

## EARLY-LIFE SEIZURES AND COGNITIVE IMPAIRMENT: A SPINY PROBLEM?

**Recurrent Seizures and the Molecular Maturation of Hippocampal and Neocortical Glutamatergic Synapses.** Swann JW, Le JT, Lee CL. *Dev Neurosci* 2007;29(1-2):168-178. Recurrent seizures in animal models of early-onset epilepsy have been shown to produce deficits in spatial learning and memory. While neuronal loss does not appear to underlie these effects, dendritic spine loss has been shown to occur. In experiments reported here, seizures induced either by tetanus toxin or flurothyl during the second postnatal week were found to reduce the expression of NMDA receptor subunits in both the hippocampus and neocortex. Most experiments focused on alterations in the expression of the NR2A subunit and its associated scaffolding protein, PSD95, since their expression is developmentally regulated. Results suggest that the depression in expression can be delayed by at least 5 days but persists for at least 3-4 weeks. These effects were dependent on the number of seizures experienced, and were not observed when seizures were induced in adult mice. Taken together, the results suggest that recurrent seizures in infancy may interrupt synapse maturation and produce persistent decreases in molecular markers for glutamatergic synapses - particularly components of the NMDA receptor complex implicated in learning and memory.

### COMMENTARY

Abundant data from experimental seizure models and clinical studies demonstrate that recurrent seizures early in life are associated with long-term cognitive and behavioral problems (1,2,3). Whether these chronic cognitive deficits are due to the etiology of the seizure or to the seizures themselves has been a controversial topic. If the mechanism of seizure-induced cognitive impairment could be identified, perhaps treatment strategies could be devised to circumvent those deficits.

Among seizure-related neuronal alterations, certain changes are known *not* to explain cognitive deficits, including cell death, axonal reorganization, and neurogenesis. While each of those cellular alterations can occur after early-life seizures, cognitive deficits are seen even when none of those cellular structural alterations are documented. Since learning and memory are hippocampal based and are dependent upon glutamatergic synaptic plasticity, it has been hypothesized that glutamatergic synaptic function (especially NMDA receptors) is altered by seizures and causes subsequent cognitive deficits. Thus, investigations focusing on these synapses and the spine-like structures that house them are extremely relevant.

The experiments described here address the hypothesis that early-life seizures cause cognitive impairment by altering the expression of NMDA receptors, particularly those located at synapses on dendritic spines. Swann and colleagues induced seizures in rodents in the second postnatal week using two methods with very different mechanisms of action that both impair GABAergic (inhibitory) synapses: intrahippocampal injection of tetanus toxin (TNTX) or inhalation of the volatile convulsant flurothyl. TNTX causes several brief limbic seizures (by block-

ing presynaptic GABA release) over a discrete time period, lasting about a week after injection. Therefore, this method assesses the effect of repeated but time-limited seizures; here, TNTX was delivered to postnatal (P) day 10 rats and spontaneous seizure frequency was monitored for the next week. Flurothyl induces seizures only during exposure to the inhalant compound (by blocking postsynaptic GABA action). This method allows the experimenter to determine the exact number and duration of seizures; in these experiments, 15 flurothyl seizures were induced in mice between P9-P13. The authors subsequently used Western blotting to examine the expression of NMDA receptor subunit subtypes NR1, NR2A, and NR2B as well as the dendritic spine-associated scaffold protein, PSD95, which has a developmental expression similar to NR2A.

The authors found that both TNTX and flurothyl seizures were associated with down-regulation of NR1, NR2A, and NR2B as well as of PSD95. Decreases in NR2A were most prominent and persistent, which is of significance given the central importance of this receptor subtype in learning and memory and its unique developmental profile. These receptor changes were sustained in both hippocampus and neocortex, but only when seizures were induced during the neonatal period. Seizures induced in adult rodents by the same methods did not alter the expression of any of those proteins. Furthermore, in the flurothyl model, the changes were only found when at least three daily seizures (15 total), but not when a single daily seizure (5 total) occurred across the five treatment days.

It was concluded that seizures early in life impair subsequent learning and cognition by disrupting the developmental trajectory of glutamate synapses. One possible mechanism for these effects on synaptic function is that network hyperexcitability engendered by the seizures interferes with normal glutamatergic synapse development (4). The finding of altered scaffold protein PSD95 points toward dendritic spines as a potential localization for the defect. Since PSD95 is the most abundant excitatory synaptic protein, down-regulation implies

either synaptic loss (5,6), altered synaptic function, or both. PSD95 directly interacts with NR2A and indirectly with other elements of the subsynaptic machinery involved in plasticity.

These results provide additional data that seizures early in life are associated with long-term cognitive impairment. Cognition was not tested directly in these experiments, though the authors' previous work established cognitive deficits in neonatal TNTX-exposed rats (7). Interestingly, down-regulation of NR2A and alteration of PSD95 expression also was documented in recent experiments in which a single neonatal seizure in rats was induced by subcutaneous kainate on P7, but in the opposite direction—PSD95 expression was increased in those experiments with no loss of dendritic spines (8). While these apparently discrepant findings confirm the inherent complexity of the experimental problems of seizure models, they now bring to the forefront the role of subsynaptic scaffolds (9), which provides a novel and unexpected target for potential pharmacologic intervention in seizure-induced cognitive changes.

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