Adult Neurogenesis: Opening the Gates of Troy From the Inside

Aberrant Hippocampal Neurogenesis Contributes to Epilepsy and Associated Cognitive Decline.

Cho KO, Lybrand ZR, Ito N, Brulet R, Tafacory F, Zhang L, Good L, Ure K, Kernie SG, Birnbaum SG, Scharfman HE, Eisch AJ, Hsieh J. *Nat Commun* 2015;26:6606.

Acute seizures after a severe brain insult can often lead to epilepsy and cognitive impairment. Aberrant hippocampal neurogenesis follows the insult but the role of adult-generated neurons in the development of chronic seizures or associated cognitive deficits remains to be determined. Here we show that the ablation of adult neurogenesis before pilocarpine-induced acute seizures in mice leads to a reduction in chronic seizure frequency. We also show that ablation of neurogenesis normalizes epilepsy-associated cognitive deficits. Remarkably, the effect of ablating adult neurogenesis before acute seizures is long lasting as it suppresses chronic seizure frequency for nearly 1 year. These findings establish a key role of neurogenesis in chronic seizure development and associated memory impairment and suggest that targeting aberrant hippocampal neurogenesis may reduce recurrent seizures and restore cognitive function following a pro-epileptic brain insult.

Commentary

The demonstration that new neurons are produced throughout life in mammals—including humans—has generated considerable excitement over the past couple of decades. Evidence of adult-neurogenesis has offered hope that neuronal loss due to injury or neurological disease might one day be repairable. Increased neurogenesis is sought after in most disease fields; however, the epilepsy field stands alone in offering an important note of caution: While neurogenesis in the dentate gyrus of the hippocampus is dramatically increased following a range of epileptogenic injuries, many of these new cells appear to integrate pathologically into the brain, developing aberrant axonal and dendrite projections that underlie the formation of recurrent excitatory connectivity. These observations have led to the hypothesis that aberrant integration of adult-generated neurons may mediate temporal lobe epileptogenesis (1).

Numerous lines of evidence have been developed to support this hypothesis. First, the hippocampal dentate gyrus is believed to act as a "gatekeeper," regulating the flow of excitation through the hippocampal circuit (2). Disruption of this structure, therefore, might be particularly pro-excitatory. Second, the appearance of abnormal granule cells in animal models of temporal lobe epilepsy roughly corresponds to the onset of spontaneous seizures, and abnormal cells are evident in resected human tissue from epilepsy surgeries (3). Finally, genetically manipulating newborn granule cells to induce them to integrate abnormally produces a seizure phenotype, establishing that abnormal cells are capable of causing the disease (4). Abnormal granule cells, therefore, have the ability to open the dentate gate to allow seizures to rampage through the brain. Demonstrating that abnormal granule cells *actually* contribute to seizure incidence in epilepsy, however, requires additional evidence.

To obtain this evidence, Cho and colleagues utilized a transgenic approach to eliminate neurogenesis prior to treating the animals with pilocarpine to induce epileptogenesis. Their hypothesis: If newborn granule cells contribute to epileptogenesis, then blocking neurogenesis should mitigate disease development. The approach selected by the group utilized nestin-δ-HSV-thymidine kinase-EGFP mice. In these animals, the nestin promoter drives thymidine kinase and EGFP expression in neural progenitor cells. This alone does not harm the cells, but treatment of the animals with ganciclovir induces apoptosis among the thymidine–kinase-expressing progenitors. To block neurogenesis, adult animals were treated with ganciclovir for 4 weeks, and then status epilepticus was induced with pilocarpine. Five weeks later, ganciclovir and vehicle-treated animals were video-EEG monitored 24/7 for 2 weeks. Blocking neurogenesis reduced spontaneous seizure frequency by 40%, supporting the hypothesis that these neurons contribute to temporal lobe epileptogenesis. Moreover, seizure frequencies were still reduced in animals examined 1 year after treatment, demonstrating a long-term beneficial effect. Finally, treated animals also exhibited improvement in hippocampal-dependent memory. The presence of abnormal granule cells profoundly alters the hippocampal circuit, so

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reducing the number of these cells might prevent them from disrupting hippocampal performance. Alternatively, improved memory might reflect secondary positive effects of reducing seizures, as seizures themselves can impair cognition. In either case, the result is encouraging.

In a second key experiment, animals were treated with ganciclovir for 4 weeks before pilocarpine-status epilepticus, and then received an additional 4 weeks of treatment after status epilepticus. This approach was undertaken to block neurogenesis occurring both before and after the insult because neurons generated during both periods integrate abnormally (5). With near-complete ablation of adult-generated cells, this treatment was predicted to result in a greater reduction in seizure frequency. Surprisingly however, EEG monitoring between 5 to 7 weeks did not reveal a significant reduction in seizure frequency with this protocol, indicating that pre-status ablation of neurogenesis was more effective than combined pre- and post-ablation.

To begin to understand the significance of this second finding, the authors explored one technical limitation of their approach; specifically, the nestin driver they used was activated in reactive astrocytes following status epilepticus, and cell birthdating studies with BrdU revealed that these new cells were killed by the treatment. The function of reactive astrocytes in epileptogenesis is uncertain, but if the new cells play a protective role—perhaps by improving ion homeostasis—their loss might mitigate the beneficial effects of ablating abnormal granule cells. An alternate and more intriguing possibility is that granule cells born at different times relative to status epilepticus perform different functions. There is already evidence in the literature that some new granule cells may act to reduce excitation in the epileptic brain (6, 7) and promote inhibitory circuit function in the normal brain (8, 9). If more of these cells are generated after status, their loss might dampen the effect of ablating aberrant granule cells. Other possibilities include the induction of compensatory changes. Ablation of granule cells impairs long-term potentiation (LTP) in the dentate but also leads to the induction of neurogenesis-independent LTP about 6 weeks later (10). Whether and how the second period of granule cell ablation interacts with compensatory changes induced by the first is likely to be complex and might account for the paradoxical results. Any inflammatory changes induced by the second period of ablation would also overlap with inflammation induced by status epilepticus, which could exacerbate epileptogenesis.

Despite the many unanswered questions, the study by Cho and colleagues provides important new proof-of-concept data that manipulating granule cell neurogenesis could be a fruitful avenue for the development of disease-modifying therapies. There is intense interest in developing therapies to prevent the development of epilepsy. Unlike many other human diseases, there are currently no FDA-approved treatments that can reduce the risk of developing epilepsy or exert disease-modifying effects. At the basic science level, further studies will be needed to delineate the temporal dynamics of ablation therapy, as well as studies to assess the impact of inflammatory and compensatory changes. At the clinical level, strategies will be needed to efficiently target adult-generated neurons in the human brain during the critical time window after an epileptogenic brain injury. Further refinement of the technique to target subtypes of adult-generated granule cells may eventually prove successful, keeping the dentate gate safely closed.

by Steven C. Danzer, PhD

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