrating a favorable cytotoxic profile. The effect of photoactivation is under evaluation by irradiating cells with ultraviolet (UV) light for 1 or 10 minutes at distances of 1 and 10 cm, 10 minutes after nanoparticle treatment. It is expected that UV exposure will enhance cytotoxicity in a time- and dose-dependent manner, particularly in the TiO₂-FA-ZnPc group. These findings may contribute to the development of targeted therapeutic strategies for gliomas using biocompatible and photoactivatable nanocarriers.

Disclosures: S. Rodriguez: None. E. Ortiz: None. C. Rodríguez-Pérez: None. M. Manriquez: None. J. Gustavo: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.15/VV14

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Enhanced Anti-seizure Efficacy by Focused-Ultrasound-mediated Blood Brain Barrier Modulation in Rat model of Temporal Lobe Epilepsy

Authors: *J. BAEK, J. PARK, S. HAN, H.-J. LEE, S. LIM, W. CHANG;
Yonsei Univ., Seoul, Korea, Republic of

Abstract: Introduction Refractory epilepsy refers to a condition where seizures cannot be controlled even with antiepileptic drugs (AEDs). AEDs, when administered at high doses, are known to cause various side effects. In particular, the second-generation AEDs, topiramate (TPM), have been reported to induce cognitive dysfunction at high doses. Recently, focused ultrasound (FUS) has gained attention for its application in the field of drug delivery. In this study, we aim to investigate whether focused ultrasound can enable low-dose AEDs to maintain their antiepileptic effects while minimizing side effects. Method 230~250g SD rats were used. SD Rats were divided into the Kainic acid group, Drug-only group, FUS-only group, and Drug+FUS group. Drug groups were divided into 10mg/kg, 15mg/kg, and 30mg/kg to set the drug concentration. The Kainic acid-derived epilepsy model was modeled by intra-hippocampal injection of kainic acid. FUS was sonicated in the hippocampus, inducing the blood-brain barrier (BBB) modulation. TPM was administered orally at the designated concentration for each group, starting one week after modeling. After the injection, all animals' seizure severity was assessed for 30 minutes every two days using the modified Racine's scale. Results Based on Racine's scale assessments, the 30mg/kg TPM group demonstrated antiepileptic effects in all subjects, while the 15mg/kg group showed effects in half of the subjects. In contrast, no antiepileptic effects were observed in the 10mg/kg group. Based on these results, the TPM dose for the FUS-treated group was set at 15mg/kg, and the study aimed to determine whether this group exhibited antiepileptic effects comparable to those at lower doses, potentially reducing drug-induced side effects. TPM is known to cause cognitive side effects when administered at high doses. Discussion Epilepsy is a chronic, non-communicable brain disorder characterized by recurrent seizures. Topiramate is known to cause cognitive side effects when administered at high doses. In epilepsy research, FUS has been investigated as a method to suppress seizures through neuromodulation, but its potential for drug delivery remains unexplored. In this study, we demonstrated that FUS-mediated drug delivery, combined with AEDs, can safely achieve antiepileptic effects. Some studies have reported that BBB opening using FUS can induce neurogenesis effects. Future experiments will be focused on confirming the neurogenesis effects induced by FUS.

Disclosures: J. Baek: None. J. Park: None. S. Han: None. H. Lee: None. S. Lim: None. W. Chang: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.16/VV15

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Implantable MicroLED-mediated Chemo-Photodynamic Combination Therapy for Glioma Treatment

Authors: *J. PARK, W. CHANG;
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Abstract: Photodynamic therapy shows promise for glioma treatment with powerful efficacy and low resistance, however, its effectiveness is significantly lowered by inadequate light delivery through the skull. Herein, a needle-type implantable microLED device and cathepsin B-responsive prodrug nanoparticles (PNPs) are newly exploited for glioma's chemo-photodynamic combination therapy. The microLED containing four small LEDs on the tip of its guide needle can be implanted into the center of glioma tissues without large area opening of skull, uniformly irradiating light to deep glioma tissues. PNPs are formulated via self-assembly of heterobifunctional prodrug composed of doxorubicin, verteporfin, and cathepsin B-cleavable peptide linker, wherein they stably maintain inactive nanoparticle structures in normal physiological conditions and specifically deliver therapeutic agent to cathepsin B-overexpressed glioma tissues. In vitro cellular assays, PNPs irradiated with showed the synergistic cytotoxicity of DOX and VFP only in cathepsin B-overexpressed cancer cells rather than normal cells. In tumor-bearing mice, PNPs showed high tumor accumulation via the nanoparticle-driven enhanced permeation and retention (EPR) effect and they also exhibited remarkable therapeutic efficacy against glioma under microLED-mediated light irradiation via chemo-photodynamic combination therapy. Accordingly, applying microLED with PNPs is an outstanding strategy to defeat glioma with cooperative chemo-photodynamic effects, minimal invasiveness, and desirable systemic/local safety.

Disclosures: J. Park: None. W. Chang: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.17/VV16

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Effects of Low-Dose Intraventricular Baclofen Delivery via Osmotic Pump in a Traumatic Brain Injury.

Authors: *H.-J. LEE¹, J. PARK², S. LIM⁴, C. SONG⁵, Y. KIM⁶, W. CHANG³;

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Abstract: Traumatic brain injury (TBI) is a leading cause of global mortality and disability, often resulting in cognitive deficits and behavioral changes. TBI is classified into primary brain injury and secondary brain injury. Primary injuries are uncontrollable so, therapeutic strategies aim to prevent secondary injuries. This study investigates baclofen, a treatment for spasticity, administered via cerebrospinal fluid (CSF) to prevent secondary brain injury. In this study, C57BL/6 mice (20-25g) were used; Sham, TBI with Intracerebroventricular (ICV) injection of baclofen via osmotic pump (0.2, 0.6, 4.5 µg/kg/day), and intraperitoneal (IP) injection of baclofen (0.2mg/kg/day). Baclofen was administered for 4 weeks. Following a 2-week rest, the open field and Y-maze tests were conducted to assess locomotor activity and spatial working memory, respectively. To evaluate neurological impairment induced by traumatic brain injury (TBI), the modified Neurological Severity Score (mNSS) was assessed weekly. After all behavioral tests were completed, animals were sacrificed for immunohistochemistry (IHC) and Western blot (WB). Our previous studies have shown that intraperitoneal administration of baclofen can prevent secondary brain injury by reducing neuroinflammation, and that a dose of 0.2 mg/kg is the most effective concentration in a TBI model. To determine intracerebroventricular (ICV) dose, we referred to other published studies shown that the dose ratio between intraperitoneal (IP) and intrathecal (IT) administration is approximately 1:300. As a result, both ICV 0.6 µg/kg and IP 0.2 mg/kg showed similar behavioral assessments. However, pronounced expression reduction of inflammatory related factors like Iba-1, GFAP, IL-1β, and C3, were observed with ICV administration of baclofen than IP injection. Moreover, ICV method resulted in less brain tissue loss. Additionally, as the concentration of baclofen increased, levels of secondary brain injury-related factors were decreased and reduced loss of lesion volume of cortex. The behavioral results demonstrated a positive correlation between the concentrations and performance except in the baclofen 4.5 µg/kg group. This study indicates that administration of baclofen via intracerebroventricular(ICV) routes leads to reduced brain parenchymal tissue loss compared to intraperitoneal delivery, despite similar results in behaviors. These results suggest the possibility of the usefulness of ICV baclofen administration as a therapeutic strategy to prevent secondary brain injury in TBI.

Disclosures: H. Lee: None. J. Park: None. S. Lim: None. C. Song: None. Y. kim: None. W. Chang: None.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.18/VV17

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Production and characterization of a target-selective biologic platform for CNS cargo delivery

Authors: ***L. WEBER**¹, A. M. RADIN², L. E. FENNO³, M. M. WELTZIN¹;

¹Univ. of Alaska Fairbanks, Fairbanks, AK; ²Numb Corp., Honolulu, HI; ³Psychiatry & Neurosci., Univ. of Texas at Austin, Austin, TX

Abstract: Chronic pain imposes substantial burdens on healthcare systems and societies due to increased use of medical services and loss of productivity. The widespread use and availability of opioids has led to a public health crisis of dependence and fatal overdose, emphasizing the critical need for safer therapeutic alternatives. To address this need, we designed novel binders that are selective for a specific receptor target expressed on defined neuronal populations involved in pain signaling. To produce the recombinant protein binders, BL21 (DE3) *E. coli* were used to express each type of binder, with IPTG being used as the inducer for protein production, and incubated at low temperatures to promote the formation of non-classical inclusion bodies. The formation of inclusion bodies in microbial cells is a common occurrence during the overexpression of recombinant proteins. We optimized the incubation conditions to favor the production of non-classical inclusion bodies, which harbor correctly folded proteins that can be solubilized by mild processes, over classical inclusion bodies. After 24hrs, cells were lysed and inclusion bodies were isolated, extensively washed, and purified until the binder proteins were solubilized using low concentrations of the anionic detergent sarkosyl. Extracted binders were further desalted with PBS and concentrated to minimize the residual sarkosyl. Binder purities were confirmed by SDS gel electrophoresis. The cytotoxicity of binder proteins was examined in neuronal-like N2a cells that express the receptor target of interest using the Alamar Blue assay. Nanomolar concentrations of the binders did not show significant effects, while micromolar concentrations decreased cell viability. To assess binder selectivity for the target receptor and cellular internalization, 100 nM - 1 μ M of each red fluorescent protein (RFP)-tagged binder was applied to HEK293 and N2a cells. Cells were visualized using live-cell confocal microscopy, and differences in fluorescence were quantified using ImageJ. Quantification of fluorescence revealed a significant increase in binder interaction with N2a cells expressing the receptor target compared to the HEK293 cells not expressing the receptor target. Each binder was found to be located inside cells within 24hrs of incubation, confirming cellular uptake. Future work includes testing of the binders in human induced pluripotent stem cells (iPSCs) and mice. Our technology is a pioneering biologic platform designed with high specificity to a receptor target expressed on neural subtypes for the directed delivery of therapeutic cargo.

Disclosures: **L. Weber:** 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.; Numb Corp. **A.M. Radin:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Numb Corp. **L.E. Fenno:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Numb Corp. **M.M. Weltzin:** 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.; Numb Corp.

Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.19/VV18

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: China Postdoctoral Science Foundation (No.2024M763656)
Science and Technology Project of Beijing Life Science Academy
(2024500CC0070)

Title: Peptide-engineered liposomes for nose-to-brain quercetin delivery targeting neuroinflammation

Authors: C. YIN^{1,2}, Y. TIAN^{1,2}, X. LI^{1,2}, H. WANG^{1,2}, S. HAN^{1,2}, L. CUI^{1,2}, ***H. CHEN**^{1,2}, H. HOU^{1,2};

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Abstract: Neuroinflammation serves as a central pathological driver in central nervous system (CNS) disorders. Current therapeutic approaches are constrained by the blood-brain barrier's limitation on traditional anti-inflammatory drugs and systemic adverse effects from prolonged administration. Quercetin (Qu) exhibits significant neuroprotective effects and disease-modifying properties in neurodegenerative disorders. However, the application of Qu is limited by the strong hydrophobicity and low bioavailability. To overcome the delivery challenges of Qu to CNS, we developed a short peptide-modified liposome-targeted delivery system (QuR-lipo) for intranasal brain-targeted delivery. This innovative platform integrated R-peptide surface functionalization to enhance transnasal epithelial penetration and CNS accumulation. Combining BV2 microglial models with LPS-induced neuroinflammatory animals, we systematically evaluated QuR-lipo's transmucosal efficiency, anti-inflammatory/antioxidant efficacy, and behavioral improvement effect. The results showed that QuR-lipo had high encapsulation efficiency and sustained-release characteristics. The R-peptide modification significantly enhanced liposomal penetration across the nasal epithelial barrier, achieving a 1.5-fold increase in brain drug accumulation compared to unmodified liposomes. Mechanistically, QuR-lipo synergistically suppressed NLRP3 inflammasome pathway activation and ROS overproduction, disrupting the vicious cycle of "neuroinflammation-oxidative stress". This dual intervention inhibited microglial morphological activation and downregulates the expression of phagocytic marker CD68. Spatial memory impairment and depression-like behavior were significantly improved in an animal model of LPS-induced neuroinflammation. This study establishes a novel strategy for delivering natural bioactive compounds to treat neurodegenerative disorders, merging nanoscale engineering with multi-level evidence from molecular pathways to functional recovery.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

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Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Support for this study was provided by the Neuroscience program, the College of Medicine, Office of Research and Sponsored Program, the E. Malcolm and Gary Leo Dunbar Endowed Chair, and the John G. Kulhavi Professorship in Neuroscience at CMU.

Title: Direct Brain Delivery of G4 70/30 PAMAM Dendrimers via the Nasal Route in C57BL/6 Mice

Authors: *A. ELMHIRST^{1,2,3,4}, N. ALLAHYARZADEH KHIABANI^{1,2,5,3}, O. DUBEY^{1,3,4}, A. POUDEL^{6,2,3}, B. SRINAGESHWAR^{7,3,8}, G. L. DUNBAR, Sr.^{9,3,2,8}, J. ROSSIGNOL^{5,3,2,4},
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Abstract: Various strategies for delivering drugs to the brain have been examined; however, the blood-brain barrier (BBB) remains a formidable obstacle to effective therapy for neurological disorders. In recent years, nanoscale carrier systems have shown promise in overcoming this barrier. Among these, intranasal (nose-to-brain) delivery has emerged as a non-invasive approach that can enhance treatment efficacy by avoiding the BBB. The olfactory nerve pathway, which leads directly into the olfactory bulbs, provides a vital channel for nanoparticles to enter the central nervous system (CNS). Dendrimers—highly branched macromolecules capable of encasing and transporting therapeutic agents—are particularly well-suited for this purpose. In particular, polyamidoamine (PAMAM) dendrimers have demonstrated the ability to traverse the BBB. Intranasal administration thus represents a non-invasive route that delivers drugs straight to the CNS via the olfactory nerve pathway, bypassing systemic circulation and minimizing peripheral side effects. The present study aimed to evaluate the in vivo impact of intranasally administered PAMAM G4 70/30 dendrimers on the brains of C57BL/6 mice. Experimental cohorts included male (n = 6) and female (n = 6) C57BL/6 mice. The treatment group received daily intranasal doses of CY5.5-labeled G4 70/30 PAMAM dendrimers, while controls were given HBSS. Over a three-week period after initiating treatment, dendrimer distribution was monitored weekly using an In Vivo Imaging System (IVIS). At the end of this period, brain, lung, liver, and kidney tissues were harvested, frozen, and sectioned for further

study. Fluorescence microscopy was then used to assess the biodistribution of the CY5.5-labeled dendrimers. Results indicated significant accumulation of G4 70/30 PAMAM dendrimers in the brain, liver, and kidney of treated mice, with male mice exhibiting higher accumulation levels than females. These observations suggest that intranasal administration effectively facilitates dendrimer delivery to the brain. These findings have important implications for drug delivery research, demonstrating that intranasal administration can serve as an efficient and minimally invasive route for targeting the brain. Moreover, the successful brain delivery of G4 70/30 PAMAM dendrimers via this method supports their potential use in transporting therapeutic agents—such as small molecules or gene therapies—to treat brain tumors. In conclusion, intranasal delivery of G4 70/30 PAMAM dendrimers enables efficient brain targeting, offering a promising strategy for the development of innovative therapies against neurological diseases.

