Synapses and Epigenetics in the Alzheimer's Brain

Diego Mastroeni,[1] Nicole C. Berchtold, PhD,[2]
Carl W. Cotman, PhD,[2] and Paul D. Coleman, PhD[1]

[1]L.J. Roberts Center for Alzheimer's Research
Banner Sun Health Research Institute
Sun City, Arizona

[2]Institute for Brain Aging and Dementia
University of California, Irvine
Irvine, California

NOTES

Correlating Synapse Density with Cognitive Status

It has been generally accepted that synapses are the best correlate of cognitive status in Alzheimer's disease (AD). This is intuitively appealing because synapses provide the mechanisms by which information is transmitted from cell to cell, processed, and stored. The concept that synapses play a vital role in cognition was reinforced by two early quantitative studies of the relationship between synapse density and cognitive status, both of which yielded correlations of approximately +0.7 (DeKosky and Scheff, 1990; Terry et al., 1991). However, this result yields an R^2 of ~0.50, indicating that synapse density accounts for only 50% of the variability in cognitive status. This conclusion then leads to the question: Where is the missing 50%?

More recent study of the relationship between synapses and cognitive scores used unbiased stereological methods to quantify total synaptic numbers in lamina 3 of the inferior temporal gyrus (Scheff et al., 2011). The results showed an even lower correlation of +0.5 between cognitive score (according to the Mini-Mental State Examination [MMSE]) and synapse numbers, yielding an R^2 of 0.25. So in this case, synapse numbers accounted for only 25% of the variance in cognition scores. Where is the missing 50–75%?

There are several potential responses to the question:

(1) The earlier studies did not use methods of unbiased stereology, suggesting that researchers did not account for the potential effects of changes in size of synapses and volume of the brain region studied;
(2) It may be presumptuous to consider that synapse density in only one brain region could account for a behavior as complex as cognitive status; or
(3) The missing percentage can be found in synapses that are structurally present but functionally impaired.

More recent data have emphasized the complexity of relationships between synapses and cognition. For example, they have reported a correlation of 0.97 between delayed nonmatching to sample and size of the spine head in thin spines in prefrontal cortex area 46 of monkey (Morrison and Baxter, 2012).

Gene Expression in Alzheimer's Disease

A wide range of studies has provided data demonstrating impaired expression in AD of genes that play major roles in synaptic function. Studies of specific molecules in AD have, for example, shown reduced expression of dynamin 1 (Yao et al., 2003), which is critical in recycling synaptic vesicles, and losses in cholinergic receptor systems (Parri et al., 2011), a system that has a significant role in memory formation.

Beyond studies of specific molecules in AD, array studies have yielded an appreciation of the wide range of synapse-related genes whose expression is affected in AD (Berchtold et al., 2008; Liang et al., 2007, 2008). These studies indicate that a wide variety of synaptic gene classes are affected in AD, including transmitter receptor systems, transmitter synthesizing enzymes, transport systems, synapse stabilizing genes, postsynaptic structural genes, and ion channels, to name a few. Data such as these show coordinated modulation of expression of a wide range of genes and raise the question—what mechanism is coordinating such a wide variety of changes?

Epigenetic Mechanisms in Alzheimer's Disease

Recent study findings

During early development of an organism, the specification of cells and tissues requires the expression of large numbers of genes, modulated by epigenetic mechanisms in a coordinated fashion to produce specified cell types and tissues (Allis et al., 2009; Olynik and Rastegar, 2012). The ability of epigenetic mechanisms to regulate chromatin structure and, consequently, the coordinated expression of large gene sets, has motivated a number of studies on the role of epigenetic mechanisms in AD.

One of the early demonstrations of a relationship between AD and an epigenetic mechanism came from the demonstration of reduced expression in AD of 10 epigenetic markers in layer II neurons of the entorhinal cortex (Mastroeni et al., 2010). This study also showed that, in the AD brain, neurons bearing neurofibrillary tangles (NFTs) showed greater decrements in DNA methylation than tangle-free neurons.

An opportunity to eliminate genetic contributions from findings of epigenetic differences in AD came with an opportunity to examine epigenetic differences between a pair of identical twins discordant for AD (Mastroeni et al., 2009). The male twins of this study were educated together as chemical engineers and died within three years of each other. The twin with a clinical diagnosis of AD showed profuse NFTs and plaques, whereas the other twin had only extremely sparse NFTs and plaques. Compared with the nondemented twin, the levels of

NOTES

global DNA methylation were significantly reduced in the temporal neocortex of the AD twin, who had spent much of his career working with pesticides. This, as well as other epigenetic studies of identical twins (Fraga et al., 2005), points to the pertinence of environmental factors affecting the epigenome and, consequently, the phenotype.

