2015

The Art of Setting Limits: Lessons from GABAergic Interneuron Transplantation

Jyoti Gupta

Janice R. Naegele
Wesleyan University, jnaegele@wesleyan.edu

The Art of Setting Limits: Lessons from GABAergic Interneuron Transplantation

Bidirectional Homeostatic Plasticity Induced by Interneuron Cell Death and Transplantation In Vivo.

Howard MA, Rubenstein JL, Baraban SC. *Proc Natl Acad Sci U S A* 2014;111:492–497.

Chronic changes in excitability and activity can induce homeostatic plasticity. These perturbations may be associated with neurological disorders, particularly those involving loss or dysfunction of GABA interneurons. In distal-less homeobox 1 (*Dlx1*$^{-/-}$) mice with late-onset interneuron loss and reduced inhibition, we observed both excitatory synaptic silencing and decreased intrinsic neuronal excitability. These homeostatic changes do not fully restore normal circuit function, because synaptic silencing results in enhanced potential for long-term potentiation and abnormal gamma oscillations. Transplanting medial ganglionic eminence interneuron progenitors to introduce new GABAergic interneurons, we demonstrate restoration of hippocampal function. Specifically, miniature excitatory postsynaptic currents, input resistance, hippocampal long-term potentiation, and gamma oscillations are all normalized. Thus, in vivo homeostatic plasticity is a highly dynamic and bidirectional process that responds to changes in inhibition.

Commentary

Neurons are endowed with the ability to dynamically regulate their ability to fire action potentials in response to alterations in overall network activity by changing the intrinsic excitability of their membranes and synapses (1). This adaptive process, termed *homeostatic plasticity*, is mediated in part by molecular signaling pathways that alter the phosphorylation of potassium ion channels, leading to changes in channel gating or clustering that alter ionic conductance in neuronal cell bodies, dendrites, or axons. Neurons can also adapt to changes in network activity by changing the number or properties of neurotransmitter receptors at afferent synapses. Yet, the role of homeostatic plasticity in abnormal conditions caused by excessive neuronal firing, such as epilepsy, is unclear.

To determine whether homeostatic plasticity can compensate for decreases in neuronal inhibition seen in epilepsy, the authors examined mutant mice with homozygous deletion of the genes encoding the transcription factor Distal-less homeobox 1 (Dlx1$^{-/-}$) [2]. In these mice, spontaneous seizures appear in an age-dependent fashion, possibly due to apoptosis of cortical and hippocampal GABAergic interneurons following or concomitant with their migration from the medial ganglionic eminence (MGE), a germinal matrix in the embryonic ventral forebrain (3). In Dlx1$^{-/-}$mice, parvalbumin interneurons innervate the somatic regions normally, but there is a pronounced deficit in subsets of MGE-derived GABAergic interneurons expressing somatostatin, calretinin, or neuropeptide Y. These populations of interneurons target both excitatory and inhibitory interneurons. By adulthood, apoptosis of GABAergic interneurons results in a 50% reduction in synaptic inhibition in cortical circuits and reduced excitatory drive to hippocampal interneurons (4).

In these mutant mice, there are deficits in both synaptic inhibition and synaptic excitation, and the question is whether homeostatic processes are involved. The authors explored this question by examining the neurochemical and electrophysiological properties of the transplanted cells and endogenous hippocampal neurons. Most of the transplanted interneurons expressed parvalbumin or somatostatin, and a smaller percentage expressed neuropeptide Y or calretinin. Remarkably, the transplanted cells eventually fired spontaneous action potentials and developed firing patterns characteristic of mature interneurons. Would these new interneurons form functional inhibitory synaptic connections with endogenous neurons in the epileptic mice? The researchers compared miniature inhibitory postsynaptic currents (mIPSCs) in *Dlx1*$^{-/-}$ and wild-type mice. This is a standard electrophysiological measure of the spontaneous release probability of synaptic vesicles containing GABA (5). mIPSC frequency was reduced in *Dlx1*$^{-/-}$ mice, as expected, and transplantation of GABAergic interneurons increased synaptic inhibition to levels comparable to wild-type mice.

