Galanin is a 29 amino acid peptide (30 in humans), which is widely distributed in the brain. Galanin has been implicated in multiple physiological actions such as appetite, mood, anxiety, and learning. Galanin is also believed to play a role in diseases including epilepsy (1). Much of the knowledge about galanin’s role in epilepsy comes from studies of status epilepticus involving the hippocampus. The hippocampus and, particularly, the dentate gyrus receive a rich innervation of galanin-containing fibers. Cholinergic fibers from septum contain galanin in addition to acetylcholine. Noradrenergic fibers from locus caeruleus innervating the hippocampus also contain the neuropeptide. Galanin appears to be secreted...
Galanin Receptors Modulate Seizures

during high frequency stimulation and modulates neurotransmitter release from the presynaptic terminals.

Initial evidence of a role for galanin in hippocampal seizures came from studies by Mazarati and colleagues (2). When galanin or its analogs were infused into the hippocampus, it prevented or terminated status epilepticus. Furthermore, status epilepticus itself depleted galanin in the hippocampus. Thus, there were effects of galanin on seizures and effects of seizures on galanin itself. Of interest, repeated seizures can induce galanin expression in neurons and interneurons over a period of time (2).

Further evidence for the role of galanin in seizures and epilepsy has come from mice overexpressing galanin and receptor knockouts, and the development galanin receptor–specific drugs. Galanin binds to three kinds of receptors: GalR-1, GalR-2, and GalR-3. Both GalR-1 and GalR-2 receptors are expressed in the hippocampus. GalR-1 receptors are linked to a pertussis toxin–sensitive G-protein, which inhibits the synthesis of cyclic AMP. The GalR-2 receptor is linked to G-proteins, which are pertussis toxin insensitive and appear to modulate inositol phosphate (IP) synthesis and turnover. These receptors also have other actions (3).

The seizure threshold is raised in mice that oversynthesize the neuropeptide. In contrast, when the GalR-1 receptor is knocked out, approximately 25% of the animals have seizures spontaneously. The remaining mice with this gene knocked out had increased sensitivity to seizures. In galanin overexpressing mice, there is a reduced release of glutamate following high-frequency stimulation, suggesting that galanin may be reducing neurotransmitter release. Other studies report reduced excitability of postsynaptic neurons in galanin-overexpressing mice. Reduction of GalR-2 receptors by RNA knock down also results in increased susceptibility of seizures. Other studies have used the galanin gene to modulate seizures. Galanin gene delivered by adeno associated virus (AAV) into the hippocampus reduces the severity and the intensity of kainate-induced seizures (4). Thus, multiple converging lines of evidence suggest an important role for galanin in modulating seizures. However, several questions remain unresolved with regards to the role of galanin in seizures and the potential use of galanin as an anticonvulsant.

Small molecule drugs that cross the blood brain barrier to modulate galanin receptors have not been available. Most analogs used in the experimental studies were small peptide fragments of the large galanin molecule, which do not cross the blood brain barrier. These peptides have limited therapeutic value for treating epilepsy. Furthermore, the goal of gene therapy with galanin remains in the distant future until the challenges of safe and effective gene delivery to central neurons are overcome. A practical solution to these problems would be to find nonpeptide molecules that would bind to specific galanin receptors and terminate seizures. However, the identity of the receptor that modulates seizures is unclear, and previous studies would suggest that GalR-1 receptors would be the targets for drug development.

The recent article by Lu et al. makes progress in this regard in a surprising direction. CYM 2503 was identified by screening compounds for activity on GalR-2 receptor-expressing Human Embryonic Kidney (HEK) cells, as a nonpeptide allosteric modulator of GalR-2 receptors. The compound increased IP accumulation in the presence of galanin but had limited intrinsic effect. It shifted the galanin concentration response curve to the left but did not displace it from binding sites.

Peripheral (intraperitoneal) administration of CYM 2503 suppressed seizures. The authors confirmed the efficacy of this new nonpeptide modulator of GalR-2 receptors in the maximal electroshock model. It increased latency to hind limb clonus, suppressed seizures, and decreased mortality. In a model of lithium pilocarpine–induced status epilepticus, it CYM 2503 was as effective as levetiracetam in terminating seizures. The treatment of status epilepticus was carried out without giving benzodiazepines. The treatment of status epilepticus almost always involves initial treatment with benzodiazepines, and a second line drug given in combination with these drugs. It is possible that the analog would have terminated status epilepticus in all animals if it had been used in combination with benzodiazepines.