Evidence from transcript expression

Although the above studies are consistent with a role for epigenetic mechanisms in altering the structure and function of synapses in AD, they are far from proving such a relationship. More detailed information about relationships between epigenetic variables and the expression of synaptic genes come from determining the correlations between expression of synaptic transcripts and the expression of selected transcripts known to regulate DNA methylation and histone acetylations. Selected aspects of these data, taken from analysis of an array study of four brain regions in brains covering the age range 20–99 years old, have already been published (Berchtold et al., 2008).

Figure 1 presents new analysis of data from Berchtold et al., 2008. It graphically represents correlations between a selection of four transcripts (Fig. 1*A–D*) related to synaptic structure and function (dynamin 1, PSD95, $AMPA_{A1}$, and synaptophysin) and ten selected transcripts that play roles in the methylation and acetylation actions of epigenetic mechanisms. These correlations are shown for four conditions: (1) AD in the postcentral gyrus (pcg, a region relatively unaffected in AD); (2) AD in the severely affected hippocampus (hipp); (3) the postcentral gyrus in nondemented, age-matched control cases; and (4) the hippocampus in nondemented, age-matched control cases.

These data show that the relationship between epigenetics and expression of synaptic genes depends on both the brain region examined and the disease state. In AD, quantification of DNA methyltransferase 1 (DNMT1) expression in hippocampus and postcentral gyrus shows a negative or low correlation with expression of all synaptic genes examined. On the other hand, the correlations are positive or low in both brain regions in age-matched controls. DNMT3a consistently yields negative correlations with all four synaptic genes, with the exception of age-matched control hippocampus, for which the correlations were either positive or low (Fig. 1*A–D*). The differences between the correlation patterns of these two DNMTs may relate to their presumptive differential functions in both *de novo* and maintenance DNA methylation.

The data for histone deacetylases (HDACs) 1, 2, 6, and 9 show varying patterns of correlation between expression of synaptic and epigenetic transcripts, depending on brain region and disease state. However, the histone acetyltransferases (HATs) MYST3 and MYST4 are fairly consistent in being negatively related to expression of the synaptic genes shown. In other words, reduced expression of these HATs is associated with increased expression of the synaptic genes shown (Fig. 1*A–D*). This finding is inconsistent with the common association of acetylation with a more open chromatin structure. It also serves to remind us of the potential complexity

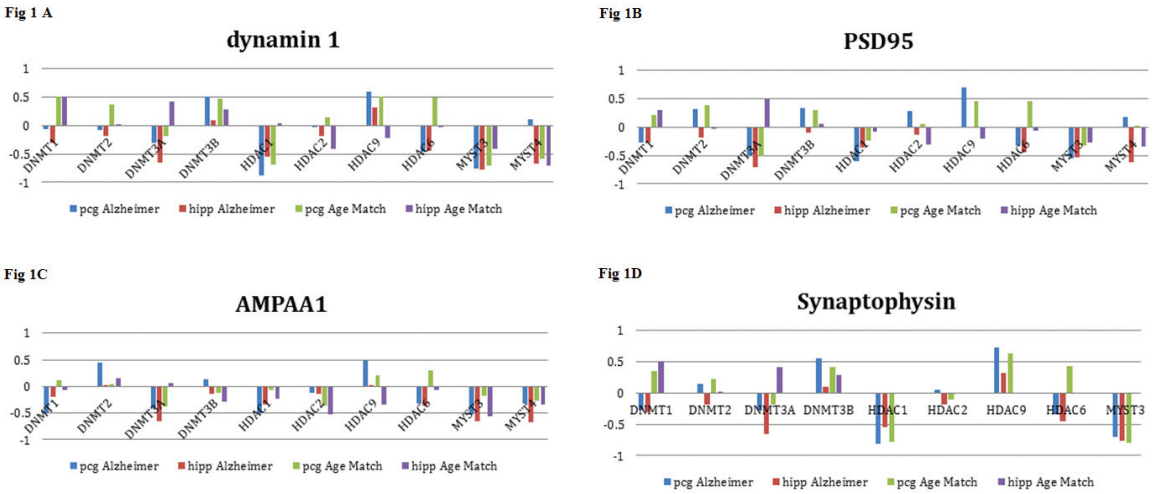

Figure 1. ***A–D***, Four plots that represent correlations (vertical axis) between the expression of 10 epigenetic molecules and 4 transcripts related to synaptic structure and function. Four data sets are represented by 4 different colors as shown: Data from AD postcentral gyrus, from AD hippocampus, from age-matched postcentral gyrus, and from age-matched hippocampus. $AMPA_{A1}$, ligand-gated ion channel, a subclass of glutamate receptor; DNMTs, a family of DNA methyltransferases; dynamin1, functions in the recycling of vesicles, especially at the synapse; HDACs, a family of histone deacetylases; MYSTs, a family of histone acetyltransferases; PSD95, postsynaptic density 95; synaptophysin, major synaptic vesicle protein.