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Poster

PSTR309: Techniques for Drug Delivery to the Brain

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR309.21/VV20

Topic: J.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Intranasal deferiprone ameliorates behavioral abnormalities in mice with brain iron loading

Authors: *R. CHENG, J. KIM;
Univ. of Massachusetts Lowell, Lowell, MA

Abstract: Brain iron accumulation is implicated in several central nervous system (CNS) disorders, including neurodegenerative diseases and neuropsychiatric conditions. Deferiprone (DFP), an FDA-approved oral iron chelator, has shown CNS activity owing to its ability to cross the blood-brain barrier. Notably, DFP has demonstrated antidepressant-like effects in mouse models, potentially by influencing neural activity without significantly altering brain iron levels. However, its oral use requires frequent high doses, which increases the risk of systemic toxicity, such as neutropenia. Therefore, we hypothesized that intranasal (IN) DFP delivery could achieve therapeutic concentrations in the brain at lower, non-toxic doses, thereby improving neurobehavioral outcomes without adverse effects. To evaluate DFP's therapeutic potential, we administered IN DFP to HFE-H67D mice, a model of brain iron overload that accompanies emotional disorders such as hyperactivity and impulsive behavior, and characterized the pharmacokinetics (PK) and pharmacodynamics of DFP. PK analysis revealed that IN administration achieved high brain concentrations of DFP within 15 minutes, suggesting a rapid and efficient brain delivery of DFP via the nose-to-brain transport pathway. Furthermore, IN delivery showed approximately six-fold higher DFP levels in the olfactory bulb and prefrontal cortex region compared with the oral route at the same dose (20 mg/kg) after 1 hour post-dose. In

addition, the elevated plus maze (EPM) task revealed that the impulsive behavior of H67D mice was normalized by IN DFP (20 mg/kg, daily for one week) but not by oral DFP (equivalent dose schedule). Similar behavioral improvements were observed when DFP was given orally at 100 mg/kg over six weeks. These results suggest that IN delivery produces more rapid and efficient therapeutic effects of DFP than the oral route. Hyperactive behavior, assessed by the mean speed in the EPM, was not significantly altered by IN DFP. Although the mechanisms by which DFP corrects the impulsive behavior remain unclear, western blot analysis of the hippocampal tissue showed a reduction in glial fibrillary acidic protein (GFAP) expression after two weeks of IN DFP treatment. These findings suggest intranasal DFP could correct behavioral alterations associated with neuroinflammation. IN DFP may provide a non-invasive, brain-targeted alternative to oral administration for CNS conditions such as neuropsychiatric disorders. Further studies are needed to elucidate the underlying mechanisms and assess the translational potential of IN DFP in treating neuropsychiatric conditions.

Disclosures: R. Cheng: None. J. Kim: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.01/VV21

Topic: J.06. Computation, Modeling, and Simulation

Support: National Research Foundation of Korea (NRF) Grant RS-2023-0280241

Title: Numerical analysis of microbubble dynamics in transcranial focused ultrasound using acoustic simulations

Authors: J. YU¹, *H. SEO²;

¹Dept. of Computer Sci. & Engin., Gyeongsang Natl. Univ., Jinju-si, Korea, Republic of; ²Dept. of Computer Sci. & Engin., Gyeongsang national Univ., Jinju-si, Korea, Republic of

Abstract: Focused ultrasound (FUS) combined with microbubbles can temporarily disrupt the blood-brain barrier (BBB) and enhance drug delivery. However, microbubble dynamics depend on multiple acoustic and bubble parameters, making controlled BBB opening challenging. Numerical studies can explore these systematically, but most models assume ideal sinusoidal waveforms, neglecting skull-induced acoustic distortions. Although some studies have simulated skull effects, their impact on microbubble behavior remains underexplored. To address this, we developed multi-scale models integrating microbubble dynamics with a 3D acoustic skull model. We conducted FUS simulations using the k-Wave toolbox in MATLAB with CT and MRI data from the Visible Human Project female dataset. Skull segmentation was performed on CT images, and MRI was co-registered to localize the thalamus. Skull acoustic properties were computed from CT-derived porosity; other tissues were assigned water properties. A single-element focused transducer (focal length: 110 mm; inner/outer diameter: 30/128 mm; 250 kHz)

was positioned perpendicularly to the skull. Microbubble dynamics were simulated with the Marmottant model using four initial radii (0.82, 1.00, 1.75, and 2.15 μm). Peak pressures of 0.1, 0.2, and 0.3 MPa were used. We quantified volumes of stable (SC) and inertial cavitation (IC). SC consistently spanned larger regions; no IC occurred at 0.1 MPa. Cavitation volume increased with bubble size. At 0.3 MPa, IC volume differed by 2.3-fold between the smallest and largest bubbles in the skull model, implying bubble-specific safety thresholds. Compared to the free-water model, skull simulations showed greater cavitation regions in most conditions. At 0.3 MPa, the IC volume increased by approximately 40% without the skull, suggesting skull-induced focusing amplifies risk. These results highlight that both acoustic pressure and bubble size influence cavitation behavior, and skull-induced distortions must be considered. Our model enables detailed prediction of these effects and may guide safe clinical use of microbubbles.

Disclosures: J. Yu: None. H. Seo: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.02/VV22

Topic: J.06. Computation, Modeling, and Simulation

Title: Is the whole more than the sum of its parts? Accounting for higher order statistical associations in brain-behavior modeling

Authors: S. RILEY¹, A. CHENG², Y.-W. WANG³, X. SHEN⁴, A. J. HOLMES⁵, R. T. CONSTABLE⁶, *S. W. YIP⁷;

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Abstract: Background: While multiple different methods for predictive brain-behavior modeling exist, variation between these approaches is generally at the algorithmic level, for example using different types of penalized regression to boost feature selection. Here, we test an alternative approach by asking whether accounting for additional information in the matrix space might also influence model performance and ask what the implications of that might mean for our understanding of the brain. Methods: Using data from the Human Connectome Project (N=515) we adopt a global, geometrically grounded measure of similarity that accounts for higher-order relationships and interactions among brain regions—the Wasserstein distance metric—and compare this metric’s utility against other standard distance metrics for: (i) successfully matching multiple different connectivity matrices from the same individual (aka ‘ID rate’) and; (ii) predicting fluid intelligence from connectivity matrices. To benchmark performance relative to two widely used brain-behavior modeling approaches, we also conducted standard connectome-based predictive modeling (CPM; i.e., vector-based linear feature selection using

correlation, no multi-dimensional scaling) and kernel ridge regression (KRR). Results: ID rate using the Wasserstein metric outperformed both standard correlation (68.6% ID rate) and other distance metrics, with an overall ID rate of 93.6%. Predicting of fluid intelligence using the Wasserstein metric ($r=.41$) also outperformed both CPM ($r=.28$) and KRR ($r=.31$, $t(8)=9.80$, $p<.001$ vs CPM; $t(8)=10.89$, $p<.001$ vs KRR). Conclusions: These results suggest that, relative to standard vector-based approaches, considering matrices as representations of their associated Gaussian distributions enables retention of features of the matrix space that meaningfully differ between individuals and that are relevant for predicting individual differences in phenotypic expressions, such as IQ.

Disclosures: S. Riley: None. A. Cheng: None. Y. Wang: None. X. Shen: None. A.J. Holmes: None. R.T. Constable: None. S.W. Yip: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.03/Web Only

Topic: J.06. Computation, Modeling, and Simulation

Support: NIH R01NS121040

Title: Integration of Surrogate-Assisted Global Optimization into a Biophysical Model for Assessing Underlying Physiological Responses Associated with BOLD HRF

Authors: *M. JUNG^{1,3}, A. VINOGRADOV¹, N. J. FESHARAKI¹, G. JUN², J. KIM¹;
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Abstract: Although our previous arterial impulse model (AIM) enabled the estimation of underlying physiological responses from blood oxygen-level dependent (BOLD) hemodynamic response function (HRF), it had several limitations. These included reliance on manual parameter initialization and inefficient exploration of the solution space, which may have led to convergence on local minima. As a result, the model's ability to reliably distinguish key physiological dynamics—such as cerebral blood flow (CBF) and the cerebral metabolic rate of oxygen (CMRO₂)—was limited. To address this, we developed a surrogate-assisted global optimization framework incorporating particle swarm optimization (PSO) guided by a gradient boosting regressor (XGBoost). Six physiological parameters were constrained within biologically plausible bounds, and a Latin hypercube sampling strategy generated >700 candidate parameter vectors per subject. We evaluated each by comparing model-predicted BOLD responses to experimental data, creating error metrics used to train an XGBoost surrogate. The surrogate model rapidly screened candidates and selected top solutions, which were perturbed $\pm 10\%$ of their ranges to initialize a high-quality swarm. PSO was then run using the full AIM model. Optimization on bootstrap-resampled BOLD data ($n = 30$) used mean-

squared deviation normalized by error estimates. Across runs, our method consistently recovered parameter sets meeting error thresholds based on confidence intervals. Post-optimization, we extracted model-derived CBF and CMRO₂ responses and quantified parameter sensitivity via outcome variance. Results indicate the proposed method improves convergence stability and mapping of feasible physiological dynamics. Our approach enables more comprehensive exploration of the AIM parameter space while reducing the computational burden of full simulations. This framework facilitates scalable application of AIM across large neuroimaging datasets and supports future extensions involving subject-specific priors and adaptive surrogate retraining.

Disclosures: M. Jung: None. A. Vinogradov: None. N. J. Fesharaki: None. G. Jun: None. J. Kim: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.04/WW1

Topic: J.06. Computation, Modeling, and Simulation

Title: Spatiotemporal Averaging Leads to Emergent Macroscopic Linearity in Networks of Correlated Neurons

Authors: *H. AHMED¹, S. AHMED², E. NOZARI³;

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Spatiotemporal Averaging Leads to Emergent Macroscopic Linearity in Networks of Correlated Neurons

Sabbir Ahmed

Hafiz Fareed Ahmed

Erfan Nozari

Abstract

Neural dynamics are inherently nonlinear at the cellular level, yet large-scale brain signals, such as those measured by functional magnetic resonance imaging (fMRI) and electroencephalography (EEG), often exhibit dynamics that are remarkably well-approximated by linear models. Whether macroscopic neural dynamics are truly linear, however, cannot be asserted based on data-driven modeling per se. In prior work [1,2], we have showed through simulations as well analytical proofs that spatial and temporal averaging indeed result in macroscopically linear behavior. Our prior results, however, applied only to the extreme case of independent and identically distributed (i.i.d.) neurons.

In this study, we develop a general theoretical framework explaining how both temporal and spatial averaging give rise to linear behavior in a wide class of nonlinear, stochastic, and heterogeneous neural systems, even when these systems are interconnected and exhibit complex correlations. For the temporal case, we prove that averaging over time smooths nonlinearities and leads to linear macroscopic behavior, even when the averaging window varies dynamically, thus overcoming limitations such as signal degeneracy, where excessive smoothing can obscure meaningful temporal transitions in neural activity.

For the spatial case, we analytically prove that spatial averaging (still) gives rise to linear macroscopic behavior, even in the presence of heterogeneous and correlated neuronal dynamics. In particular, our results extend to geometrically-embedded, three-dimensional spatially distributed arrays of neurons with arbitrary spatial and network topologies. The only key assumption is the decay of pairwise correlations with at least one notion of distance (Euclidean, synaptic, or even functional) in order to ensure the lack of global synchrony.

Together, our results offer the first unified theoretical explanation for how linearity emerges from nonlinear neural dynamics through intrinsic averaging processes, and provide a mechanistic explanation for why linear models remain effective in capturing large-scale neural dynamics despite the complex, nonlinear nature of individual neuronal components.

[1] S. Ahmed and E. Nozari, "On the Linearizing Effect of Spatial Averaging in Large-Scale Populations of Homogeneous Nonlinear Systems," 2022 IEEE 61st Conference on Decision and Control (CDC), Cancun, Mexico, 2022, pp. 641-648.

[2] S. Ahmed and E. Nozari, "On the Linearizing Effect of Temporal Averaging in Nonlinear Dynamical Systems," 2023 American Control Conference (ACC), San Diego, CA, USA, 2023, pp. 4185-4190.

Abstract:

Disclosures: H. Ahmed: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.05/Web Only

Topic: J.06. Computation, Modeling, and Simulation

Support: UNAM-PAPIIT TA101224

Title: High-quality distortion correction in diffusion MRI using artificial intelligence with preservation of local resolution

Authors: *J. MONTES RIOS¹, P. OCAMPO LUNA¹, M. GARCIA-GOMAR¹, L. CONCHA², M. J. FAIR¹;

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Abstract: Diffusion weighted imaging (DWI) MRI is a fundamental component of neuroscience research and clinical practice for probing white matter integrity. However, its reliance on echo planar imaging (EPI) makes it highly susceptible to geometric distortions in regions with strong magnetic field inhomogeneities, especially near air and tissue interfaces such as those at the frontal and temporal poles. These artifacts can compromise neuroanatomical precision and introduce bias in group level analyses.

Traditional post processing techniques, including corrections based on field maps or image registration, can mitigate such distortions but are constrained by additional acquisition requirements, high computational cost, and residual deformation. In this work, we optimized a deep learning based correction framework capable of learning the complex and nonlinear mapping from distorted to undistorted DWI images.

We implemented a modified two dimensional U-Net, based on the work of Hu et al. (2020), trained and tested on both preliminary synthetic data with simulated distortions as well as IRB approved prospectively acquired data (3T Phillips; 212 images for training, 27 validation, 27 testing). The model was optimized by analyzing parameters including patch size, patch overlap, dataset size, number of training epochs, loss functions, and evaluation metrics.

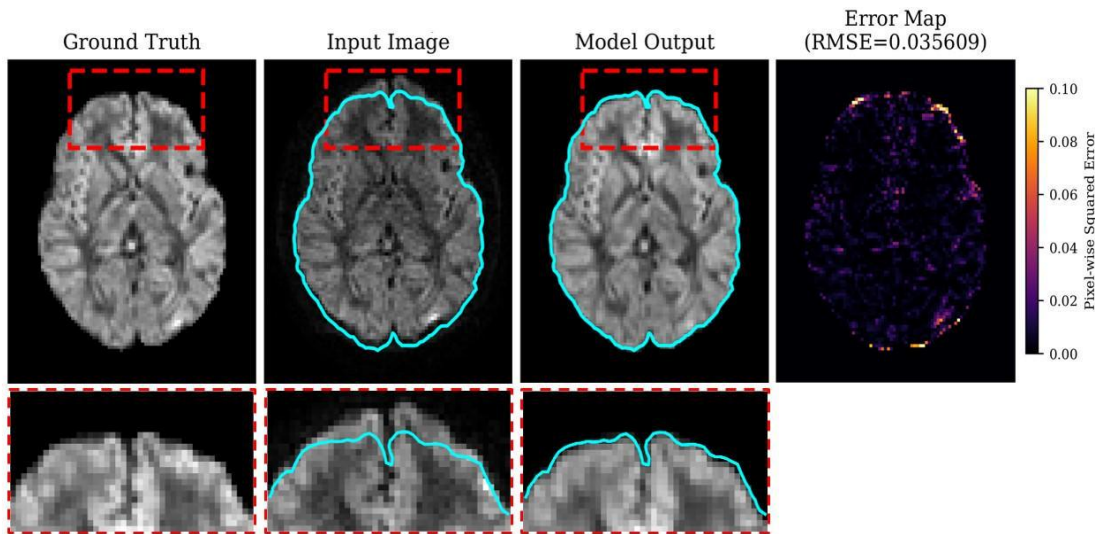
In both datasets, the architecture effectively corrected image distortions while preserving the fine spatial features that are critical for tractography and anatomical segmentation. The optimized model achieved an average root mean squared error (RMSE) of $4.7 \pm 1.2\%$ and a structural similarity index (SSIM) of 0.94 ± 0.02 on the test set, indicating high fidelity reconstructions with no perceptible visual artifacts. These results suggest high reliability for diffusion derived metrics and support a more efficient neuroimaging workflow.

Future work will focus on addressing anatomically complex distortions, utilizing distortion-free acquisitions to improve model training.

This work was supported by UNAM-PAPIIT TA101224.

Model Correction Performance Example

RMSE: 0.035609 | SSIM: 0.9679 | MAE: 0.013520



Disclosures: J. Montes Rios: None. P. Ocampo Luna: None. M. Garcia-Gomar: None. L. Concha: None. M.J. Fair: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.06/WW2

Topic: J.06. Computation, Modeling, and Simulation

Title: Predicting endocrine state from functional brain organization within and between individuals

Authors: *I. S. GEPHART, M. D. ROSENBERG;
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Abstract: While functional brain connectomes are highly stable within individuals (Gratton et al. 2018), they can simultaneously vary depending on the state of the individual. Factors like current task (Gonzalez-Castillo et al. 2015), typical sleep duration (Mummaneni et al. 2023), and ongoing thoughts (Gonzalez-Castillo et al. 2024) can alter functional connectivity in the brain. But what about other biological processes occurring in the body? In women, female sex hormones (estradiol, progesterone, FSH and LH) vary greatly over the 28 day menstrual cycle and correspond with changes in functional connectivity (Pristchet et al. 2020). Beyond naturally produced hormones, 80% of women in the United States take oral contraceptives (OCs) at some point in their life (Daniels and Jones, 2013). It has also been shown that within a single individual OC use can be predicted based on functional connectivity, and the predictive model

generalizes to a small number of similarly sampled individuals (Bottenhorn et al. 2025). However, it is unknown if these networks generalize to large samples with different hormonal contraceptive formulations. In this work, we analyze resting-state fMRI data from a single subject precision neuroimaging sample which includes hormone levels for each scan (n=60; Pritschet et al. 2020) to investigate what brain networks differ while naturally cycling compared to when taking OCs. Our results indicate that there are widespread differences in network connectivity depending on OC use within an individual. Using network based statistics (Zalesky et al. 2010), we identified two networks, a “Naturally Cycling Network” and an “OC Network” that include 2238 and 3241 edges, or 6% and 9% of all possible edges, respectively. These edges are spread across all networks in the brain, aligning with previous results (Bottenhorn et al. 2025). However, we then applied the identified networks to the Human Connectome Project dataset (Van Essen et al. 2013), which includes resting-state fMRI scans paired with self report OC use (largely “yes” or “no” without further detail). We find no generalization of the within-subject networks to the cross-sectional data. This suggests that general self-report characterizations of OC may not be sufficient to identify these differences in functional connectivity, highlighting the importance of increasing the specificity of endocrine-related measures in demographic questionnaires. This advancement is critical not only because women’s health topics are understudied in cognitive neuroscience (Taylor et al. 2020), but because it will lead to better understanding of functional brain network stability and variability in general.

