

IS POSTTRAUMATIC EPILEPSY THE BEST MODEL OF POSTTRAUMATIC EPILEPSY?

A Model of Posttraumatic Epilepsy Induced by Lateral Fluid-Perussion Brain Injury in Rats

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Although traumatic brain injury is a major cause of symptomatic epilepsy, the mechanism by which it leads to recurrent seizures is unknown. An animal model of posttraumatic epilepsy that reliably reproduces the clinical sequelae of human traumatic brain injury is essential to identify the molecular and cellular substrates of posttraumatic epileptogenesis, and perform preclinical screening of new antiepileptogenic compounds. We studied the electrophysiologic, behavioral, and structural features of posttraumatic epilepsy induced by severe, non-penetrating lateral fluid-perussion brain injury in rats. Data from two independent experiments indicated that 43% to 50% of injured animals developed epilepsy, with a latency period between 7 weeks to 1 year. Mean seizure frequency was

0.3 ± 0.2 seizures per day and mean seizure duration was 113 ± 46 s. Behavioral seizure severity increased over time in the majority of animals. Secondarily generalized seizures comprised an average of $66 \pm 37\%$ of all seizures. Mossy fiber sprouting was increased in the ipsilateral hippocampus of animals with posttraumatic epilepsy compared with those subjected to traumatic brain injury without epilepsy. Stereologic cell counts indicated a loss of dentate hilar neurons ipsilaterally following traumatic brain injury. Our data suggest that posttraumatic epilepsy occurs with a frequency of 40% to 50% after severe non-penetrating fluid-perussion brain injury in rats, and that the lateral fluid percussion model can serve as a clinically relevant tool for pathophysiologic and preclinical studies.

COMMENTARY

The need to develop better animal models of epileptogenesis and chronic epilepsy has been widely acknowledged. However, what a better model might look like largely depends on what aspect of the epileptic condition is intended for modeling. Merely reproducing a presumed etiological factor, such as status epilepticus, cortical malformation, stroke, fever, or traumatic brain injury, may not be that challenging. However, as the ultimate goal of an animal model is to understand the pathophysiology of the disease and to develop effective therapeutic interventions, a better model of epileptogenesis would be one that comes closest to reproducing the mechanisms and pharmacological profile. The key question is whether an etiological approach necessarily implies mechanistic and therapeutic congruence between experimental and clinical scenarios.

The continuous interest in developing better models for posttraumatic epilepsy is understandable, as traumatic brain injury (TBI) represents one of the apparent factors that can be retrospectively linked to epilepsy. A number of experimental studies have found that TBI induces alterations in hippocam-

pal excitability and pathology that resemble those of other animal models and of human temporal lobe epilepsy (1–3). Furthermore, recent advances in the techniques of long-term EEG monitoring have helped to reveal epileptiform events in animals subjected to TBI (4).

Kharatishvili and colleagues performed a detailed investigation of the occurrence, progression, and histopathology of experimental posttraumatic epilepsy. The authors found that all animals that had been subjected to and survived severe TBI exhibited typical hallmarks of chronic epilepsy, such as neuronal injury and synaptic reorganization in the hippocampus; in addition, half of these animals developed spontaneous recurrent seizures. Correlation among the three mentioned features of chronic epilepsy allowed some important conclusions to be drawn. First, the extent of TBI-induced cell loss in the hilus of the dentate gyrus did not correlate with the occurrence of spontaneous seizures. Second, while all experimental animals demonstrated mossy fiber sprouting in the injured dentate gyrus, the extent of synaptic reorganization in rats with epilepsy was more pronounced than in posttraumatic animals that failed to develop spontaneous seizures. Moreover, in contrast to nonepileptic rats, mossy fiber sprouting in epileptic animals was bilateral. Hence, TBI-induced hippocampal injury and synaptic reorganization do not necessarily lead to epilepsy. Some other mechanisms, which are yet to be identified, seem to

trigger spontaneous seizures following trauma. Epileptogenesis, in turn, might lead to the progression of mossy fiber sprouting, further exacerbating epilepsy.

Chronic epilepsy that developed in a subset of the post-TBI animals had general characteristics that are similar to those found after other types of precipitating insults, such as status epilepticus (induced, e.g., by pilocarpine, kainic acid, or electrical stimulation). Status epilepticus is frequently used in the laboratory setting because of the simplicity of induction, relatively high survival rate, fast progression, high incidence, and frequency of spontaneous seizures, relative resistance of epileptogenesis to standard antiepileptic drug therapy, and pathological findings that resemble patients with temporal lobe epilepsy. The TBI model might have provided insight into the mechanisms of posttraumatic epileptogenesis through comparison of epileptic rats to those who did not develop spontaneous seizures, despite trauma and hippocampal cell loss; however, several methodological difficulties complicate such comparison. In particular, there is an apparent lack of uniformity in the precipitating insult itself, including significant variations in the duration of posttraumatic apnea; presence of acute posttraumatic seizures in some animals and absence of such seizures in others; some variability in the impact volume and direction; unpredictable location; and difference in the extent of extravasations.

In the case of the protocol used by Kharatishvili and colleagues, it is unclear whether mimicking the etiological factor led to distinct pathophysiological outcome or not. Coulter et al. (1) found that changes in hippocampal excitability after TBI (induced by a similar technique) closely resembled those occurring following pilocarpine-induced status epilepticus and that the differences between the two models were mostly quantitative (i.e., less severe after TBI than after status epilepticus). Histopathological findings of Kharatishvili et al. confirmed previously reported selective loss of neurons in the hilus of the dentate gyrus (3); however, as the authors showed, such histopathological sequela alone cannot account for chronic epilepsy. Furthermore, the clinical relevance of isolated hilar injury to human posttraumatic epilepsy is not definitive, as epilepsy patients with TBI in the anamnesis show various patterns of both hippocampal and extrahippocampal (particularly neocortical) abnormalities.

Do the benefits of the model outweigh methodological problems associated with the induction of posttraumatic epilepsy, as acknowledged by Kharatishvili and colleagues? This question is an important, practical one to consider when introducing a protocol into an epilepsy research laboratory. Currently, there is no compelling evidence that posttraumatic spontaneous seizures are mechanistically different from those induced, for example, by status epilepticus. At this point, it is not

known whether posttraumatic spontaneous seizures have different resistance to antiepileptic drugs, as compared with seizures that develop after status epilepticus. Furthermore, such factors as low survival rate, low percentage of animals that develop epilepsy, and a slow disease progression represent substantial challenges in applying the described protocol to the evaluation of the efficacy of therapeutic interventions, while the apparent variability of precipitating insult might significantly complicate validation of the model.

Although the studies are unquestionably useful, the protocol employed by Kharatishvili and colleagues as well as similar techniques used by others (4) have not yet become both valid models for studying mechanisms of posttraumatic epilepsy and practical tools for developing new therapeutic strategies. Given the technical difficulties and low reproducibility, additional studies are required to prove that this model indeed imitates features that are unique to posttraumatic epilepsy, as opposed to imitating epilepsy induced by other means but with fewer technical challenges. Alternately, focusing on certain sequelae of brain trauma, such as hemorrhage-associated iron deposition in brain tissue (5), might offer mechanism-oriented approaches to modeling posttraumatic epileptogenesis. All the mentioned problematic issues, however, do not diminish but rather emphasize the importance of the work of Kharatishvili and colleagues. After all, the question: "What good are animal models? (6)" can only be answered through trials and comparisons, and the authors have undertaken an important step in this direction.

by Andrey Mazarati, MD, PhD

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