Changes in mIPSCs and sIPSCs after kainate treatment: status epilepticus–induced neuronal loss or direct activation of kainate receptors?

To the Editor: We are excited to see that our study has attracted the interests of a colleague from a field close to but different from the neurophysiology of epilepsy. The comments by Rodriguez-Moreno propose an alternative interpretation for our observation, an interpretation that is surprising when one closely examines our data and the published data on kainate receptors. That is, Rodriguez-Moreno has proposed that the short- and long-term (i.e., days and months, respectively) reduction of miniature inhibitory postsynaptic current (mIPSC) frequency of the dentate granule cells after kainate-induced status epilepticus (Shao and Dudek 2005) is attributed to direct activation of presynaptic kainate receptors, rather than the loss of GABAergic interneurons caused by status epilepticus. We discuss this issue in more detail below.

First, the core of the hypothesis of Rodriguez-Moreno is based on the earlier observations that bath application of kainate reduced the amplitude of evoked IPSCs and both the frequency and amplitude of mIPSCs in cultures and hippocampal slices (Rodriguez-Moreno et al. 1997, 1998). Activation of metabotropic kainate receptors on the axonal terminals of the GABAergic neurons was proposed to cause reduction of \( \gamma \)-aminobutyric acid (GABA) release (Rodriguez-Moreno et al. 2000). However, several questions remain regarding this hypothesis. 1) Subsequent studies from different laboratories have reported that kainate has little or no effect on mIPSCs, but a dramatic increase in the frequency of spontaneous (s)IPSCs (Bureau et al. 1999; Cossart et al. 1998; Frerking et al. 1999; Semyanov and Kullmann 2001). These results contradict the hypothesis of Rodriguez-Moreno that kainate acts directly on presynaptic terminals, but does imply excitation of presynaptic interneurons at the somatodendritic level. 2) Kainate-induced reduction of evoked IPSC amplitude has several different possible explanations besides a reduction of GABA release (Frerking et al. 1999; Kang et al. 2004). 3) No compelling anatomical evidence supports the existence of kainate receptors on the axon terminals of the hippocampal interneurons. Therefore other laboratories have provided data concerning the acute effects of kainate that were different from or contradictory to the results of Rodriguez-Moreno and these results on acute effects of kainate are unlikely related to the data from our chronic experiments (see following text).

Second, the effects of kainate in the experiments of Rodriguez-Moreno and coworkers (1997, 1998) were reversible within a few minutes. Therefore even if the mechanism proposed by Rodriguez-Moreno did occur when kainate was injected into rats during our experiments, it is hard to understand how the kainate effect would last for \( \geq 72 \) h in our studies (i.e., three to four orders of magnitude longer than the observed time course of Rodriguez-Moreno). Furthermore, there is simply no reasonable basis to suggest that the reversible kainate effect would last for months after injection, as necessary to explain our data from animals studied months after kainate-induced status epilepticus.

Third, and related to the second point, direct application of kainate in acute slice experiments and kainate injection into an intact animal to induce chronic epilepsy are two completely different concepts, with interneuron loss being the key issue previously raised by histopathological data after kainate-induced status epilepticus. The essence of using kainate (or other chemoconvulsants, such as pilocarpine, a muscarinic receptor agonist) to establish an animal model of chronic epilepsy is not by means of its direct effects on neurons (as would occur in acute slice experiments), but rather, by means of kainate-induced status epilepticus (i.e., repeated seizures), which causes brain damage (see following text) and triggers a pathological process of epileptogenesis (for review, see Dudek et al. 2002). The most notable and well-studied damage caused by kainate- or pilocarpine-induced status epilepticus is neuronal loss, particularly in the hippocampus. Numerous studies using the kainate or pilocarpine models, or even the kindling model (i.e., by electrical stimulation), have shown a significant loss of hippocampal interneurons, including interneurons in the dentate gyrus (Buckmaster and Dudek 1997a; Dinocourt et al. 2003; Kobayashi and Buckmaster 2003; Sayin et al. 2003). Based on extensive anatomical evidence of loss of some hippocampal interneurons after status epilepticus, one would predict a partial but significant decrease in mIPSC frequency in hippocampal principal neurons. In fact, the key result of our study (Shao and Dudek 2005) was a reduction in the frequency of mIPSCs (but not amplitude of mIPSCs, and not the frequency and amplitude of the sIPSCs) in dentate granule cells in short-term (4–7 days recovery) and long-term (>3 mo recovery) groups after kainate-induced status epilepticus. Therefore extensive histopathological data suggest that the most parsimonious explanation of a decrease in mIPSC frequency in the granule cells is a partial loss of dentate interneurons. This may be balanced by compensatory mechanisms, such as an increase of interneuron firing, possibly through a loss of inhibitory input to interneurons.

Finally, the lack of a substantial increase in sIPSCs in dentate granule cells in our study was not a “surprising” result and was not a failure to “attest the presence and function of somatodendritic ionotropic kainate receptors” as Rodriguez-Moreno suggested. Rather, it further confirms that the effect of kainate-induced status epilepticus was to kill some of the interneurons that project to dentate granule cells (this death of interneurons may have partly arisen from the direct effects of kainate on the somatodendritic region of some hippocampal interneurons) and was not a months-long direct activation of kainate receptors to cause an extensive increase of sIPSCs, as seen in acute in vitro studies on brain slices (Bureau et al. 1999; Cossart et al. 1998; Frerking et al. 1999; Semyanov and Kullmann 2001).

In conclusion, the mechanism that Rodriguez-Moreno proposes for kainate modulation of GABA transmission in acute experiments is inconsistent with work from other laboratories, and quite different from and not directly relevant to our study. That is, our observation of a reduction in the frequency of mIPSCs after kainate-induced status epilepticus was the result of a mechanism that is different from the acute kainate-induced reduction in mIPSC frequency that Rodriguez-Moreno reported in in vitro experiments.

REFERENCES