Disclosures: I.S. Gephart: None. M.D. Rosenberg: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.07/WW3

Topic: J.06. Computation, Modeling, and Simulation

Support: NIH Grant P01NS131131 (A.S., B.M., J.M.S., A.N.)
H.H. received salary support from TOP-TIER grant T32- EB021955 during the study period

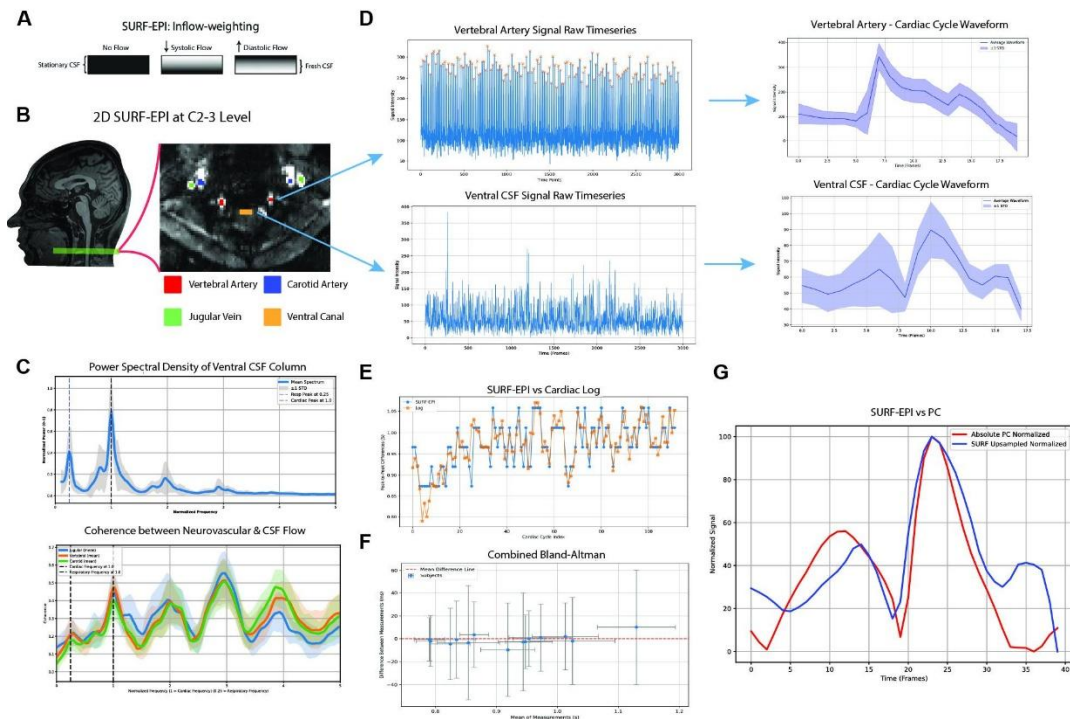
Title: Simultaneous real-time Imaging of neurofluid and neurovascular dynamics using ultrafast flow-weighted echo-planar imaging

Authors: *A. SHAKER¹, H. HOSSEINI¹, C. ELDENIZ¹, B. A. MARTIN³, A. SOTIRAS², J. M. STRAHLE⁴, A. NAZERI⁵;

¹Radiology, ²Washington Univ. in St. Louis, St. Louis, MO; ³Dept. of Chem. and Biol. Engin., The Univ. of Idaho, Moscow, ID; ⁴Dept. of Neurosurg., Washington Univ. Sch. of Med., St. Louis, MO; ⁵Washington Univ. Sch. of Med., Saint Louis, MO

Abstract: Understanding the connection between cerebrospinal fluid (CSF) and blood flow is crucial for how the brain regulates pressure, clears waste, and maintains homeostasis. Phase-

contrast MRI measures CSF flow but relies on gating and velocity encoding, limiting temporal resolution and simultaneous vascular-CSF capture. We present a self-gated ultrafast echo planar imaging (SURF-EPI; Fig. 1A), method enabling bi-directional inflow-weighted imaging with 47 ms temporal and 1.3 mm² spatial resolution, with a wide dynamic range that captures both neurovascular and CSF flow (Fig. 1B). This approach was performed alongside PC MRI at the C2-C3 level in 21 participants on a 3T Siemens Prisma scanner. Frequency-domain and event-related analyses were used to assess vascular-CSF coupling and reconstruct CSF waveforms across the cardiac cycle. Spectral analysis of CSF signals showed clear respiratory and cardiac peaks, including higher-order cardiac harmonics. Magnitude Squared Coherence analysis revealed stronger CSF-neurovascular coupling at cardiac frequencies, peaking at the third harmonic, suggesting arterial and venous pulsations drive spinal CSF flow (Fig. 1C). Systolic peaks from vertebral artery waveforms enabled event-related CSF cycle reconstruction without external gating. Time segments around these events were averaged to generate voxel-wise CSF waveforms (Fig. 1D). We compared cardiac cycle lengths from SURF-EPI with physiological logs ($n = 15$; 14 ICC > 0.60; error < 47 ms in 14; Fig. 1E, F). In addition, waveform comparisons with PC MRI showed strong or very strong correlation ($r > 0.60$) in 75% of participants, and moderate correlation ($r = 0.49$ -0.58) in the rest (Fig. 1G). SURF-EPI enables fast, simultaneous imaging of CSF and neurovascular flow (>20 Hz), capturing cardiac-driven dynamics and flow phases without external gating. By avoiding velocity-encoding constraints, it overcomes the limited dynamic range and high phase noise of real-time PC MRI, providing a robust, efficient alternative for CSF and vascular flow assessment.



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Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.08/WW4

Topic: J.06. Computation, Modeling, and Simulation

Support: NSF EFRI grant 2223495
UW + Amazon Science Hub grant
Templeton World Charity Foundation Frameworks grant
Air Force Office of Scientific Research under award number FA9550-24-1-0313

Title: Data Heterogeneity Limits the Scaling Effect of Pretraining in Neural Data Transformers

Authors: L. P. JIANG, S. CHEN, E. TANUMIHARDJA, X. HAN, W. SHI, E. T. SHEA-BROWN, *R. P. RAO;
Univ. of Washington, Seattle, WA

Abstract: A key challenge in analyzing neuroscience datasets is the profound variability they exhibit across sessions, animals, and data modalities—i.e., heterogeneity. Several recent studies have demonstrated performance gains from pretraining neural foundation models on multi-session datasets, seemingly overcoming this challenge. However, these studies typically lack fine-grained data scaling analyses. It remains unclear how different sources of heterogeneity influence model performance as the amount of pretraining data increases, and whether all sessions contribute equally to downstream performance gains. In this work, we systematically investigate how data heterogeneity impacts the scaling behavior of neural data transformers (NDTs) in neural activity prediction. We found that explicit sources of heterogeneity, such as brain region mismatches among sessions, reduced scaling benefits of neuron- and region-level activity prediction performances. For tasks that do exhibit consistent scaling, we identified implicit data heterogeneity arising from cross-session variability. Through our proposed session-selection procedure, models pretrained on as few as five selected sessions outperformed those pretrained on the entire dataset of 84 sessions. Our findings challenge the direct applicability of traditional scaling laws to neural data and suggest that prior reports of multi-session scaling benefits may need to be re-examined in light of data heterogeneity. This work both highlights the importance of incremental data scaling analyses and suggests new avenues toward optimally selecting pretraining data when developing foundation models on large-scale neuroscience datasets.

Disclosures: L.P. Jiang: None. S. Chen: None. E. Tanumihardja: None. X. Han: None. W. Shi: None. E.T. Shea-Brown: None. R.P. Rao: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.09/WW5

Topic: J.06. Computation, Modeling, and Simulation

Support: Seattle Children's Seed Grant

Title: Self-map: dissociative self-report geometry reveals a cortical action-to-memory gradient

Authors: *C. TANG¹, W. LIU², A. LU³, M. SONG², A. SHIN², A. FU⁴, R. LEELADHARAN⁵, Z. ZHENG⁶, L. MAKARYAN², J. LIU², L. KE⁴, A. TAN², T. N. TRAN², Q. GE², T. LEE⁷, M. KOH², M. ROUT², S. MUSINOVIC², W. CHAMBERS², G. SCIMECA⁸, I. OANE⁹, I. MINDRUTA¹⁰, M.-A. KATE¹¹;

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Abstract: A central challenge for consciousness science is translating patterns in first-person symptom reports into concrete brain predictions. Dissociation questionnaires are ideal for this purpose because their items span disruptions of agency, self-reference, self/non-self perception and autobiographical memory. To test whether the hidden geometry of such reports mirrors cortical layout, we developed **SELF-MAP**—a similarity-based embedding framework that extracts latent topologies from cross-individual variation. The approach was first validated on problems with known structure. SELF-MAP recreated the two-dimensional layout of synthetic objects and recovered the three-dimensional fold shared by nanobody scaffolds directly from nanobody-antigen interface profiles, showing that snapshots of “self-to-other” interactions can encode genuine physical form. Item-level response frequencies from two independent cohorts who completed the 60-item Multidimensional Inventory of Dissociation (MID-60) were then analysed. The resulting SELF-MAP configuration was highly reproducible. Crucially, we oriented this subjective space by leveraging item-specific associations between MID-60 scores and functional-MRI activations; this alignment to cortical coordinates revealed a clear anterior-to-posterior gradient. Along the newly oriented principal axis, statements tied to disrupted agency and self-referential control mapped to frontal loci, whereas statements linked to autobiographical memory and altered reality mapped to posterior midline regions. Convergent evidence came from intracranial stimulation: sites whose perturbation produced action or agency disturbances clustered anteriorly, while sites inducing memory or reality distortions clustered posteriorly. As a semantic control, embeddings produced solely by a state-of-the-art language model preserved psychological relationships yet failed to align with the cortical gradient, indicating that the anatomically relevant ordering arises specifically from human response variation. Together, these results show that dissociative self-reports harbour a latent spatial topology that, once anchored by fMRI data, mirrors a macroscale cortical action-to-memory

gradient. SELF-MAP thus offers a quantitative bridge between subjective phenomenology and the neural interface of self-experience.

Disclosures: C. Tang: None. W. Liu: None. A. Lu: None. M. Song: None. A. Shin: None. A. Fu: None. R. Leeladharan: None. Z. Zheng: None. L. Makaryan: None. J. Liu: None. L. Ke: None. A. Tan: None. T.N. Tran: None. Q. Ge: None. T. Lee: None. M. Koh: None. M. Rout: None. S. Musinovic: None. W. Chambers: None. G. Scimeca: None. I. Oane: None. I. Mindruta: None. M. Kate: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

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Program #/Poster #: PSTR310.10/WW6

Topic: J.06. Computation, Modeling, and Simulation

Support: NRF Grant RS-2022-NR070832

Title: Hemodynamic Analysis Using Integral Parameters in 4D Flow MRI

Authors: *J. KOO¹, S. LEE²;

¹Korea Brain Res. Inst. (KBRI), Daegu, Korea, Republic of; ²Seoul Natl. Univ. Boramae Med. Ctr., Seoul, Korea, Republic of

Abstract: Four-dimensional flow magnetic resonance imaging (4D flow MRI) is a non-invasive imaging technique that enables the measurement of patient-specific, three-dimensional blood flow velocity fields. Its superior flow visualization capabilities have led to widespread applications in both engineering and hemodynamic research. However, the clinical utility of 4D flow MRI is hindered by its inherently low spatial resolution, which affects the accuracy of key hemodynamic parameters such as wall shear stress (WSS) and oscillatory shear index (OSI). These parameters, commonly used in cardiovascular disease diagnosis, are highly sensitive to spatial resolution and noise due to their differential-based nature. To overcome these limitations, we propose an alternative integral-based hemodynamic parameter analysis method that preserves the diagnostic effectiveness of conventional WSS and OSI while significantly improving robustness against spatial resolution variations.

Disclosures: J. Koo: None. S. Lee: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.11/WW7

Topic: J.06. Computation, Modeling, and Simulation

Support: NIGMS 5P30GM149405

Title: Voxalign: automated single-voxel MR spectroscopy prescriptions for reproducible voxel placement

Authors: *E. S. LORENC, F. E. MCELENEY, G. L. LEBLANC, M. S. WORDEN;
Brown Univ., Providence, RI

Abstract: Single-voxel MR spectroscopy (MRS) measures concentrations of neurometabolites and neurotransmitters within a relatively large volume of $\geq 1 \text{ cm}^3$. A single voxel often samples both gray and white matter, and multiple neuroanatomical structures; even a small change in placement can differently sample the underlying anatomy and change the measured spectrum. Therefore, particularly for longitudinal studies, it is crucial that voxel placements be reproduced accurately across sessions.

MRS voxels are typically prescribed manually. However, there are automated options: older versions of Siemens software include AutoAlign for spectroscopy, although current versions do not (XA30+). Alternatively, CMRR provides a specialized sequence with automated MRS voxel prescription (AutoVOI)¹. While AutoVOI improves within-subject voxel placement accuracy over manual prescriptions (~90% vs ~85% voxel overlap)¹, it remains far short of exact replication and does not support product sequences.

Our python package voxalign (<https://github.com/brown-bnc/voxalign>) is designed to ensure reproducible within-participant voxel placements across scan sessions. Running on a separate computer, voxalign takes in a T1-weighted anatomical DICOM and spectroscopy DICOM(s) from the first MR session, and a T1 from an in-progress second session. All DICOMs are converted to NIFTI format, and the T1s are coregistered. The resulting transform, combined with the affine information from the T1s and MRS voxel(s), determines the new voxel position and orientation that will exactly match the first session. Finally, this is translated into Siemens notation (e.g. Position: L11.1 A50.9 H58.8; Orientation: C > T-23.1 > S2.0; Rotation: -1.03°) for entry on the MR console. Voxalign can also prescribe based on MNI coordinates, which is useful for standardization across participants, and for voxel placements in the first session of a longitudinal study.

Here, we illustrate usage and present voxalign accuracy for a demonstration dataset in which 3 human participants each completed 2 scan sessions. Voxalign was used to center the session 1 voxels at MNI coord [-8, 38, 31], and then to exactly match each participant's session 1 prescription for the session 2 scan. Within-subject overlap was quantified with the Dice coefficient (0=no overlap; 1=perfect overlap). The resulting Dice coefficients (1.0, 1.0, 0.99) indicate that voxel positions were exactly reproduced across sessions.

Multiple studies at Brown have already adopted voxalign, and we are facilitating its use by other research sites.

¹ Y. W. Park et al., Magn. Reson. Med. 80, 1787-1798 (2018).

Disclosures: E.S. Lorenc: None. F.E. McEleney: None. G.L. LeBlanc: None. M.S. Worden: None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.12/WW8

Topic: J.06. Computation, Modeling, and Simulation

Title: A Bidirectional Machine Learning Framework for Mapping Fetal Brain Connectivity and Molecular Features

Authors: *Y. WEI, C. ZHANG;
Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Microscale molecular variations critically influence macroscale brain connectivity during human fetal development; however, direct mechanistic mapping between these scales remains lacking. We developed a comprehensive bidirectional computational framework integrating single-cell/nucleus RNA sequencing (sc/snRNA-seq) and fetal MRI to bridge this gap and enhance early diagnostic capabilities for neurodevelopmental disorders. Transcriptomic datasets from gestational weeks (GW) 6-38 spanning diverse brain regions were harmonized using the Seurat-Harmony pipeline and spatially mapped to standard fetal brain atlases. Three biochemical features were then systematically derived: cell-type composition via Azimuth annotation, neurotransmitter synthesis capacity through KEGG pathway enrichment, and metabolic flux estimates using a physics-informed neural network. These features served as inputs to a Temporal Graph Neural Network (TGNN), featuring memory modules and gated recurrent units (GRUs) for capturing temporal dependencies and spatial interactions. TGNN robustly predicted macroscale structural connectivity from fetal diffusion MRI data (GW21-38), significantly outperforming static models (mean squared error [MSE] < 0.05; $p < 0.001$, paired t-test). Conversely, MRI-derived connectivity matrices were analyzed using an Evolving Graph Convolutional Network (EvolveGCN) to generate dynamic embeddings, subsequently enabling multilayer perceptrons (MLPs) to accurately infer biochemical profiles (MSE < 0.05). Model validation included rigorous time-series cross-validation, biological perturbation simulations mimicking autism spectrum disorder (ASD) connectivity disruptions, and explicit controls against static and linear baselines. Sc/snRNA-seq data ($n > 10$ datasets) incorporated biological variability across brain regions; biological sex was included where available. The framework is publicly available as reproducible software, enhancing mechanistic understanding and enabling targeted early interventions in neurodevelopmental disorders.