Several ways that neurons might compensate for decreased inhibitory drive include decreasing the strength of synaptic excitation or altering intrinsic membrane properties. To test for changes in synaptic excitation, the investigators recorded miniature excitatory postsynaptic currents (mEPSCs) from CA1 pyramidal cells. As with the trend for mIPSCs, the frequency of mEPSCs was decreased after interneuron loss and returned to

Epilepsy Currents, Vol. 15, No. 6 (November/December) 2015 pp. 347–348

wild-type levels after transplantation of newborn GABAergic interneurons. Reduced mEPSC frequency in CA1 pyramidal cells probably results from compensatory homeostatic plasticity in response to the genetically-induced decrease in synaptic inhibition. However, decreased mEPSCs could also result from abnormal development of excitatory synapses, altered presynaptic release of glutamate, or other mechanisms. The authors distinguished between these alternatives by measuring the paired pulse ratio, a test of presynaptic release probability. The paired pulse ratio was comparable in *Dlx1*$^{-/-}$ mice with or without MGE cell transplants, ruling out this mechanism. In contrast, the ratio of EPSCs due to AMPA versus NMDA postsynaptic receptors was reduced in *Dlx1*$^{-/-}$ mice but was normal in mice with MGE cell transplants. In comparison to WT mice, EPSP slopes in *Dlx1*$^{-/-}$ mice were decreased, further supporting the reduction in postsynaptic glutamate receptors as an explanation. These findings suggest that reduced synaptic inhibition through the loss of inhibitory interneurons may lead to compensatory changes in postsynaptic excitatory receptors—consistent with homeostatic plasticity as a mechanism for some of the observed changes in *Dlx1*$^{-/-}$ mice.

Prior work in neuronal cell cultures had already established that homeostatic plasticity, following bicuculline-induced blockade of inhibitory currents, accounts for changes in intrinsic excitability of neurons by regulating the strength of excitatory currents (6). The present study extends our understanding of the signals that regulate homeostatic plasticity to an *in vivo* mouse model of developmental epilepsy caused by interneuron loss (interneuronopathy). However, the ability of hippocampal pyramidal cells to reduce their excitatory currents in response to decreased inhibition was not sufficient to normalize cortical circuits.

In addition to characterizing intrinsic changes in neuronal plasticity resulting from decreased inhibition, these authors also showed that *Dlx1*$^{-/-}$ mice exhibit reduced gamma frequency oscillations (GFOs), a form of cortical brain activity conducted by fast spiking parvalbumin-expressing basket cells when firing in synchrony. These rhythms are implicated in spatial and working memory; the disruption of GFOs is thought to underlie cognitive deficits in some types of human epilepsy (7). GFO disturbances might also result from disruptions in the ratio of different subpopulations of interneurons, or from altered inhibitory input to different regions of membrane, such as the soma or dendrites. *Dlx1*$^{-/-}$ mice do not appear to lose parvalbumin interneurons, so the observed deficits in GFOs likely result from altered levels of perisomatic inhibition from the PV interneurons.

This elegant study shows that homeostatic plasticity is an important mechanism that balances excitation and inhibition in cortical and hippocampal networks. It also demonstrates a mechanism for how neurons adapt their excitability following interneuron transplantation in pathological conditions such as epilepsy. Stem cell–based treatments for severe epilepsy with intractable seizures would be considered only long after the development of interneuronopathy and epileptogenesis and after less invasive pharmacological attempts had been tried and failed. Whether homeostatic plasticity is maintained throughout life in individuals with epilepsy is an important unresolved issue because such treatments might not be effective for adult-onset epilepsy if homeostatic plasticity is diminished or lost in chronic epilepsy. These concerns would likely need to be addressed when devising future stem cell–based clinical treatments for severe, intractable forms of adult-onset epilepsy.

by Jyoti Gupta, MS, and Janice R. Naegele, PhD

References

1. 1. Turrigiano GG, Nelson SB. Homeostatic plasticity in the developing nervous system. *Nat Rev Neurosci* 2004;5:97–107.
2. Surmeier DJ, Foehring R. A mechanism for homeostatic plasticity. *Nat Neurosci* 2004;7:691–692.
3. Cobos I, Calcagnotto ME, Vilaythong AJ, Thwin MT, Noebels JL, Baraban SC, Rubenstein, JLR. Mice lacking Dlx1 show subtype-specific loss of interneurons, reduced inhibition and epilepsy. *Nat Neurosci* 2005;8:1059–1068.
4. Jones DL, Howard MA, Stanco A, Rubenstein JL, Baraban SC. Deletion of Dlx1 results in reduced glutamatergic input to hippocampal interneurons. *J Neurophysiol* 2011;105:1984–1991.
5. Zhang J, Yang Y, Li H, Cao J, Xu L. Amplitude/frequency of spontaneous mEPSC correlates to the degree of long-term depression in the CA1 region of the hippocampal slice. *Brain Res* 2005;1050:110–117.
6. Turrigiano GG, Leslie KR, Desai NS, Rutherford LC, Nelson SB. Activity-dependent scaling of quantal amplitude in neocortical neurons. *Nature* 1998;391:892–896.
7. Worrell GA, Parish L, Cranstoun SD, Jonas R, Baltuch G, Litt B. High-frequency oscillations and seizure generation in neocortical epilepsy. *Brain* 2004;127(Part 7):1496–1506.