NOTES

of interactions among histone modifications, DNA methylations, and other molecules affecting transcription at specific sites in the genome.

Another caution against generalizing these data is that the initial data came from homogenates of selected brain regions. Thus, these data represent not only neurons but also glia and vascular cells. Also, although more neuron-specific data have been derived from laser capture microdissection of single neurons (Liang et al., 2008), the amplification required for single-cell data may have differentially affected the selected genes of interest.

Implications

The correlations shown here do not, of course, prove causality. They do, however, offer suggestions for further studies, experimental manipulation, and potential therapeutic intervention for AD. At the same time, they providing a cautionary tale about the potential for complex interactions among the many epigenetic molecules, specific sites of synapse-related genes, brain regions, and disease states.

References

Allis CD, Jenuwein T, Reinberg D. (2009) Epigenetics. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press.

Berchtold NC, Cribbs DH, Coleman PD, Rogers J, Head E, Kim R, Beach T, Miller C, Troncoso J, Trojanowski JQ, Zielke HR, Cotman CW (2008) Gene expression changes in the course of normal brain aging are sexually dimorphic. Proc Natl Acad Sci USA 105:15605–15610.

DeKosky ST, Scheff SW (1990) Synapse loss in frontal cortex biopsies in Alzheimer's disease: Correlation with cognitive severity. Ann Neurol 27:457–464.

Fraga MF, Ballestar E, Paz MF, Ropero S, Setien F, Ballestar ML, Heine-Suñer D, Cigudosa JC, Urioste M, Benitez J, Boix-Chornet M, Sanchez-Aguilera A, Ling C, Carlsson E, Poulsen P, Vaag A, Stephan Z, Spector TD, Wu YZ, Plass C, Esteller M (2005) Epigenetic differences arise during the lifetime of monozygotic twins. Proc Natl Acad Sci USA 102:10604–10609.

Liang WS, Dunckley T, Beach TG, Grover A, Mastroeni D, Walker DG, Caselli RJ, Kukull WA, McKeel D, Morris JC, Hulette C, Schmechel D, Alexander GE, Reiman EM, Rogers J, Stephan DA. (2007) Gene expression profiles in anatomically and functionally distinct regions of the normal aged human brain. Physiol Genomics 28:311–322.

Liang WS, Dunckley T, Beach TG, Grover A, Mastroeni D, Ramsey K, Caselli RJ, Kukull WA, McKeel D, Morris JC, Hulette CM, Schmechel D, Reiman EM, Rogers J, Stephan DA (2008) Altered neuronal gene expression in brain regions differentially affected by Alzheimer's disease: A reference data set. Physiol Genomics 33:240–256.

Mastroeni D, McKee A, Grover A, Rogers J, Coleman PD. (2009) Epigenetic differences in cortical neurons from a pair of monozygotic twins discordant for Alzheimer's disease. PLoS One 4(8):e6617.

Mastroeni D, Grover A, Delvaux E, Whiteside C, Coleman PD, Rogers J. (2010) Epigenetic changes in Alzheimer's disease: Decrements in DNA methylation. Neurobiol Aging 31:2025–2037.

Morrison JH, Baxter MG (2012) The aging cortical synapse: Hallmarks and implications for cognitive decline. Nat Rev Neurosci 13:240–250.

Olynik BM, Rastegar M. (2012) The genetic and epigenetic journey of embryonic stem cells into mature neural cells. Front Genet 3:81.

Parri HR, Hernandez CM, Dineley KT. (2011) Research update: Alpha7 nicotinic acetylcholine receptor mechanisms in Alzheimer's disease. Biochem Pharmacol 82:931–942.

Scheff SW, Price DA, Schmitt FA, Scheff MA, Mufson EJ. (2011) Synaptic loss in the inferior temporal gyrus in mild cognitive impairment and Alzheimer's disease. J Alzheimers Dis 24:547–557.

Terry RD, Masliah E, Salmon DP, Butters N, DeTeresa R, Hill R, Hansen LA, Katzman R. (1991) Physical basis of cognitive alterations in Alzheimer's disease: synapse loss is the major correlate of cognitive impairment. Ann Neurol 30:572–580.

Yao PJ, Zhu M, Pyun EI, Brooks AI, Therianos S, Meyers VE, Coleman PD. (2003) Defects in expression of genes related to synaptic vesicle trafficking in frontal cortex of Alzheimer's disease. Neurobiol Dis 12:97–109.