Disclosures: Y. Wei: A. Employment/Salary (full or part-time): Oregon Health & Science University. C. Zhang: A. Employment/Salary (full or part-time): Oregon Health & Science University.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR310.13/WW9

Topic: J.06. Computation, Modeling, and Simulation

Support: DGAPA-PAPIIT UNAM IN207923

Title: Development of brain template and atlases for African Americans populations

Authors: ***P. ROSALES ZALDÍVAR**¹, D. ATILANO-BARBOSA², A. HERNANDEZ¹, A. RICO CHÁVEZ¹, F. A. BARRIOS³;

¹Univ. Autonoma de Queretaro, Queretaro, Mexico; ²Natl. Autonomous Univ. of Mexico, Queretaro, Mexico; ³Univ. Nacional Autonoma de Mexico, Querétaro, Mexico

Abstract: In the field of neuroimaging, structural brain templates are commonly used as they provide a standardized 3D coordinate framework for analyzing data from diverse subjects. This can be particularly applicable to the morphological analysis of a specific population with a defined age range and geographical location, thereby reducing bias in neuroimaging studies that focus on populations with diverse ethnic and racial backgrounds. In this study, a brain atlas was constructed using a T1 structural magnetic resonance imaging template with voxel sizes of 0.7 mm and 1 mm from an African American population (n = 167), obtained from the Human Connectome Project (HCP) database. The AFNI make template `dask.py` script was used for template creation. The process for generating the brain atlas involved using atlases generated in FreeSurfer, containing cortical parcellations for the identification and analysis of different brain regions. As a result, an average template, a group variance template, and a typical template were obtained, along with the FreeSurfer parcellation-based atlases corresponding to the Desikan-Killiany Atlas and the Destrieux Atlas. The template and atlases can be applied in the field of neuroimaging to enhance clinical studies or research focused on minorities, as a population-specific template may facilitate accurate diagnosis for improved treatment. **Keywords** brain atlases, brain templates, cortical parcellations, neuroimaging

Disclosures: **P. Rosales Zaldívar:** None. **D. Atilano-Barbosa:** None. **A. Hernandez:** None. **A. Rico Chávez:** None. **F.A. Barrios:** None.

Poster

PSTR310: Computational Approaches in fMRI or EEG II

Location: SDCC Halls B-H

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Program #/Poster #: PSTR310.14/WW10

Topic: J.06. Computation, Modeling, and Simulation

Support: NIH Grant R01MH60974
NIH Grant UM1MH130981

Title: Drums - a new tool for inter-subject cortical surface registration

Authors: *Y. WANG¹, C. YANG², T. COALSON³, D. C. VAN ESSEN⁴, M. F. GLASSER⁵;

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Abstract: In human neuroimaging, surface-based registration of cerebral cortex to a template is critical for enabling accurate comparisons across individuals despite folding variability. We introduce DRUMS (Diversely Reconstructed U-net with Multimodal Supervision), a deep learning framework for cortical surface registration. We used data from 1,071 subjects from the WU-Minn Young Adult Human Connectome Project. Multi-modal features were generated for each individual subject, including FreeSurfer “sulc” map, T1w/T2w myelin map, cortical curvature, cortical thickness and 76 components derived from PROFUMO-based resting-state fMRI networks. Subjects were split into three groups (475 for training, 475 for testing, 121 for validation); 45 subjects in the testing set were scanned twice, yielding two versions of the surfaces and feature maps for evaluating registration reproducibility. Also, 40 of the 45 test-retest subjects were scanned with task-fMRI, and these data will be used to evaluate the generalizability of the registration to held out modalities. We made three versions of the model: DRUMS-Sulc using only folding patterns, DRUMS-SupSulc using folding patterns but supervised with multi-modal features during training, and DRUMS-All using multi-modal features. The algorithm extracts and merges features from multiple scales using a U-net style network with geometric convolution. The resulting deformation field is generated from extracted features and positional encoding for vertices. Cross-correlation is used as similarity loss and a physically inspired strain energy regularization is used to penalize distortion. We evaluated results using similarity (cross-correlation), distortion (change of triangular area and shape on the sphere), reproducibility (cross-correlation) and task fMRI feature alignment (sum of vertex-wise t-statistics). DRUMS-Sulc outperformed MSMSulc with sulc feature alignment (0.84 vs 0.83) and reproducibility (0.99 vs. 0.98). DRUMS-SupSulc outperformed MSMSulc with myelin, RSN features and task features alignment (myelin: 0.75 vs. 0.71; RSN: 0.59 vs. 0.56; task: 219k vs. 206k) and reproducibility (myelin: 0.90 vs. 0.86; RSN: 0.59 vs. 0.58; task: 0.65 vs. 0.63). DRUMS-All outperformed MSMAll with myelin, RSN features and task feature alignment (myelin: 0.76 vs. 0.74; RSN: 0.64 vs. 0.62; task: 249k vs. 231k) and reproducibility (myelin: 0.90 vs. 0.85; RSN: 0.59 vs. 0.57; task: 0.64 vs. 0.62). In conclusion, DRUMS outperformed MSM in feature alignment for both trained and held out modalities. Also, we demonstrated the capability to supervise registration using modalities different from the input.

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Poster

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Program #/Poster #: PSTR310.15/WW11

Topic: I.04. Executive Functions

Support: AFOSR: Award# FA9550-23-1-0453

Title: Twobrain: a multimodal fnirs-fmri dataset to model deep-brain activity from surface signals

Authors: *M. CEKO, J. CRUM, L. HIRSHFIELD;
Univ. of Colorado Boulder, BOULDER, CO

Abstract: Functional MRI (fMRI) and functional near-infrared spectroscopy (fNIRS) are complementary neuroimaging modalities that measure hemodynamic responses. fMRI offers high spatial resolution and access to deep-brain structures, while fNIRS provides an ecologically valid approach towards real-world neuroscience but is limited to surface cortical signals. To bridge this gap, we aim to develop computational models that infer subcortical (deep-brain) activity from fNIRS data by leveraging concurrent fMRI data collection. We are collecting a large-scale multimodal dataset ('TWOBRAINS') combining 3T fMRI (Siemens Prisma, 32-channel head coil) and MRI-compatible fNIRS (NIRx Borealis, 110-channel cap covering frontal, parietal, temporal, and occipital cortices). Participants perform a battery of validated tasks spanning cognitive, affective, and sensorimotor domains that broadly engage cortical and subcortical networks: N-Back, Finger Tapping, Go/No-Go, Delayed Discounting, Negative Affect using aversive thermal and visual stimuli, and Naturalistic Affect & Cognition using movie clips. This dataset will serve as both a functional brain localizer and a training set for machine learning models capable of predicting deep-brain activity from fNIRS data. Brain responses will be related to individual differences in personality, cognitive function, and emotional regulation assessed via self-report. fMRI data are preprocessed using a standard fMRIPrep pipeline. fNIRS data are preprocessed to remove baseline shifts and motion artifacts, followed by a band-pass filter (Butterworth [.01, .5], 4th order) to remove physiological noise, and resampled from 5 Hz to 2.17 Hz to match the fMRI sampling rate. Preliminary GLM analyses of the fNIRS data during the N-back task in 14 subjects revealed significant group-level increase in activation for the 2-Back > Baseline contrast over the right vLPFC (Channel 52; BA45), $t(13) = -2.57$, $p = .03$, consistent with benchmark fMRI findings from the Human Connectome Project (HCP). However, activation in several other cortical areas implicated in the HCP dataset were not observed, likely due to a combination of insufficient sample size and increased signal noise observed during concurrent recording. Our goals with this work are to (1) benchmark the capacity of combined fMRI-fNIRS to map functional brain networks; (2) support machine-learning models to infer deep-brain activity from fNIRS; (3) provide an open resource dataset modeled after the HCP; and (4) bridge fNIRS and fMRI communities by providing a roadmap for multimodal brain mapping in real-world and laboratory settings.

Disclosures: M. Ceko: None. J. Crum: None. L. Hirshfield: None.

Poster

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Program #/Poster #: PSTR310.16/WW12

Topic: I.04. Executive Functions

Support: FA9550-23-1-0453

Title: Assessing fNIRS signal quality during concurrent fMRI-fNIRS and between testing environments

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Abstract: Previous research has used functional magnetic resonance imaging (fMRI) to validate functional near-infrared spectroscopy (fNIRS) as a hemodynamic method for human brain mapping (e.g., Noah et al., 2015), with the deoxygenated hemoglobin (HbR) signal from fNIRS best representing the BOLD signal. However, few studies have used concurrent imaging in which BOLD and fNIRS signals like HbR are collected simultaneously (e.g., Bulgarelli et al., 2018). The concurrent fMRI-fNIRS studies have been carried out have had limited spatial coverage over the cortex in terms of fNIRS channel configurations. It is unclear whether maximizing a montage of fNIRS channels to approach whole-head coverage will retain adequate signal quality during simultaneous recordings. The present work therefore assessed differences in the quality of fNIRS signals covering most of the cerebral cortex when moving from a lab-based setting to a scanner-based one. Using channel configurations containing over 100 fNIRS channels to obtain near whole-head coverage, two independent studies used n-back and somatomotor protocols from the Human Connectome Project (HCP) to evaluate changes in signal quality when moving from lab-based fNIRS to scanner-based fNIRS. Study 1 obtained lab-based fNIRS data from 36 healthy participants and Study 2 used concurrent imaging to acquire fMRI and fNIRS data from an independent sample of 14 healthy participants. Signal quality of the fNIRS data was evaluated using a coefficient of variation (CV) of 30%, where $CV(\%) = 100 \times SD(data)/Mean(data)$. The fNIRS data also consisted of two different wavelengths (850 nm, 770 nm), corresponding to HbO and HbR, respectively. If one wavelength for a given fNIRS channel exceeded the CV threshold, then both wavelengths were excluded. In Study 1, the lab-based fNIRS data indicated that 7% of channels should be excluded from further analysis. A subsample of this dataset was matched to the concurrent imaging datasets ($n = 14$), which indicated that 6% of channels were of poor quality. In contrast, the fNIRS data in Study 2 indicated that 36.27% of channels were of unacceptable signal quality. Although the signal quality results of Study 2 suggested that 63.73% of channels were still acceptable, the combined results of both studies indicated a ~30% decrease in signal quality when moving from a lab-based setting to that of a scanner. This decrease might be due to favoring whole-head coverage, which makes optimal and constant optode-scalp coupling more challenging. Opting for precision channel placement over regions of interest that are determined a priori might address this issue in future research using concurrent imaging.

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Poster

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Program #/Poster #: PSTR310.17/Web Only

Topic: I.04. Executive Functions

Support: FA9550-23-1-0453

Title: Towards Integrating fMRI and fNIRS Modalities: Evaluating Spatial Smoothing and Channel-Based Atlases for Enhancing BolT Model Classification

Authors: *A. ARNOLD, J. CRUM, M. CEKO, L. HIRSHFIELD;
Univ. of Colorado Boulder, Boulder, CO

Abstract: Transformer models have shown groundbreaking results in fMRI cognitive state decoding. Large fMRI datasets such as the HCP S1200 dataset enable training of deep learning models. In contrast, fNIRS datasets are typically small, private, and vary in brain coverage across labs, making them difficult to use in deep learning without overfitting due to model complexity and limited data. Recent work using transformer models on fMRI data has shown promising results in discriminating between different tasks (i.e., N-back vs. Motor) as well as between conditions within tasks (i.e., 0 vs. 1 vs. 2-Back). These models, such as BolT (Bedel H, et al., Medical Image Analysis. 2023), work by using extracted ROIs from fMRI data. We aim to develop a model that combines the spatial richness of fMRI with the portability of fNIRS by evaluating the effect of using different parcellations and spatial smoothing kernels on BolT model performance. To evaluate the model on fMRI data, we compared 0-Back and 2-Back conditions and the motor finger tapping conditions from the HCP S1200 data release. We also collected fNIRS datasets (n=28). The fNIRS channels were spatially registered into MNI space and used to create ROIs centered on these MNI coordinates. ROIs were extracted from the fMRI data using standard atlases (AAL or Brodmann) and from a 30mm sphere centered around fNIRS channel locations. Data were spatially smoothed using Gaussian kernels with full width at half maximum values of 0, 6, and 10 mm. Each model was evaluated by classifying task conditions (0- vs. 2-back; right vs. left finger tapping). For the N-Back task, the model performed well regardless of what atlas was used (AAL atlas, 94% accu.; 30mm spheres, 88% accu.; Brodmann atlas 91% accu.). For the motor task, both the AAL and 30mm spheres performed well with 94% and 95% accu. respectively, while the Brodmann atlas only achieved 66% accu.. This is likely because the Brodmann atlas does not differentiate between the two hemispheres of the brain, treating both motor cortices as a single region despite their distinct roles in the motor task. Smoothing had no significant effect on model performance, with accuracy differences across smoothing levels remaining within 2%. Given that the 30mm spheres performed well for both tasks, we believe this approach shows promise for pre-training transformer models on fMRI and fNIRS data, then fine-tuning on fNIRS data with the goal of developing a transformer model that generalizes to fNIRS data. While the resulting model still struggles to generalize to fNIRS data alone, our work shows the importance of atlas choice and how aligning signal locations across modalities is a critical step toward a unified model.

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Poster

PSTR310: Computational Approaches in fMRI or EEG II

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Program #/Poster #: PSTR310.18/WW13

Topic: J.06. Computation, Modeling, and Simulation

Title: Power Law Scaling Represents Hierarchical Structural Growth in Biologically Inspired Neural Networks Learning and Development

Authors: *S. F. LEITE^{1,2}, B. MOTA³, A. RAMOS SILVA⁴, A. THORNTON⁵;
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Abstract: Power law scalings are robust features of hierarchical networks with long-range connections, observed in the brain at both neuroanatomical and cognitive levels. At the **cognitive level**, the **Model of Hierarchical Complexity (MHC)** describes task performance as progressing through hierarchical developmental stages, indexed by the **Order of Hierarchical Complexity (OHC)**. Each stage enables higher-order learning by **coordinating at least two instances** of the previous stage in a **non-arbitrary manner**. At the **neural organization level**, the cortex is structured iteratively as a **circuit-of-circuits**, integrating neurons into motifs, motifs into columns, columns into functional areas, and areas into whole-brain architectures. In neurodevelopment, generations of outward-migrating neurons bootstrap increasingly complex networks. **Mammalian gyrification** follows a universal power law, driven in part by long-range axonal connectivity, resulting in **self-similar (fractal) cortices**. This principle likely generalizes to other networks where power-law scaling implies **self-similarity, optimizing for efficiency and simplicity** despite apparent structural complexity. The existence of scaling laws suggests deep, possibly shared underlying structures across these systems, which we seek **characterize and assess for their degree of universality**. We investigated whether artificial neural network learning exhibits **similar developmental scaling** after training on the **balance beam problem**. Using **MHC-informed task segregation**, inputs were divided into four OHC subsets (concrete, abstract, formal, and postformal). Minimal neural networks were trained independently, and the best-performing models were analyzed to assess **structural progression**. Results showed **structural progression** from abstract to formal and to postformal stages of complexity. **Three distinct self-organized structural progressions** emerged, **avoiding catastrophic forgetting**. Scaling dynamics showed a 120% neuron/connection increase followed by a 50% rise, an 80% increase in both cycles, and an 80% increase followed by 50% increase in neuronal growth and 25% in connection growth. Postformal structures **outperformed benchmarks**. Network complexity scaled with stage as $C(n) \propto (n-8)^{0.95}$. Its sub-linear growth mirrors biological brain development and reflects an **evolutionarily tuned balance** between **capacity** and

efficiency as cognitive tasks increase in hierarchical complexity. Our method offers a **scalable framework** for modelling **developmental and evolutionary growth** in artificial systems, potentially uncovering universal learning principles across domains.

