2010

Differential effects of ApoE isoforms on dendritic spines in vivo: Linking an Alzheimer's disease risk factor with synaptic alterations

Jacob M. Basak
Washington University School of Medicine in St. Louis

Jungsu Kim
Washington University School of Medicine in St. Louis

Journal Club

Editor's Note: These short, critical reviews of recent papers in the *Journal*, written exclusively by graduate students or postdoctoral fellows, are intended to summarize the important findings of the paper and provide additional insight and commentary. For more information on the format and purpose of the Journal Club, please see http://www.jneurosci.org/misc/ifa_features.shtml.

Differential Effects of ApoE Isoforms on Dendritic Spines *In Vivo*: Linking an Alzheimer's Disease Risk Factor with Synaptic Alterations

Jacob M. Basak[1,2] **and Jungsu Kim**[2]

[1]Program in Molecular Cell Biology and [2]Department of Neurology, Washington University School of Medicine, St. Louis, Missouri 63110

Review of Dumanis et al.

The ε4 allele of apolipoprotein E (APOE) is the strongest genetic risk factor for the development of Alzheimer's disease (AD). Accumulation of amyloid-β (Aβ) is hypothesized to initiate synaptic and neuronal dysfunction that ultimately lead to neuronal cell death in AD, and several lines of evidence strongly suggest that the differential effects of apoE isoforms on Aβ aggregation and/or clearance plays a major role in AD pathogenesis (Kim et al., 2009). However, the apoE isoforms could influence the risk for AD via other mechanisms as well. For example, different apoE isoforms might produce differences in synaptic structure and function that make neuronal cells more susceptible to the toxic insults that occur with AD.

In a recent article published in *The Journal of Neuroscience*, Dumanis et al. (2009) characterized the effect of the apoE isoforms on dendritic spine density and branching in the murine brain. Dendritic spines, small protrusions along the length of the dendrite, play an important role in synapse function as the site of excitatory glutamatergic synaptic input. Changes in spine shape and number are postulated to contribute to the synaptic plasticity underlying learning and memory. The effect of apoE isoforms on dendrite structure and function has been extensively investigated *in vitro* with widely differing results depending on a variety of factors (Kim et al., 2009). The study by Dumanis et al. (2009) is the first to systematically compare dendritic spines *in vivo* in mice in which the human APOE2, APOE3, or APOE4 gene has been substituted into the mouse APOE locus. Using the Golgi impregnation method, the authors analyzed spine density and morphology in brain sections from various regions obtained from mice at multiple ages. Several important observations from this study shed light on how the apoE isoforms differentially alter synaptic complexity.

One of the principal findings was that, at all ages examined, mice expressing APOE4 had significantly fewer dendritic spines in layer II/III cortical pyramidal neurons than mice expressing APOE2 or APOE3 [Dumanis et al. (2009), their Fig. 1, Fig. 3]. No differences in cortical spine density between APOE2 and APOE3 mice were observed [Dumanis et al. (2009), their Fig. 1, Fig. 3]. Based on these results, Dumanis et al. (2009) proposed that apoE isoforms have differential effects on spine formation in the cortex. However, spine density reflects the balance between spine formation and elimination (Alvarez and Sabatini, 2007). The technique used by Dumanis et al. (2009) only allowed quantification of spine number at a single time point. Therefore, the decreased number of cortical spines in APOE4 mice could be caused by a decrease in spine formation and/or an increase in elimination. Because the addition of a new stable spine is a rare event in adult mice (Alvarez and Sabatini, 2007), it is possible that the decreased spine density in older APOE4 mice results from increased spine elimination.

Understanding how apoE4 affects spine dynamics *in vivo* is critical, as enhanced elimination of weak synaptic connections may not necessarily be detrimental to neuronal function. A previous study using two-photon microscopy to measure spine turnover in the living mouse brain analyzed spine formation and elimination following a decrease in barrel cortex activity due to sensory deprivation (Zuo et al., 2005). The authors found that long-term sensory deprivation dramatically reduced spine elimination, but had no effect on spine formation. This finding suggests that synapse connectivity during experience-dependent processes could predominantly be shaped by the extent of spine loss. However, more recent two-photon studies demonstrated that both formation and elimination of spine are associated with learning and could play a role in memory formation (Xu et al., 2009; Yang et al., 2009). Long-term two-photon imaging of spines in the APOE targeted replacement mice could

Received Jan. 26, 2010; revised Feb. 26, 2010; accepted March 3, 2010.

We thank our mentor Dr. David Holtzman for support and discussion, and Alison Finger and Juhyun Kim for careful reading of the manuscript.

Correspondence should be addressed to Jungsu Kim, Department of Neurology, Washington University School of Medicine, 660 South Euclid Avenue, Campus Box 8111, St. Louis, MO 63110. E-mail: kimj@neuro.wustl.edu.

DOI:10.1523/JNEUROSCI.0505-10.2010

 0270-6474/10/304526-02$15.00/0

help elucidate how the presence of apoE4 regulates spine remodeling in comparison to the other apoE isoforms.