Schauwecker’s study describes another emerging role for galanin receptors (5). An important consequence of prolonged seizures or status epilepticus is the loss of principal neurons of the hippocampus and interneurons in the hilus. This seizure-induced neuronal loss is believed to be mediated by excitotoxins. Schauwecker and colleagues in a seminal study demonstrated that susceptibility to neuronal loss caused by excitotoxins is genetically determined (5). Certain genetic background mice are resistant to excitotoxic cell death, whereas others are susceptible. In a systematic set of studies over a decade, the group has searched for the genes that confer susceptibility to seizure-induced cell death. The region of interest was localized to an expanson on mouse chromosome 18 that contains the GalR-1 receptor gene.

Schauwecker demonstrates in this study that animals resistant to excitotoxic cell death become sensitive to it when the GalR-1 receptor is knocked-out out. These GalR-1 knockouts were characterized in detail, and they did demonstrate compensatory overexpression of GalR-2 receptors or galanin. Latency, severity, and duration of severe seizures were similar in wild-type and knock-out mice. Pharmacologic block of GalR-1 receptors also rendered wild-type mice susceptible to neuronal damage.

These studies suggest that the susceptibility to excitotoxic cell death is mediated at least in part by the GalR-1 receptor. It is possible that there are polymorphisms or mutations of the GalR-1 receptor in susceptible mice, which render the receptor less effective. However, it is also possible that the second messenger systems activated by Gal-R-1 receptors are less sensitive to galanin activation. Further studies are needed to figure out the molecular mechanisms of susceptibility. However, these findings are very exciting because they begin to unravel the mystery of differential sensitivity to excitotoxic cell death.

Finally, the two studies together raise further questions about the role of GalR-1 and GalR-2 receptors in seizures and epilepsy. The study by Lu et al. suggests that GalR-2 receptors are important for seizure termination and should be targeted for further drug development. Presumably, prolonged...
seizures were responsible for excitotoxic cell death observed in mice. However, the study by Schauwecker suggests that GalR-1 receptors should be targeted for preventing cell death. It is possible that generation of seizures and generation of cell death are mediated by two distinct pathways even though both may involve glutamnergic transmission. A further careful analysis of electrographic seizures in these knock-out mice and perhaps generation of GalR-2 receptor knock-out mice will address these questions. Overall, these studies point to galanin receptors as an important therapeutic target for treatment of seizures and status epilepticus and perhaps a pathway towards preventing seizure-induced cell death in people.

by Jaideep Kapur, MD, PhD

References
Baltimore, MD
Baltimore Convention Center
December 2 - 6, 2011

2011
American Epilepsy Society
65th Annual Meeting

Important Deadlines
October 28 Early Bird Discount
October 30 Hotel Reservations
November 17 Pre-Registration

www.AESNET.org

Future Annual Meeting Dates

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San Diego Convention Center
November 30 – December 4

2013
Washington, D.C.
Washington Convention Center
December 6 – 10

2014
Seattle, WA
Washington State Convention and Trade Center
December 5 – 9

2015
Philadelphia, PA
Pennsylvania Convention Center
December 4 – 8

2016
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American Epilepsy Society

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1. Today’s Date: _____________ April 5th 2011 ______________________

2. First Name _______ Jaideep ____________ Last Name ________ Kapur ________ Degree ___ MD, PhD ___

3. Are you the Main Assigned Author? _X_ Yes _____ No

If no, enter your name as co-author _______________________________________________________

4. Manuscript/Article Title: _______ Galanin receptors modulate seizures _________________________________

5. Journal Issue you are submitting for: 11.4 ____________________________________________________________________

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Did you or your institution at any time receive payment or services from a third party for any aspect of the submitted work (including but not limited to grants, data monitoring board, study design, manuscript preparation, statistical analysis, etc.)?

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* This means money that your institution received for your efforts on this study.
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Place a check in the appropriate boxes in the table to indicate whether you have financial relationships (regardless of amount of compensation) with entities as described in the instructions. Use one line for each entity; add as many lines as you need by clicking the “Add” box. You should report relationships that were present during the 36 months prior to submission.

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