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Poster

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Program #/Poster #: PSTR310.19/WW14

Topic: J.06. Computation, Modeling, and Simulation

Support: Ministry of Trade, Industry & Energy (MOTIE, Korea), under Grant 20012355 and NTIS 1415181023

Title: Diffusion-based reconstruction of high-density EEG signals in an EEG with a limited number of electrodes for portable brain-computer interfaces

Authors: *J. LEE, S.-P. KIM;

Biomed. Engin., Ulsan Natl. Inst. of Sci. and Technol., Ulsan, Korea, Republic of

Abstract: Wearable BCI systems need compact EEG systems for applications in daily life settings. However, reducing the number of electrodes largely sacrifices spatial resolution. We introduced a new method using denoising diffusion probabilistic models (DDPM) to synthesize high-density EEG recordings from sparse electrode setups. We assumed that DDPM might capture the spatial sparsity of scalp EEG and precisely recover direction information of missing channels by conditional generation. Five healthy participants participated in visual event-related potential (ERP) tasks and were recorded with 64-channel EEG. We have developed the inpainting task on top of a DDPM with conditionally sampled inputs. For the first experiments, either 30%, 50% or 70% of channels were masked randomly and models were trained on recovering the full scalp topography. For second spatial reconstruction experiments, we also select a reduced number of electrodes (6 channels) in either the temporal or occipital regions. The DDPM was trained with 1000 steps. Reconstruction performance was quantified by MSE between original and reconstructed signals in every 500ms window of the length 500ms. Analysis of the subject-level MSE results demonstrated that the MSE performed consistently with very low between-subject variability. Random channel selection at 50% also revealed very strong ERP component reconstruction, if you consider the respective characteristic components, such as P100, N170 and P300. It was shown that 6-channel configurations of either occipital or temporal regions could decode whole-brain activity patterns with a fair level of accuracy while preserving the spatiotemporal dynamics of visual processing. These results suggest that spatial correlations embedded in EEG signals can be exploited efficiently by the DDPM to recover high-resolution estimations with a limited number of recording electrodes. This methodology may

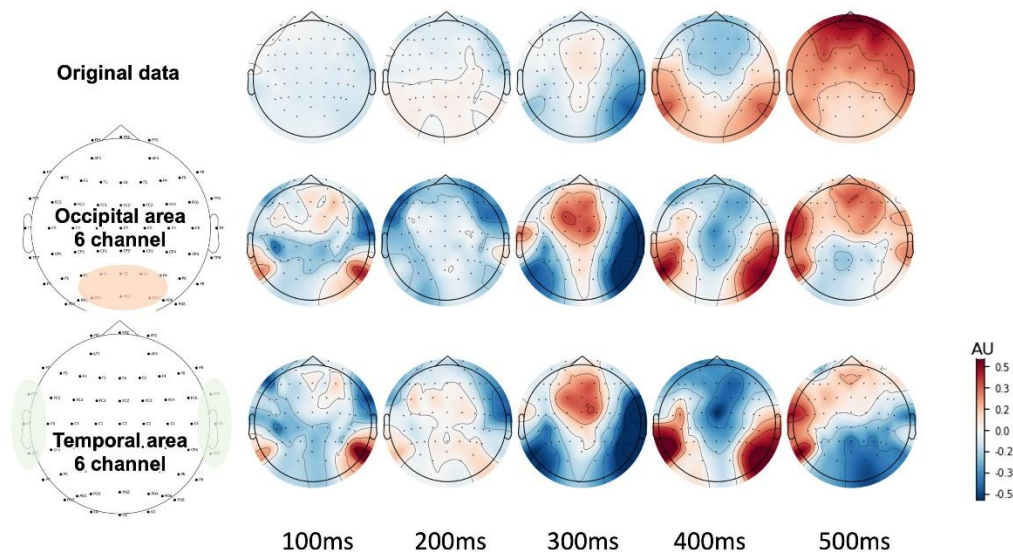
open the doors for the lightweight and portable BCI systems while reducing the hardware complexity.

Generative AI for Spatial resolution Enhancement

Inpainting test using scalp EEG

◆ Inpainting test : Conditional input

■ Area selection (6 channels)



Brain-Computer Interfaces Lab (BCI Lab)

Disclosures: J. Lee: None. S. Kim: None.

Poster

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Program #/Poster #: PSTR310.20/WW15

Topic: J.06. Computation, Modeling, and Simulation

Support: Del Monte Institute for Neuroscience, University of Rochester

Title: Estimating the Explainable Variance of EEG Responses to Natural Speech

Authors: *J. DOU¹, E. C. LALOR²;

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Abstract: How the human brain so effortlessly processes natural speech is incompletely understood. One useful approach to studying this centers on estimating linear encoding models - sometimes known as temporal response functions (TRFs) - that allow one to predict brain responses to speech based on various features of that speech. However, interpreting the predictive accuracy of such models is complicated by large inter-individual differences in the signal-to-noise of brain measurements and an inability to know what might constitute optimal model performance. Both issues would be addressed if it were possible to estimate the maximum explainable variance in brain responses to natural speech. A common approach to estimating the explainable variance involves computing correlations between responses to repeated presentations of the same stimulus. However, this approach is not best suited for natural speech, where repeated presentation of stimuli can lead to differences in brain activity due to changes in attention and the encoding of linguistic information. An alternative strategy is to assume that the brains of people who speak the same language should produce similar responses to the same stimuli. Indeed, using this strategy, previous studies estimated the explainable variance in ECoG and fMRI by calculating how accurately a selected subject's brain signal (target) can be decoded from the signals of other (source) subjects. Specifically, these studies: 1) fit decoders for a target's brain signals using different numbers of source subjects; 2) fit curves to model the relationship between the number of source subjects and the target subject's decoding accuracy. Here, we tailored this framework for EEG by 1) first applying multi-way canonical correlation analysis on source subjects' EEG to reduce the data dimensionality, 2) time-lagging the transformed data before ridge regression to incorporate the temporal differences between source and target EEG, 3) adopting a hierarchical version of bootstrapping to account for the inter-subject differences in EEG SNR. We tested this method on an EEG dataset that was collected from 19 subjects who each listened to about 1 hour of an audiobook. The obtained estimate of explainable variance was then compared with the prediction accuracy of a TRF model that was fit on different acoustic and linguistic speech features. The results showed that the explainable variance at centroparietal EEG channels was not fully explained by the TRF models. This established an important benchmark for future models of EEG responses to natural speech and provides a useful framework for controlling for inter-individual differences when using such models.

Disclosures: J. Dou: None. E.C. Lalor: None.

Poster

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Program #/Poster #: PSTR310.21/WW16

Topic: J.06. Computation, Modeling, and Simulation

Support: Air Force Research Laboratory (AFRL) under Contract No. FA8650-22-C-6441

Title: Development of Analytical and Realistic (Head-Shaped) Phantoms for Multimodal Calibration of Novel MEG and EEG Sensors

Authors: ***M. P. WEISEND**¹, B. RICHARDS², M. BRIDGES³, I. GERRARD⁴, D. M. EIDUM⁵, D. LABANOWSKI⁶, P. M. CONNOLLY⁷;

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Abstract: Motivation: Calibration of emerging sensors is critical for characterization of brain activity with electroencephalography (EEG) and magnetoencephalography (MEG). We present a suite of novel analytical (not head-shaped) and realistic (head-shaped) phantoms, along with supporting systems, to evaluate sensor performance in terms of signal Frequency, Amplitude, Latency, Location, and Duration (FALL-D). Approach: The analytical MEG phantom is a printed circuit board (PCB) configured with 20 magnetic dipole sources. Dipoles are 1 mm conductors spaced at 1 mm intervals. The dry EEG phantom is a PCB with an interface to connect directly to electrodes or headboxes from any EEG system. All phantoms have electronics that generate signals appropriate for EEG/MEG from standard function generators. We also developed head-shaped phantoms for EEG and MEG that have internal source distributions. The dry MEG configuration accommodates the analytical PCB phantom inserted into indexed slots in a head-shaped fixture. The wet configuration produces both MEG and EEG signals by embedding stripped coaxial cables in conductive gel (Leahy et al., 1998). Sources can be placed at any location inside the head. The 10-20 scalp locations have modular, indexed mounts to position EEG/MEG sensors at standard recording sites. Phantom output has custom software that maps recorded signals onto the surface geometry of the phantom in real time. Results: FALL-D using these phantoms have been validated on superconducting quantum interference devices (SQUIDS), optically pumped magnetometers (OPMs), and acoustically driven ferromagnetic resonance (ADFMR) sensors. Simultaneous EEG/MEG data have been acquired using OPM systems. Dry and wet EEG phantoms assessed signal quality on OpenBCI, Brain Vision, and custom Teledyne EEG systems. Conclusions: The modular, multimodal nature of this phantom suite enables flexible cross-modal calibration, reproducibility across platforms, and systematic benchmarking of emerging sensor technologies. Together, these tools support rigorous validation of next-generation EEG and MEG systems. Support: This material is based upon work supported by the Air Force Research Laboratory (AFRL) under Contract No. FA8650-22-C-6441. Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of AFRL.

Disclosures: **M.P. Weisend:** A. Employment/Salary (full or part-time): Teledyne Intelligent Systems Laboratory. **B. Richards:** None. **M. Bridges:** None. **I. Gerrard:** None. **D.M. Eidum:** None. **D. Labanowski:** None. **P.M. Connolly:** None.

Poster

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Program #/Poster #: PSTR310.22/XX1

Topic: J.06. Computation, Modeling, and Simulation

Support: NIH Grant 1R16NS140310-01

Title: Predicting Human Gait Events Onset using Deep Neural Networks

Authors: T. GIAN¹, H. FEKRI AZGOMI², D. D. WANG², *G. KUMAR¹;

¹Chem. and Materials Engin., San Jose State Univ., San Jose, CA; ²Neurolog. Surgery, Univ. of California San Francisco, San Francisco, CA

Abstract: Introduction: Gait impairment is a debilitating symptom of Parkinson's disease (PD), profoundly affecting patients' quality of life. This study develops a deep neural network (DNN) framework to predict gait events using DBS parameters and the time-series data of canonical frequency band power derived from local field potential (LFP) recordings from globus pallidus internus (GPi) and electrocorticography (ECoG) from motor and premotor cortices from a subject during walking. Our goal is to utilize the developed model within a model predictive closed-loop control framework to optimize the DBS settings, achieving an optimal pattern of gait events during ambulation. Accurate prediction of these events is crucial for improving gait kinematics, particularly stride timing and bilateral symmetry-key indicators of motor function in PD.

Methods: We developed DNN models to predict the onset of Right Toe-Off (RTO), Right Heel-Strike (RHS), and Left Toe-Off (LTO) using neural signals recorded from GPi and motor and premotor cortices in both hemispheres. Data were collected from one PD patient with implanted DBS over 6 walking sessions, each using distinct left/right DBS amplitude and frequency, resulting in 524 gait cycles. LFP and ECoG signals from 6 recording channels were decomposed into delta (1-4 Hz), theta (4-8 Hz), alpha (8-13 Hz), beta (13-30 Hz), and low gamma (30-50 Hz) bands, yielding 30 neural features. These were combined with 4 DBS parameters, resulting in a 34-dimensional input per gait cycle. For event prediction, we used time-resolved spectral features: the first 60 ms of a gait cycle was used to predict RTO onset; the segment from LHS up to the onset of RTO for RHS; and the segment from LHS to RHS for LTO. Two modeling approaches were used. In the first, each of the 6 sessions was split into 90:10 for training/testing, with pooled data utilizing all 34 features. In the second, we assessed cross-hemisphere generalization by training on all the gait cycles using 15 neural features from the left hemisphere and 4 DBS parameters, and testing on the corresponding features from the right hemisphere. Performance was measured using mean absolute percentage error (MAPE), averaged over 10 runs.

Results: The optimized models successfully predicted gait event timing with low error. In the full-channel model, MAPEs were 5.361%, 2.320%, and 1.352% (± 0.473 , ± 0.212 , ± 0.175) for RTO, RHS, and LTO, respectively. The cross-hemisphere model achieved MAPEs of 5.734%, 2.406%, and 1.142% (± 0.211 , ± 0.123 , ± 0.039).

Conclusion: DNNs can accurately predict gait event onsets. Integrating such models into a closed-loop adaptive DBS system may enable real-time improvements in gait function.

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Poster

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Program #/Poster #: PSTR310.23/XX2

Topic: J.06. Computation, Modeling, and Simulation

Title: Capturing Global and Local Features from low and high frequency ECoG signals with ResNet and Transformer

Authors: *W. YANG^{1,3}, C. ZHENG¹, H. CUI²;

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Abstract: Electrocorticogram (ECoG) is a promising long-term signal acquisition method for decoding brain activity, enabling applications in rehabilitation and external device control. Recent advances in deep learning have demonstrated the potential of decoding these signals. However, the inherent spatio-temporal variability of ECoG signals presents challenges in accurately extracting meaningful characteristics for various tasks. Here, we adopted two distinct classification models based on the principle that global and local features can be extracted from low and high frequency components, respectively. One model is an 8-layer Residual Network (ResNet), preceded by two 1D convolution layers (Conv1D): the first with a 64×64 kernel for the 4-30 Hz frequency range, and the second with a 32×32 kernel for the 30-70 Hz range. The second model is a hybrid of CNN and Transformer, with the Transformer module based on the HiLo framework. The prediction times for the two models are approximately 2 ms for ResNet and 30 ms for the hybrid model, both suitable for online decoding. The data used for training the models was recorded from a paralyzed patient implanted with a 128-channel flexible epidural ECoG electrode. The four motor intentions decoded include extending the right wrist and opening the fist, lifting the right leg, moving the left thumb, and lifting the left leg. Data from seven consecutive days was selected and used as follows: day 1 to predict days 6-7; days 1-3 to predict day 6-7; and days 1-5 to forecast days 6-7. As the number of days increased, the decoding accuracy could exceed 85%. Our analysis revealed that, with smaller data volumes, ResNet outperformed the CNN-Transformer in decoding performance. However, as the data accumulated, Transformer's decoding performance improved progressively. Using PCA and UMAP dimensionality reduction to analyze the features extracted by ResNet and CNN-Transformer, we found that the representational drift across days was only captured by the CNN-Transformer features but not ResNet. Furthermore, the CNN-Transformer-based model was able to capture more nonlinear information compared to the ResNet-based model. The CNN-

Transformer model proved more capable of capturing subtle changes in brain activity from ECoG signals, whereas ResNet was less sensitive to such fine-grained changes. For simple classification tasks with insufficient data, ResNet seems to perform better. However, when ample data is available and more signal variation is required to capture for complex analysis, the Transformer model is more appropriate.

Disclosures: W. Yang: None. C. Zheng: None. H. Cui: None.

Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.01/XX3

Topic: J.08. Methods to Modulate Neural Activity

Support: GE HealthCare Research Funding

Title: Assessment of Deep Learning MRI Reconstruction for Connectomic-Guided Deep Brain Stimulation in Depression

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Abstract: Subcallosal cingulate deep brain stimulation (SCC-DBS) is a promising intervention for treatment-resistant depression (Choi and Mayberg, 2023). Connectomic DBS targeting uses individual diffusion tractography to optimize electrode placement, aiming to modulate the intersection of key white matter tracts: the cingulum bundle, forceps minor, and uncinate fasciculus (Riva-Posse et al., 2014). However, conventional diffusion-weighted imaging (DWI) has well-known limitations that compromise tractography accuracy due to low signal-to-noise ratio and image artifacts (Maier-Hein et al., 2017). To address these issues, we applied a novel deep learning-based MR reconstruction method, AIRTM Recon DL, which enhances SNR and image sharpness while reducing truncation artifacts (Lebel, 2020), and evaluated its feasibility in SCC-DBS surgical planning. Raw DWI data from seven patients who underwent SCC-DBS were processed using both conventional and DL reconstruction pipelines. Probabilistic tractography was performed for each dataset to quantify the amount of white matter stimulation using bilateral SCC seeds, based on prior coordinates associated with therapeutic response (Song et al., 2023). The stimulation maps were normalized by the total number of streamlines (waytotal) and thresholded at 0.001%, indicating voxels structurally connected to the SCC. The amount of stimulation was assessed in key white matter tracts targeted in SCC-DBS surgical planning. Results demonstrated that conventional reconstruction failed to capture complete trajectories of the uncinate fasciculus, particularly its projections from the SCC to the amygdala, compared to DL reconstruction. Quantitative analysis using nonparametric paired tests confirmed

significantly greater connectivity and sensitivity proximal to the insular cortex with DL reconstruction compared to the conventional method ($p = 0.016$). These findings support the use of DL reconstruction to enhance the anatomical fidelity of diffusion tractography in clinical DBS planning. By mitigating acquisition artifacts without increasing scan time, this method enables more precise connectomic targeting and may improve treatment outcomes in connectome-guided neuromodulation. Furthermore, integrating DL-enhanced imaging into standard clinical workflows may broaden access to personalized, circuit-based DBS approaches.