Dumanis et al. (2009) also found that spine length differed between mice expressing different APOE isoforms. At 1 month of age, APOE2 mice had longer cortical spines than APOE3 and APOE4 mice [Dumanis et al. (2009), their Fig. 1*F–H*]. In the 1-year-old cohort, the mice expressing the APOE4 gene had shorter spines than APOE2 and APOE3 mice [Dumanis et al. (2009), their Fig. 3*H*]. Whether these structural differences reflect functional changes in synaptic activity was not analyzed. However, one can postulate that the different spine lengths may reflect isoform-dependent alterations in the populations of spine subtypes, which are believed to represent different levels of synapse integrity. Spines are generally categorized as thin, stubby, or mushroom type based on their shape. It has been proposed that mushroom spines represent more stable synaptic connections (memory spines), whereas thin spines are more dynamic and respond to changes in synaptic connections (learning spines) (Bourne and Harris, 2007). Young mice have many long, thin dendritic protrusions known as filopodia, which may be spine precursors (Alvarez and Sabatini, 2007). Insight into how the APOE isoforms influence the distribution of spines in these subgroups may lead to a better understanding of apoE's role in modulating synaptic connectivity. For instance, it may be possible that the increased spine length in 1-month-old APOE2 mice results from an increase in filopodia number and subsequent synapse formation. However, it is difficult to conclude that the structural differences observed between isoforms actually represent variations in synaptic strength and integrity, because the authors did not distinguish between spine subtypes in their analysis.

Whether the presence of apoE4 represents a gain of toxic function or loss of a neuroprotective function is still debated (Kim et al., 2009). The data presented by Dumanis et al. (2009) lends some support to the latter hypothesis, at least in regard to dendritic spine morphology. They compared dendritic spine number and length between neurons from mice lacking apoE altogether and mice in which a human APOE isoform replaced the mouse APOE gene, and demonstrated that the presence of any human apoE isoform led to increased spine number at 3 months of age in comparison to mice that lack apoE [Dumanis et al. (2009), their Fig. 3*A–C*]. However, APOE-null mice were not included in the 1-year-old cohort, so it is premature to conclude that the lower spine density in the APOE4 mice represents the loss of a neuroprotective function.

Several studies have shown that the amount of apoE protein in the brain differs among mice expressing different human isoforms, with the highest levels in APOE2 mice and the lowest in APOE4 mice (Kim et al., 2009). As Dumanis et al. (2009) point out, the different levels of apoE protein could account for the differences observed between isoforms in their study. However, a recent study by Korwek et al. (2009) did not find a difference in apoE protein levels in the hippocampus of mice expressing different human APOE isoforms. It is worth validating this observation, as it may explain why Dumanis et al. (2009) found no differences in spine density in the hippocampus of the mice expressing different isoforms [Dumanis et al. (2009), their Fig. 2, Fig. 3*I–L*]. Regardless, future studies will need to directly determine whether the effect of APOE isoforms on spine number and morphogenesis result from intrinsic functional differences in the apoE isoform proteins, or simply differences in the level of apoE protein. One method of analyzing the effect of apoE level would be to compare mice that have two alleles of APOE4 with those that have one allele of APOE4.

In summary, it appears that the APOE isoforms differentially modulate dendritic spine number and morphology *in vivo* in the otherwise normal mouse brain. It remains to be seen whether similar effects will be observed in the human brain in the presence of different APOE isoforms and whether this is relevant to AD pathogenesis. Recently, it was demonstrated that an increase in the levels of Aβ secreted from axons and dendrites decreases spine number significantly in local dendrites (Wei et al., 2010). As a result, it is possible that the differential effects of the APOE isoforms on dendritic spine may be altered or absent in the presence of high Aβ levels. Further studies using APOE-isoform mice that express human Aβ will be important in determining whether the different APOE isoforms exhibit an effect on dendrite number and structure during the onset and progression of AD pathogenesis.

References

Alvarez VA, Sabatini BL (2007) Anatomical and physiological plasticity of dendritic spines. Annu Rev Neurosci 30:79–97.

Bourne J, Harris KM (2007) Do thin spines learn to be mushroom spines that remember? Curr Opin Neurobiol 17:381–386.

Dumanis SB, Tesoriero JA, Babus LW, Nguyen MT, Trotter JH, Ladu MJ, Weeber EJ, Turner RS, Xu B, Rebeck GW, Hoe HS (2009) ApoE4 decreases spine density and dendritic complexity in cortical neurons *in vivo*. J Neurosci 29:15317–15322.

Kim J, Basak JM, Holtzman DM (2009) The role of apolipoprotein E in Alzheimer's disease. Neuron 63:287–303.

Korwek KM, Trotter JH, Ladu MJ, Sullivan PM, Weeber EJ (2009) ApoE isoform-dependent changes in hippocampal synaptic function. Mol Neurodegener 4:21.

Wei W, Nguyen LN, Kessels HW, Hagiwara H, Sisodia S, Malinow R (2010) Amyloid beta from axons and dendrites reduces local spine number and plasticity. Nat Neurosci 13:190–196.

Xu T, Yu X, Perlik AJ, Tobin WF, Zweig JA, Tennant K, Jones T, Zuo Y (2009) Rapid formation and selective stabilization of synapses for enduring motor memories. Nature 462:915–919.

Yang G, Pan F, Gan WB (2009) Stably maintained dendritic spines are associated with lifelong memories. Nature 462:920–924.

Zuo Y, Yang G, Kwon E, Gan WB (2005) Long-term sensory deprivation prevents dendritic spine loss in primary somatosensory cortex. Nature 436:261–265.