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Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.02/XX4

Topic: J.08. Methods to Modulate Neural Activity

Support: American Lebanese Syrian Associated Charities (ALSAC) in the form of salaries to all of the authors, equipment, and other expenses
St. Jude Cancer Center Developmental Funds Award – winter 2025.

Title: Cognitive rehabilitation for survivors of childhood medulloblastoma using real-time fMRI neurofeedback: a design for a pilot study

Authors: ***C. TORRES ROJAS**¹, P. PANDEY¹, S. S. MCAFEE¹, P. ZOU¹, J. DALBONI DA ROCHA¹, J. M. ASHFORD², Y. LI³, R. GILES⁴, H. CONKLIN², R. SITARAM¹;

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Abstract: Introduction: While survival rates for children diagnosed with brain tumors have increased to over 75% [1], there is growing concern about the high risk of cognitive deficits as sequelae of disease and cancer treatment [2,3]. Neurocognitive assessments demonstrate impaired working memory (WM) in medulloblastoma survivors [2,3,4]. WM is a cognitive function that holds and manipulates information for short periods. We lack a complete understanding of treatment-related effects on this brain function. Our long-term research goals are to characterize and predict the impact of cancer and its treatment on brain function and

develop cognitive interventions to mitigate neurocognitive impairments in this population.

Methods: This project implements a randomized, 2:1 (intervention: control) double-blind controlled trial. Using closed-loop fMRI neurofeedback, the intervention group (n=20) will receive neurofeedback from the bilateral dorsolateral prefrontal cortex (DLPFC), and the control group (n=10) will receive neurofeedback from the primary auditory cortex, a region considered not directly involved in WM. **Aim 1** will address the feasibility, survivor interest, and acceptability of rt-fMRI neurofeedback during three training sessions. **Aim 2** seeks to estimate from fMRI images the percent change in DLPFC activity and its effect sizes during up-regulation compared to the baseline condition. **Aim 3** will broadly assess the efficacy of neurofeedback training on WM performance measures and parent-reported symptoms of executive dysfunction.

Results: We designed a Bayesian monitoring rule to monitor whether the data suggests the participation rate is lower than 60%. A modulation index (up-regulation minus rest) will be calculated for activity in DLPFC in each neurofeedback run, and changes in the modulation index over time will be used to quantify neurofeedback learning. The percentage of Blood Oxygenation Level-Dependent (BOLD) changes in the DLPFC will be compared across time and between study groups to determine the effect of neurofeedback training on DLPFC activation during N-back (i.e., 2-back vs. 0-back condition). The correlation between brain activation and cognitive performance will be evaluated in all participants to help identify regional and network activity contributing to good cognitive performance in these survivors. **Summary:** This study will provide evidence of the feasibility of rt-fMRI neurofeedback training in childhood medulloblastoma survivors regarding methodology, participation rate, adherence, and side effects.

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Poster

PSTR311: Network Activity Modulation

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Program #/Poster #: PSTR311.03/XX5

Topic: J.08. Methods to Modulate Neural Activity

Support: Grant from Samsung Open Innovation Initiative

Title: Slow wave entrainment with a smartwatch improves sleep quality and mood

Authors: N. W. WHITMORE¹, S. CHAN³, A. DULSKI², A. PODRUG², N. HIDALGO², N. OBI², V. VISWANATH⁴, V. NATHAN⁴, P. MAES²;

¹Media Lab., ²MIT, Cambridge, MA; ³Nanyang Tech. Univ., Singapore, Singapore; ⁴Samsung Res. America, Mountain View, CA

Abstract: In the past decade, research has shown that slow electrical oscillations in the cortex (slow waves) play an important role in the restorative functions of sleep. Key evidence for the causal role of slow waves has emerged from studies of slow wave entrainment in which rhythmic sounds during non-REM sleep are used to increase slow wave amplitude. Slow wave entrainment has been shown to improve multiple cognitive and physiological functions including memory, executive function, and immune function. These findings suggest that slow wave entrainment could be a valuable intervention for improving health; however the current systems used for slow wave enhancement are expensive, cumbersome, and fragile, limiting their use outside of research. To improve the usability of slow wave stimulation, we created a smartwatch app to stimulate slow waves using rhythmic vibration and sound. Stimuli are controlled using a machine learning model which uses motion and heart rate data to predict the optimal stimulus timing and intensity. In our first study with 96 participants ages 18-92 years, we tested whether smartwatch-based stimulation could increase EEG delta power comparable to previous results. Stimulation increased frontal EEG delta power compared to no stimulation, replicating previous findings. Participants also showed less sleep fragmentation in stimulation periods compared to matched non-stimulation periods. Participants with a large delta increase during stimulation showed improvements in self-rated sleep quality and mood and reduced errors on a trail-making task following stimulation. Participants with poor baseline mental health showed significant improvements on confusion and tension axes of the Brunel Mood Scale following stimulation. Our second study involved 663 participants ages 18-80 who used their own smartwatches. Participants received nights with no entrainment, entrainment with sound, and entrainment with vibration in randomized order. Ratings of overall mood and fatigue were improved following sound entrainment compared to no entrainment; vibration entrainment did not improve these measures. Our results provide additional evidence for the restorative role of slow wave activity in large, diverse cohorts and also demonstrate that slow wave entrainment using a smartwatch is a promising approach for improving sleep quality and mental health.

Disclosures: **N.W. Whitmore:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **S. Chan:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **A. Dulski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **A. Podrug:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **N. Hidalgo:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **N. Obi:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **V. Viswanath:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **V. Nathan:** A. Employment/Salary (full or part-time): Employee of Samsung Research America. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder,

excluding diversified mutual funds); Inventor named in the patent for the smartwatch slow wave entrainment system. **P. Maes:** 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.; Received grant from Samsung to conduct the study.

Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.04/XX6

Topic: J.08. Methods to Modulate Neural Activity

Title: Maestro-eeg: methods for brain-body coupling in piano music listening over multiple sessions

Authors: ***A. UPPAL**¹, V. JALTARE¹, J. HUANG², M. LEE¹, G. FRANK³, S. SEBT¹, Y. XU⁴, J. LEUGERING⁴, V. R. DE SA⁵, G. CAUWENBERGHS¹;

¹Shu Chien-Gene Lay Dept. of Bioengineering, ²Computer Sci. and Engin., ³Electrical and Computer Engin., ⁴Inst. for Neural Computation, ⁵Cognitive Sci., UCSD, La Jolla, CA

Abstract: Music perception occurs at the intersection of sensory stimuli and predictions generated from past listening experiences (Vuust et al., 2022). But how does music perception change with experience, as we become more familiar with music stimuli through listening repetitions separated over days? To address this, we record brain (EEG), cardiac (ECG), and respiratory (nasal) rhythms from subjects listening to music longitudinally. EEG is recorded non-invasively from the scalp using a dry-contact wireless headset (CGX Quick-32r) with 32 channels in standard 10-20 montage (reference=A1), including an auxiliary channel recording chest ECG (gel pad). Respiration is recorded as nasal pressure (airflow) with subjects breathing into a nasal cannula connected to a pressure sensor, synchronized with EEG through LabStreamingLayer (Kothe et al., 2024). Music stimuli are from the publicly-available Magenta MAESTRO virtuoso piano performance dataset (Hawthorne et al., 2018), providing audio and ground-truth annotated MIDI from virtuoso pianists performing on a Yamaha Disklavier piano. Data collection is ongoing, and here we present pilot analysis for six listening sessions (3 hours) spaced over 3 weeks from N=1 musician subject (male, age=31). For each listening session lasting approx. 30 minutes, the subject listened to complete piano performances randomly selected and ordered from a subset of 12 MAESTRO stimuli (6 compositions, two performers each). The subject was instructed to listen to the music with eyes closed, with breaks and 1-minute long eyes-closed silent resting trials between listening trials. The subject reported never having heard the played music before, but being able to recognize the repeating pieces between sessions, and developing a liking or disliking for certain pieces. To explore changes in the subject's brain-body coupling with increasing familiarity to the stimuli, we assess (1) magnitude-squared coherence of scattering spectral coefficients (Andén et al., 2019) for all sensed modalities, and (2) phase-locking between ECG R-peaks and underlying musical beat (Heydari

et al., 2021). Finally, we propose adopting cortical slice models (Neymotin et al., 2020) to explore how ongoing EEG rhythms could be shaped by variations in corticocortical inputs over time, assuming preserved stimulus-derived thalamic inputs across listening sessions. Our methods could inform the design of sound-based neurofeedback therapies to gauge how a subject's receptivity to stimuli changes with acclimatization to music samples over time.

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Poster

PSTR311: Network Activity Modulation

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Program #/Poster #: PSTR311.05/XX7

Topic: J.08. Methods to Modulate Neural Activity

Support: NIH R16 Award 1R16NS140310-01

Title: Closed-loop model predictive control of pathological beta-oscillations in a computational model of cortico-basal ganglia circuit

Authors: *D. Q. TRAN¹, T. GIANG², G. KUMAR²;

¹Computer Engin., ²Chem. and Materials Engin., San Jose State Univ., San Jose, CA

Abstract: Introduction: Pathological beta-oscillations (13-30 Hz) in the subthalamic nucleus (STN) have been found to be strongly correlated with motor impairments in Parkinson's disease (PD). Traditionally, Deep Brain Stimulation (DBS) uses fixed stimulation parameters, which limit its adaptability to the brain's dynamic neural activity. In contrast, adaptive (or closed-loop) DBS, which dynamically adjusts stimulation parameters based on real-time neural feedback, has been shown to be promising in suppressing pathological beta-oscillations and easing Parkinson's symptoms. In this work, we propose a predictive model-based multi-input multi-output (MIMO) closed-loop optimal control approach that is designed to proactively control pathological beta oscillations. Our method seeks to dynamically optimize DBS parameters by taking into account several therapeutic goals within a single optimization framework.

Methods: We used the biophysical computational model of the cortico-basal ganglia (CBG) circuit, validated with PD patients' data in earlier research, to generate synthetic data under various DBS parameter settings (amplitude, frequency). With this collected data, we developed a data-driven Long Short-Term Memory (LSTM) neural network model to predict beta-oscillation patterns in response to the DBS parameters. By integrating this model within a model predictive control framework, we designed an optimal control policy to optimize the DBS parameters in a closed-loop to achieve the desired beta oscillations.

Results: Our results show that the LSTM model can predict the beta-oscillation patterns with high accuracy. By integrating the LSTM model within the model predictive control. We

demonstrate the efficacy of our closed-loop control design in controlling the beta oscillations effectively under various scenarios.

Conclusions: Our model-based predictive closed-loop strategy is capable of incorporating various clinically relevant objectives and constraints, and could potentially open the door to the development of the next generation DBS protocols that can deliver autonomous, real-time neuromodulation based on the individual's specifications

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Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

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Program #/Poster #: PSTR311.06/XX8

Topic: J.04. Physiological Methods

Title: Electrophysiological Correlates of Nadamay Meditation Compared to Mind Wandering

Authors: *K. MEGHA¹, A. SASIDHARAN², G. KUMAR², A. ROJ¹, S. PATRA¹;

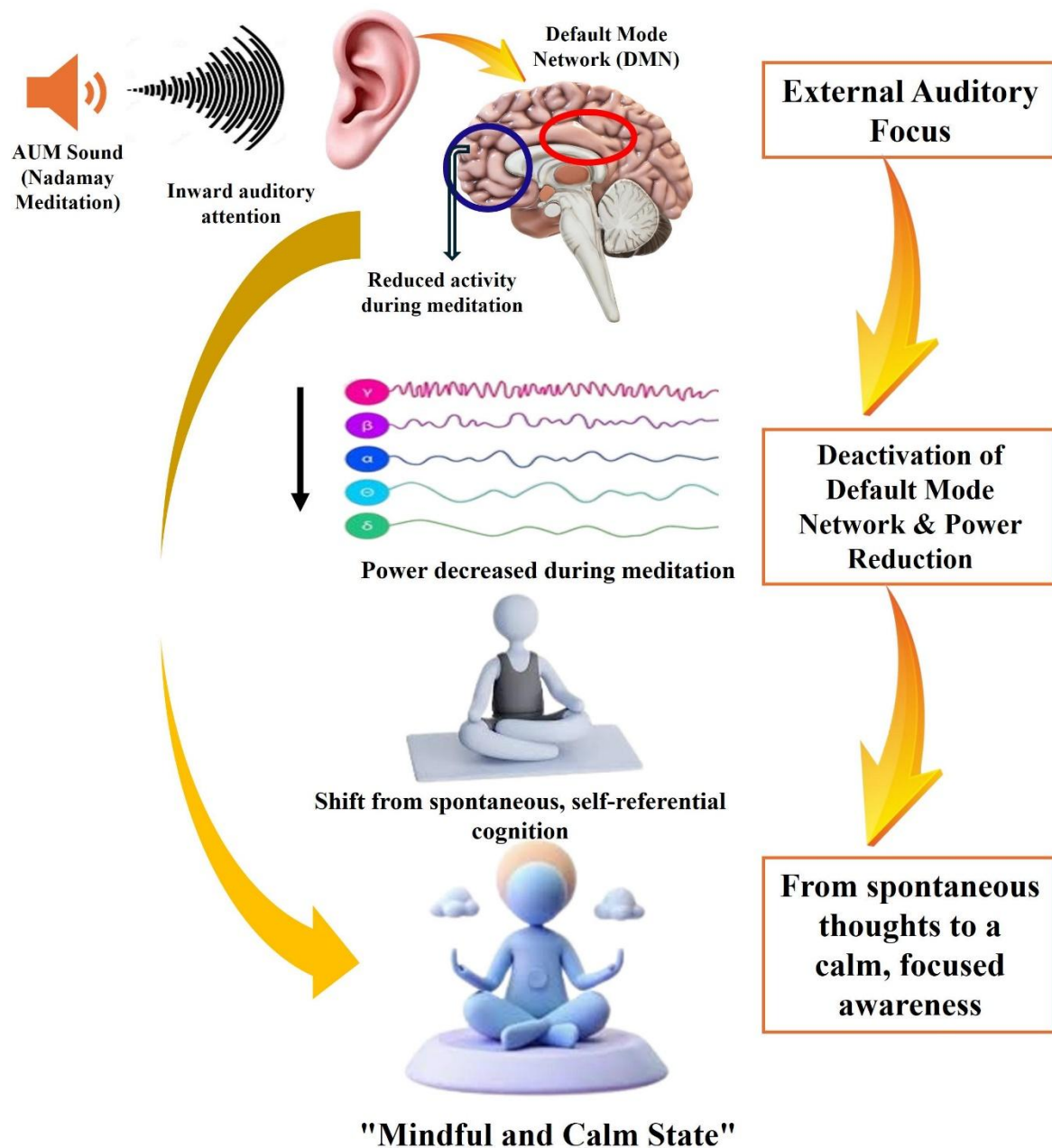
¹Dept. of Yoga, Central Univ. of Rajasthan, Ajmer, India; ²Dept. of Neurophysiol., Natl. Inst. of Mental Hlth. and Neurosci., Bengaluru, India

Abstract: Background: Meditation fosters attention and emotional regulation, often marked by increased EEG alpha and theta power, reflecting relaxed focus. Sound-based practices, rooted in ancient traditions like the Nāda Bindu Upanishad, leverage auditory stimuli such as AUM recitation to deepen internal awareness, yet their neural dynamics are underexplored.

Objectives: This study explored the brain activity and subjective experience of Nadamay Meditation, an auditory-guided practice, compared to a mind-wandering condition. The study aimed to uncover distinct EEG spectral patterns and changes in alertness during this unique meditation technique. **Methods:** Fifteen healthy adults (age 24.8 ± 3.6 years, 9 female, 6 male) completed two 26-minute sessions (Nadamay Meditation and mind wandering) in a randomized, counterbalanced setup. EEG data were recorded with a 64-channel actiCHamp system and power spectral density was calculated across delta, theta, alpha, beta, and gamma bands. Subjective alertness was assessed before and after sessions using the Stanford Sleepiness Scale (SSS).

Yuen's paired t-test with FDR correction was used for statistical comparisons. **Results:** Nadamay Meditation significantly lowered EEG power across all frequency bands, especially in prefrontal and frontocentral areas, compared to mind wandering ($p < .05$, FDR-corrected). Reduced delta, theta, and alpha power attributed to decreased cortical idling and improved attentional focus, while decreased beta and gamma power suggested decreased cognitive effort. Participants reported feeling more alert after Nadamay Meditation (93.3% alert) compared to less alert after mind wandering (73.3% alert). **Conclusions:** Nadamay Meditation produces distinct brain activity changes, marked by widespread EEG power reduction and increased alertness, likely reflecting suppressed default mode network activity. These results highlight its potential as

an effective, sound-based practice for boosting attention and mental clarity, adding valuable insights to the neuroscience of meditation.



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Poster

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Topic: J.08. Methods to Modulate Neural Activity

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R01DA055439
R01NS118461

Title: Closed-loop control of prefrontal population activity via patterned microstimulation in awake primates

Authors: I. MORAN MARTINEZ¹, G. BARZON³, A. DE⁴, C. CARNAHAN³, L. MAZZUCATO⁵, *R. KIANI²;

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Abstract: Electrical stimulation of brain circuits is widely used to treat neurological disorders, including Parkinson's disease, epilepsy, and depression. However, current approaches usually rely on single-electrode stimulation and trial-and-error to identify effective patterns. A critical gap remains in our ability to predict and shape the effects of complex spatiotemporal stimulation on neural population activity and behavior, needed to achieve neural control and deliver targeted therapies. Here, we designed a real-time, closed-loop neural control system capable of precisely sculpting spatiotemporal patterns of population spiking activity in the prefrontal cortex of awake monkeys. We developed a Brain-Computer Interface for simultaneous electrophysiological recording and electrical microstimulation, using a 96-channel Utah array chronically implanted in the prearcuate gyrus. Stimulation was delivered via the same device. We developed Input/Output Control (I/O-Ctrl), a novel algorithm enabling efficient, data-driven control of evoked neural activity, in three steps. First, we identified a subset of stimulating electrodes ("hubs") based on causal functional connectivity estimated on-the-fly during rest (Nejatbakhsh et al., 2023), avoiding trial-and-error identification of the most effective electrodes. Second, using a short train of random stimulation patterns, we modeled the input-output relationship between stimulation and evoked population activity. Third, we synthesized optimal stimulation patterns to evoke desired target responses and tested their effects. We found that the evoked population activity significantly correlated with the target patterns and this correlation scaled with the amplification of the target pattern, as predicted by our algorithm. The inferred "stimulation fields" characterized the spatiotemporal population response to multi-electrode stimulation, revealing principles of spatial and temporal summation underlying evoked responses. These findings establish the feasibility of control theory for predicting and shaping population activity in real time, paving the way for rational design of stimulation protocols tailored to achieve desired circuit level activity patterns, with implications for both neuroscience and clinical neuromodulation.

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Poster

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Topic: J.08. Methods to Modulate Neural Activity

Support: NIH Grant R01MH127375

Title: Causal Flow: Fast and Robust Estimation of Directed Interactions in Complex Neural Systems

Authors: *C. CARNAHAN¹, R. KIANI³, L. MAZZUCATO², A. DE⁴, G. BARZON⁵; ²biology, ¹Univ. of Oregon, Eugene, OR; ³Ctr. for Neural Sci., New York Univ., New York, NY; ⁴Inst. of Neuroscience, Univ. of Oregon, Eugene, OR; ⁵Padova Neurosci. Ctr., Padova, Italy

Abstract: Biological neural circuits are characterized by dense recurrent connectivity, weak pairwise correlations, and nonlinear dynamics, making it challenging to infer how activity at one node influences others in the network. Yet, such inference is critical for designing effective neural perturbation strategies and for developing neuromodulation therapies for conditions like Parkinson's disease and depression. Here, we quantify directed functional interactions within neural circuits using Causal Flow (CF), an extension of Convergent Cross Mapping. CF estimates the causal influence one node exerts on another solely based on short recordings of spontaneous activity. Crucially, CF can distinguish genuine causal interactions from common input effects, can quantify both interaction strength and time lags, and it is robust to nonlinear dynamics and partial observability. We validated CF on diverse simulated dynamical systems and recurrent neural networks, where it consistently outperformed alternative methods, including Granger Causality (GC) and Transfer Entropy (TE). To enable real-time use, we developed an efficient implementation of CF that reduces computational cost while preserving accuracy, enabling the rapid identification of "hub" nodes, which exert the strongest causal influence on the rest of the network. Hub nodes can be used to guide targeted intervention. Finally, we applied CF to multi-electrode recordings from the macaque pre-arcuate gyrus. Using recording and electrical microstimulation with chronically implanted Utah arrays, we found that stimulation of hub nodes influenced the population activity more significantly. Further, hub nodes remained largely consistent over days and weeks, indicating a stable causal network architecture in the primate prefrontal cortex. Our results establish CF as a powerful tool for uncovering functional architecture in recurrent circuits and optimizing closed-loop neuromodulation strategies in both basic and clinical neuroscience.

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Poster

PSTR311: Network Activity Modulation

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Program #/Poster #: PSTR311.09/XX11

Topic: J.08. Methods to Modulate Neural Activity

Title: Optimal control of neural activity in circuits with excitatory-inhibitory balance

Authors: *G. BARZON¹, R. KIANI², L. MAZZUCATO³;

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Abstract: Two influential theories in neuroscience are the "computation through dynamics" framework and the "efficient coding hypothesis." The former posits that neural computations are revealed by the geometry of low-dimensional trajectories of activity in state space (Mante et al., 2013; Vyas et al., 2020). The latter holds that sensory systems maximize information transmission under biological constraints (e.g., metabolic cost and limited bandwidth) by reducing redundancy and producing sparse representations (Barlow, 1961; Olshausen & Field, 1996). However, the efficient coding hypothesis is largely static and does not account for the dynamic nature of neural responses. Here, we propose a novel dynamical extension of efficient coding: the "minimum input energy principle." According to this principle, the incoming projections to a local neural circuit are sculpted during development and learning to minimize deviations from the input baseline while maximizing the information content of the resulting trajectory in state space. This principle emerges naturally from interpreting input signals as "minimum-energy optimal controllers" that steer local circuit dynamics. We apply this framework to cortical circuits characterized by complex excitatory-inhibitory (E/I) interactions, which are known to shape both neural dynamics and coding efficiency (Sanzeni et al., 2020; Denève & Machens, 2016). We demonstrate how E/I balance and recurrent interactions sculpt a circuit's controllability and how this manifests as balanced amplification of input-driven responses. We capture the spatiotemporal features of cell-type specific control in spatially extended networks by introducing two new observables: controllability horizon and Gramian dimensionality. Extending these insights to networks performing cognitive tasks, we highlight control theory as a unifying framework for understanding both information processing and causal circuit manipulations. We suggest that the minimum input energy principle also extends to inter-areal interactions, illuminating the logic of i) inter-areal communication, ii) top-down and bottom-up contextual modulations, and iii) whole-brain dynamics. Together, this work proposes a biologically grounded dynamical theory of efficient coding and offers new tools for probing the functional architecture of neural circuits.

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Poster

PSTR311: Network Activity Modulation

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Topic: J.08. Methods to Modulate Neural Activity

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Pew Innovation Fund (00037214)
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Title: Model based design of causal manipulations of recurrent network activity with high spatiotemporal precision

Authors: *A. DE¹, R. KIANI³, L. MAZZUCATO²;

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Abstract: Recent advancements in neurotechnology have made it possible to deliver precise spatiotemporal perturbations to neural circuits with single-cell resolution. However, current methods for selecting stimulation sites commonly rely on laborious trial-and-error procedures that do not scale well to multi-site, temporally-structured perturbations. Efficient methods to design complex perturbation patterns are urgently needed. To address this limitation, we present a computationally efficient framework for designing multi-site spatiotemporal stimulation patterns by combining approaches from the theory of dynamic mode decomposition (DMD) and control theory (Proctor et al., 2016). Our goal is to steer neural population activity—and ultimately behavior—toward a desired target state through optimized input patterns. We demonstrate the feasibility of our approach using recurrent neural network (RNN) models with nonlinear and chaotic dynamics. First, we linearize the nonlinear dynamics by embedding the activity state in a high-dimensional delay space, where the evolution of network states can be approximated as a linear transformation. In this space, we apply linear control theory to design optimal stimulation sequences that drive the system from an initial state to a target state. Our simulations reveal a trade-off between the number of stimulated neurons, the duration of stimulation, and the distance between the desired target and final states, similar to classical linear systems. We systematically quantify the minimum number of stimulated neurons and the control duration required to minimize the error between the desired and actual final states. Our results establish the feasibility of applying model-based control to high-dimensional nonlinear neural systems and pave the way for achieving targeted manipulations of brain activity. This framework has broad implications for experimental neuroscience and neuromodulation therapies, offering a scalable alternative to trial-and-error stimulation approaches.

Disclosures: A. De: None. R. Kiani: None. L. Mazzucato: None.

Poster

PSTR311: Network Activity Modulation**Location:** SDCC Halls B-H**Time:** Tuesday, November 18, 2025, 8:00 AM - 12:00 PM**Program #/Poster #:** PSTR311.11/YY1**Topic:** J.08. Methods to Modulate Neural Activity**Title:** Toward Seizure Prevention Using Passivity-Based Control of Epileptiform Activity**Authors:** ***G. ACHARYA**¹, A. HUANG¹, V. SANTHAKUMAR², E. NOZARI³;²Mol. Cell and Systems Biol., ³Mechanical Engin., ¹Univ. of California, Riverside, Riverside, CA

Abstract: While neurostimulation has shown great promise for treating various neurological and psychiatric disorders, its efficacy in treating epileptic seizures has remained limited. In particular, existing neuromodulation systems that are FDA-approved for drug-resistant epilepsy seek to prevent seizures by, ironically, injecting energy into an already hyperexcitable and/or unstable system. While this may occasionally succeed via desynchronizing or hyperpolarizing neuronal populations and/or long-term plasticity, rates of seizure freedom have remained limited to about 1 in every 5 implanted patients. In this work, we propose a passivity-based control (PBC) scheme for the treatment of drug-resistant focal epilepsy via neuromodulation. In contrast to active pulse-based stimulation, PBC drains energy from the epileptic tissue, hence “cooling” the tissue and stopping the initiation/propagation of an impending/ongoing seizure. Rooted in classical control theory, passivity-based control approaches have been extensively studied and successfully applied in a variety of engineering systems for over half a century, but ironically have not yet been used for treating epileptic seizures. In this work, we combine PBC with a model-based seizure predictor to form a closed-loop, analogue control system for seizure monitoring and suppression. We demonstrate the power of PBC using a biophysically realistic dentate gyrus model (Santhakumar et al., 2005) that is capable of generating both evoked and spontaneous focal seizure-like activity. For the case of evoked seizure-like activity, we have consistently found that PBC is sufficient to fully suppress ictal spiking with only 20 ms of stimulation, even when control was delayed up to 30 ms after seizure onset, reducing ictal spiking by 94%. In the spontaneous case where seizure-like events originated randomly within the network, PBC remained highly effective, achieving a 93% reduction in seizure duration and a 92% reduction in ictal spiking. Our findings demonstrate the promise of PBC as a robust, paradigm-shifting closed-loop control strategy for the prevention and suppression of focal epileptic seizures and motivate its subsequent testing in humans and animal models.

Disclosures: **G. Acharya:** None. **A. Huang:** None. **V. Santhakumar:** None. **E. Nozari:** None.**Poster****PSTR311: Network Activity Modulation****Location:** SDCC Halls B-H**Time:** Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.12/YY2

Topic: J.08. Methods to Modulate Neural Activity

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National Natural Science Foundation of China U20A20221
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Title: Brain-wide Mesoscale Functional Connectivity of Primate V2 Stripes Mapped by Infrared Neural Stimulation and 7T fMRI

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Abstract: Motivation: The submillimeter functional stripes in V2 are well-established as distinct gateways to dorsal and ventral visual pathways. Previous work using focal electrical stimulation and optical imaging (Hu & Roe, 2022) demonstrated local columnar connectivity, revealing highly functionally specific connectivity (stripe-matched and functionality matched), topographic network organization, and sparsity. However, the brain-wide mesoscale functional networks of V2 stripes remain poorly characterized due to methodological limitations. To address this gap, we mapped at whole brain scale, activations in response to single column stimulation using ultrahigh-field 7T fMRI with infrared neural stimulation (INS, 1875nm). **Methods:** In anesthetized macaques, thin and thick stripes were localized using functional mapping (0.8 mm isotropic) in response to color vs luminance (red/blue vs dark/light at 2cyc/deg, 1.5cyc/sec) and fast vs slow motion (0.5cyc/deg, 8cyc/sec vs 1cyc/sec, moving in 0°, 90°, 180°, 270° directions), respectively. Using 200µm optical fibers, we targeted a series of stripes in V2 from foveal to peripheral cortical locations while imaging in 7T MRI. Significant activations were identified via GLM (HRF regression, $p < 0.001$, FDR corrected). **Results:** INS evoked retinotopically matched activation in contralateral V2. Stimulation of thick stripe locations elicited discrete mesoscale nodes of activation along the dorsal pathway (V3, MT, MST, LIP). In contrast, thin-stripe stimulation selectively engaged ventral stream area V4, TEO, and temporal stream loci. Notably, INS-fMRI revealed mono- and di-synaptic functional networks, including distal parietal and prefrontal regions not typically observed with conventional tract tracing. Convergence of activation in parietal and prefrontal areas is being evaluated. **Conclusion:** Stripe-targeted INS-fMRI successfully mapped brain-wide functional connectivity of primate V2 at mesoscale resolution, confirming dorsal/ventral segregation of mesoscale loci while revealing broader mesoscale integrative networks beyond direct anatomical connected sites. This provides a mesoscale basis for both functional specificity and integration in brain-wide networks.

Disclosures: F. Tian: None. M. Chen: None. Y. Liu: None. A.W. Roe: None.

Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.13/YY3

Topic: F.02. Cerebellum

Support: JPMJMS2012

Title: Strength-duration profiles of endovascular cerebellar neurostimulation

Authors: *W. Qi¹, J. VILLALOBOS¹, J. LIU², H. XIN¹, M. SPENCER¹, S. KAZEMI³, S. HO⁴, J. WEST¹, D. B. GRAYDEN⁵, S. JOHN²;

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Abstract: Electrical stimulation and neural recording are foundational to next generation neural prostheses and therapeutic interventions for treatment-resistant neurological disorders. Traditionally, such methods have relied on invasive surgical approaches, limiting their clinical reach. Recently, the cerebral vasculature has emerged as a promising, minimally invasive route for intracranial neuromodulation. This study presents the first detailed strength-duration profiling of cortical potentials evoked by endovascular stimulation using a novel polymer-based stent-electrode array. The device comprises five platinum disc electrodes affixed to a polyimide stent strut, deployed into the left transverse sinus adjacent to the cerebellum in an ovine model. Biphasic pulses (1-3 mA, 100-800 μ s, delivered 30 times at 2.11 s intervals) were applied via the stent electrodes. Subdural ECoG recordings from a 32-channel cortical grid captured robust, time-locked evoked potentials, with consistent peaks near 100 ms and 300 ms post-stimulation. Latencies were comparable to transcranial magnetic and direct current stimulation studies. Electrode viability was confirmed through impedance monitoring. Strength-duration analysis revealed effective and efficient neural activation, aligning well with established physiological parameters for neural activation. These findings affirm the safety and efficacy of endovascular stimulation for precisely activating brain circuits. This work establishes a framework for future minimally invasive neuromodulation technologies. By combining safe vascular access with high-fidelity neural activation, endovascular stimulation holds translational promise for closed-loop applications including seizure forecasting, brain stimulation for depression, and epilepsy management.

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Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.14/YY4

Topic: J.08. Methods to Modulate Neural Activity

Support: NSERC Grant RGPIN/6506-2018

Title: Mesoscopic Calcium Imaging of Visual Cortex Demonstrates an Effective Transient Chemogenetic Retinal Ganglion Cell Silencing in Mice

Authors: *G. LALIBERTE, D. BOIRE;
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Abstract: Focal disruptions in retinal output are observed in several pathologies that induce retinal ganglion cell (RGCs) degeneration. They lead to spatially confined deficits in cortical processing. However, few experimental models allow for both spatially selective and temporally reversible manipulations of retinal ganglion cell activity. Here, we present a chemogenetic strategy to transiently silence RGCs in specific retinal quadrants, effectively mimicking localized vision loss in a controlled and reversible manner.

To target RGCs, we performed intravitreal injections of an adeno-associated viral vector (AAV2.2-NEFL-HA-hM4D(Gi)) into the posterior chamber of adult C57Bl/6J mice (n = 7; age P60-P90). This led to a selective expression of the inhibitory DREADD receptor hM4Di under the RGC selective promotor NEFL. Systemic administration of deschloroclozapine (DCZ, 0.1 mg/kg, i.p. Hellobio) induced rapid, robust, and reversible suppression of RGC activity within spatially confined retinal regions.

To assess alterations in visual activation of cortex, we used wide-field calcium imaging in awake, head-fixed Thy1-GCaMP6s mice. Localized visual stimulation with white Gabor patches revealed a significant reduction in evoked activity within the retinotopically corresponding regions in the primary visual cortex (V1), with an average decrease of $51.8 \pm 16.0\%$ ($p = 0.009$, Bonferroni-corrected), ranging from 23.1% to 77.3%. In contrast, stimulation of non-silenced retinal areas produced no significant cortical changes. Additionally, contrast response functions to full-field sinusoidal gratings (0.04 cpd, 2 Hz; contrast 12.5%-100%) were significantly attenuated in the affected V1 region ($p = 0.0181$, two-way ANOVA), with a uniform reduction across contrasts but no interaction.

Postmortem immunohistochemistry of flat-mounted retinas confirmed specific expression of hM4Di in RGCs, validating anatomical targeting. Cortical suppression was reproducible and reversible across sessions, confirming model robustness.

This chemogenetic approach provides a non-invasive, reversible, and spatially precise model of focal RGC lesions. It replicates key features of a central scotoma and serves as a valuable platform to investigate cortical plasticity, sensory network adaptation, and to evaluate targeted vision restoration strategies.

Disclosures: G. Laliberte: None. D. Boire: None.

Poster

PSTR311: Network Activity Modulation

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Program #/Poster #: PSTR311.15/YY5

Topic: J.08. Methods to Modulate Neural Activity

Support: KAKENHI JP23H05450
KAKENHI JP22KK0180

Title: Combining implantable CMOS imaging and scaffold technologies for multimodal neural and cellular observation

Authors: *Y. SUNAGA¹, K. TSO¹, V. G. CASTILLO¹, Y. OHTA¹, P.-C. CHEN², M.-F. LIN³, Y.-C. HUANG³, W. SU³, J. OHTA¹;

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Abstract: Imaging techniques, including those using GCaMP, have become powerful tools in neuroscience research. However, conventional imaging methods face limitations when applied to freely behaving animals. To overcome these challenges, we have developed a compact measurement system based on a CMOS imaging device that can be directly implanted into the mouse brain. This implantable device is small, lightweight, and enables minimally invasive monitoring of neural activity. Using this device, we have successfully observed neural activity in freely moving mice [1,2]. In this study, we aim to establish a new platform that enables the monitoring of cellular growth and regeneration in addition to visualizing neural activity. Toward this objective, we are developing an in vitro monitoring system capable of monitoring the cellular growth process by leveraging our implantable imaging device technology. Specifically, we have initiated the development of an experimental platform that combines the implantable imaging device with a polypeptide-based scaffold. This setup allows for continuous observation in a three-dimensional environment while recreating the natural relationships between structure and function through cell adhesion and cellular interactions. In this platform, we fabricated a thin scaffold for cell culture directly on the pixel area of the imaging device using electrospinning technique. The device was fixed onto a parylene-coated glass slide and covered with a 3D-printed cell culture well, which was also coated with parylene. This configuration enables direct cell culture on the device surface. As an initial experiment, we confirmed that cells can be successfully cultured on a scaffold placed on a parylene-coated dummy device, which served as a substitute for a functional imaging device. In the next phase, we plan to culture cells on a functional implantable imaging device equipped with a scaffold and to observe cell growth and regeneration under in vitro environment. The final objective is to enable real-time monitoring of cell growth and regeneration under in vivo environment. [1] Y. Ohta et al., Neuroelectronics, vol.2, no.1, pp.1-33, 2025 [2] Y. Sunaga et al., IEEE Access, vol.8, pp.68013-68020, 2020

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Poster

PSTR311: Network Activity Modulation

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Topic: J.08. Methods to Modulate Neural Activity

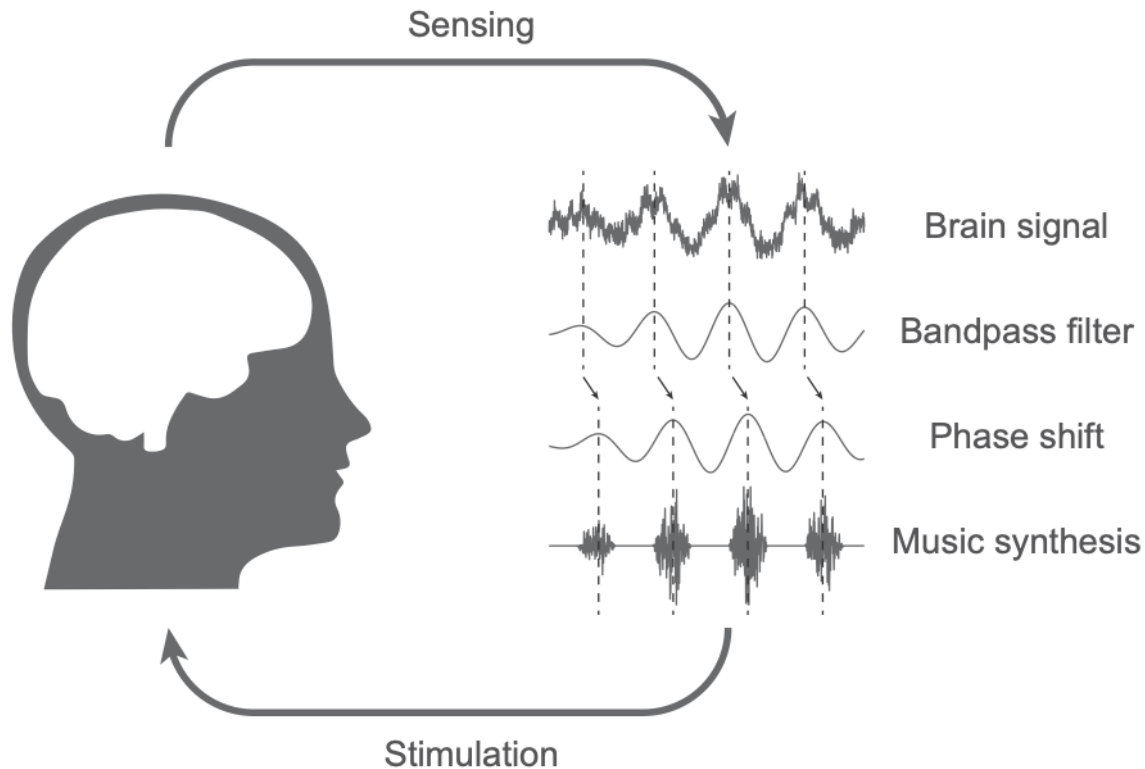
Support: EPSRC EP/R511584/1

Title: Brain-responsive music for non-invasive, targeted neurostimulation

Authors: *A. JACKSON¹, B. ZAAIMI², J. RICKERT^{3,4};

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Abstract: We are developing a new closed-loop brain stimulation method by embedding, within music, auditory elements that respond to the listener's brain activity. Here we show that this brain-responsive music has systematic and targeted effects on neural oscillations implicated in a variety of neurological and mental health disorders. We recorded magnetoencephalogram (MEG) or electroencephalogram (EEG) signals from participants as they listened to music synthesized by commercial audio software. Brain signals were bandpass filtered, phase-shifted and used to control the timbre and/or timing of notes within the music. Listening to brain-responsive music induced peaks and troughs in spectral power at frequencies that depended systematically on the phase-shift applied to the brain signal. Phase-dependent modulation was greatest at the centre frequency of the filter. As a result, by calibrating these parameters we could achieve selective enhancement or suppression of either theta (5 Hz) or alpha (10 Hz) oscillations. Moreover, by choosing different sensor locations we could target power modulation to either frontal or temporal cortex. The phase-dependent power modulation observed with brain-responsive music was significantly attenuated when participants listened to identical music as a conventional, open-loop stimulus. Finally, we demonstrate that brain activity could be modulated by more complex compositions combining a variety of brain-responsive musical elements controlled by a wireless, wearable EEG headband suitable for home use. Brain-responsive music provides an unobtrusive and targeted method of modulating neural oscillations in the listener's brain, and may enable both creative and therapeutic applications of neural interface technologies.



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Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

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Program #/Poster #: PSTR311.17/YY7

Topic: J.08. Methods to Modulate Neural Activity

Support: NIH Grant R01NS110893

Title: Volume control for a cortical network

Authors: ***R. WANG**¹, G. FOXWORTHY¹, G. FRIDMAN^{1,2};

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Abstract: Excitability fundamentally shapes cortical network responses, influencing brain functions such as attention, learning, and decision-making. Dysregulation of excitability underlies disorders including epilepsy and autism. Traditional pharmacological treatments are broad-acting and slow, making it difficult to control local cortical circuits in real-time. Ionic direct current (iDC) stimulation uses an electrolyte-filled microcatheter placed on the pia mater, enabling rapid and spatially precise modulation of cortical excitability. To investigate the ability of low-amplitude iDC to modulate network excitability, we recorded evoked multi-unit activity (MUA) in response to electrical stimulation on the foot across cortical layers in the primary somatosensory cortex hindlimb region (S1HL) of urethane-anesthetized adult rats (n=7; 4 males, 3 females). Recordings were performed using a 32-channel electrode array before and during iDC stimulation delivered at the cortical surface adjacent to the recording site. iDC stimulation produced amplitude-dependent and layer-specific bidirectional modulation of cortical activity. Anodic iDC (+50 μ A) significantly increased MUA in layer 5 channels by $66.8 \pm 7.3\%$, while cathodic (-50 μ A) significantly decreased it by $25.4 \pm 4.6\%$ (mean $\pm$ SEM; $p < 0.05$, t-test). iDC modulated the input-output gain of the cortical response, shifting the MUA patterns evoked by moderate foot stimulation to resemble those generated by stronger or weaker unmodulated inputs. Layer-specific analysis revealed the strongest modulation in layer 5, with progressively less effect in layers 4, 2/3, and 6. These findings were reproduced by computational modeling based on neuronal morphologies. Modeling suggested that the observed laminar specificity arises from differential dendritic integration at the axon initial segment, driven by cell-type-specific somatodendritic structures. In vivo spatial decay of iDC also closely matched model predictions, experimentally validating the predicted submillimeter spatial precision. iDC stimulation offers a new method to rapidly and reversibly modulate local cortical network excitability, facilitating causal investigations of how dynamic shifts in excitability influence brain function and behavior, and offering potential translational relevance for disorders characterized by altered cortical excitability.

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Poster

PSTR311: Network Activity Modulation

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Program #/Poster #: PSTR311.18/YY8

Topic: J.08. Methods to Modulate Neural Activity

Support: UH3NS121565

Title: Demonstration of neural plasticity effects of neuromodulation with beta phase-specific stimulation via the Cortec Brain Interchange

Authors: *H. CHO¹, B. L. GRANNAN⁵, A. L. KO², S. C. CRAMER⁶, J. G. OJEMANN³, J. A. HERRON⁴;

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Abstract: There continues to be growing interest in neural connectivity due to its role in understanding the underpinnings of neurological conditions and its potential to inform neuromodulation paradigms towards improving therapeutic outcomes. While there have been advances in the means to better investigate the nature of neural plasticity, the underlying mechanisms driving plasticity changes have yet to be fully characterized, particularly within clinical applications. Prior work utilized adaptive stimulation approaches within preclinical models to induce plasticity effects and cortico-cortical evoked potentials (CCEPs) to measure these changes. Modulation of particular oscillatory components of a CCEP has been used to assess changes in the strength of connections between cortical sites and can provide feedback to guide stimulation towards inducing targeted plasticity effects. In this work, we build off these previous efforts by assessing plasticity induction outcomes of a beta phase-specific stimulation paradigm delivered via an investigational research device, the CorTec Brain Interchange (CorTec GmbH, Freiburg, Germany). Patients with epilepsy undergoing intracranial monitoring with implanted stereo-encephalography electrodes were approached to assess the efficacy of this adaptive stimulation paradigm to modulate neuroplasticity. Stimulation pulses were delivered during specific phases of patients' beta oscillations, and CCEP responses were captured before and after stimulation. We then evaluated changes in CCEP morphology in response to adaptive stimulation delivery. Our findings show that with reliable administration of beta phase-triggered stimulation, we observed significant changes in CCEP components that would indicate clinical modulation of neural connectivity. These outcomes demonstrate the Brain Interchange's potential to induce plasticity with adaptive stimulation paradigms, and present preliminary data demonstrating the platform's abilities to be an effective research tool for neuroplasticity investigation.

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Poster

PSTR311: Network Activity Modulation

Location: SDCC Halls B-H

Time: Tuesday, November 18, 2025, 8:00 AM - 12:00 PM

Program #/Poster #: PSTR311.19/YY9

Topic: J.08. Methods to Modulate Neural Activity

Title: Standardized m-Torquer synthesis for consistent magnetogenetic modulation

Authors: *S. LEE¹, J. CHEON², M. KWAK³;

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Abstract: The m-Torquer is a well-established magnetic nanoparticle system, extensively characterized, widely adopted, and documented in numerous publications for precision magnetogenetic stimulation. To further enhance its applicability and reliability, this project focuses on optimizing synthesis processes, enforcing rigorous quality control standards, and ensuring improved colloidal stability under physiological conditions. We specifically addressed key parameters, including magnetic strength, nanoparticle assembly precision, and surface functionalization accuracy, to achieve robust performance and reproducibility. This optimized m-Torquer system has the capability for precise tunability of mechanical forces and effective cell-specific targeting through enhanced surface chemistry, enabling reliable outcomes in advanced in vitro and in vivo experiments.

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Poster

PSTR311: Network Activity Modulation

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Program #/Poster #: PSTR311.20/YY10

Topic: J.08. Methods to Modulate Neural Activity

Title: Radiolytic Neuromodulation of Cell Activity

Authors: *D. ZANELLA¹, S. J. MABRY^{5,6}, A. M. BUCHANAN², C. ROWE⁷, C. GONZÁLEZ-ARANCIBIA³, H. GOFFINETT⁴, M. I. HOSSAIN⁹, B. D. ROBINSON⁴, S. MENA ORTEGA¹⁰, A. MAROTTA¹¹, G. GALLI¹², S. A. ANDRABI⁴, P. HASHEMI¹³, A. CARTER⁸, M. BOLDING¹⁴, A. GALLI¹⁵;

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Col. London, London, United Kingdom; ¹¹; ¹²Brentwood, TN, ; ¹³Imperial College, London, London, United Kingdom; ¹⁴U of AL at Birmingham, Birmingham, AL; ¹⁵UAB, Brentwood, TN

Abstract: Biological techniques (e.g., optogenetics, chemogenetics) that enable the regulation of genetically defined neurons offer powerful opportunities to investigate how neural circuits control specific behaviors. Optogenetics allows for precise control of neuronal activity and neurotransmitter release in living tissue and freely moving animals. However, it has several limitations: it requires permanent intracranial implants, its effects are confined to the illuminated region of the brain, and the intensity of stimulation is difficult to regulate. To address these limitations, we developed a novel technology platform using X-ray (Xray) irradiation. Our data demonstrate that Xray stimulation increases intracellular Ca^{2+} , causes membrane potential depolarization/action potential firing, and promotes neurotransmitter release and associated behaviors in biological systems expressing the *Drosophila* transient receptor potential ankyrin 1 (dTRPA1) channel. dTRPA1 is an evolutionarily conserved detector of reactive oxygen species (ROS). The *dTRPA1* locus has two promoters (A and B) that drive expression of at least five transcripts. Among these, the dTRPA1(A)10a and dTRPA1(A)10b isoforms—derived from the A promoter—are highly sensitive to hydrogen peroxide (H_2O_2). Notably, the dTRPA1(A)10b splicing variant is not a direct heat sensor, making it preferable for applications in mammals. Importantly, Xray irradiation of physiological (aqueous) solutions rapidly generates H_2O_2 . Our findings show that the increases in intracellular Ca^{2+} , membrane depolarization/action potential firing, and neurotransmitter release induced by Xray in dTRPA1(A)10b-expressing preparations are mediated through Xray-induced H_2O_2 production and subsequent activation of dTRPA1 channels. Xray stimulation does not activate mouse TRPA1 (mTRPA1) or induce neurotransmitter release in murine brain preparations. Therefore, targeted viral expression of dTRPA1(A)10b in specific neurons in murine models may allow Xray to control their activity and associated behaviors. We provide a mechanistic description of how Xray activation of dTRPA1(A)10b leads to increases in intracellular Ca^{2+} , membrane depolarization, neurotransmitter release, and behavioral changes, using cells, neurons, and *Drosophila* as model systems. We define this approach as *radiolytic neuromodulation*. Finally, we began translating this technology to mammals by virally expressing dTRPA1(A)10b in vivo in defined mouse brain regions, enabling Xray to selectively activate these neuronal populations and drive neurotransmitter release.

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