In Search of a New and Improved Target for Antiepileptic Drugs: Sialic Acid?

Role of Extracellular Sialic Acid in Regulation of Neuronal and Network Excitability in the Rat Hippocampus. Isaev D, Isaeva E, Shatskih T, Zhao Q, Smits NC, Shworak NW, Khazipov R, Holmes GL. *J Neurosci* 2007;27(43):11587–11594. The extracellular membrane surface contains a substantial amount of negatively charged sialic acid residues. Some of the sialic acids are located close to the pore of voltage-gated channel, substantially influencing their gating properties. However, the role of sialylation of the extracellular membrane in modulation of neuronal and network activity remains primarily unknown. The level of sialylation is controlled by neuraminidase (NEU), the key enzyme that cleaves sialic acids. Here we show that NEU treatment causes a large depolarizing shift of voltage-gated sodium channel activation/inactivation and action potential (AP) threshold without any change in the resting membrane potential of hippocampal CA3 pyramidal neurons. Cleavage of sialic acids by NEU also reduced sensitivity of sodium channel gating and AP threshold to extracellular calcium. At the network level, exogenous NEU exerted powerful anticonvulsive action both in vitro and in acute and chronic in vivo models of epilepsy. In contrast, a NEU blocker (*N*-acetyl-2,3-dehydro-2-deoxyneuraminic acid) dramatically reduced seizure threshold and aggravated hippocampal seizures. Thus, sialylation appears to be a powerful mechanism to control neuronal and network excitability. We propose that decreasing the amount of extracellular sialic acid residues can be a useful approach to reduce neuronal excitability and serve as a novel therapeutic approach in the treatment of seizures.

COMMENTARY

It is well known that over the last 20 years, the availability of several new antiepileptic drugs (AEDs) has not greatly reduced the percentage of patients who are considered intractable or pharmacoresistant (1). A central tenet in the search for new AEDs is that novel targets for selectively blocking epileptic seizures are needed in order to provide better treatment for people with epilepsy. The study by Isaev and coworkers aims to provide evidence that neuraminidase (NEU), which is a bacterial enzyme that cleaves sialic acid residues, may reveal potential new strategy for blocking seizures. The authors offer a rigorous and comprehensive analysis of the effects of NEU on sodium currents, action potential threshold, and in vitro and in vivo seizures, using the high-potassium and kindling models. The key finding is that NEU caused a depolarizing shift in action potential threshold without altering resting membrane potential, which in turn, elevated seizure threshold, blocking the occurrence of stimulated seizures in both the in vitro and in vivo models tested.

One of the strengths of the report by Isaev and colleagues is the degree to which it is designed to identify a new molecular target. The work essentially starts with an assessment of the effects of NEU on sodium currents and ends with an analysis of drug effects on kindled seizures. Although NEU effectively suppresses sodium currents and blocks these experimental seizures, the findings leave open the question of whether NEU or a related molecule will be effective for chronic epileptic seizures. The kindling model has been used to study the progressive component of epileptogenesis, but kindled seizures still are electrically evoked seizures—possibly quite different from the spontaneous seizures observed in an appropriate animal model or in human studies of chronic epilepsy. Virtually all of their trials involved acute dosing, which does not address the issue of whether chronic dosing would be effective at suppressing spontaneous seizures. Furthermore, in the in vitro experiments, the brain slices were bathed for 2 h in the NEU solution, and the in vivo experiments involved NEU injections 12 h prior to stimulation. Thus, NEU itself is not going to be a clinical treatment for epilepsy, but sialic acid could conceivably be a target

Epilepsy Currents, Vol. 8, No. 3 (May/June) 2008 pp. 79–80
Blackwell Publishing, Inc.

for a new AED. Unfortunately, a great deal of work will be necessary to move from an enzyme that cleaves sialic acid residues to a small molecule that might perturb sialic acid residues but probably will not cleave them.

The findings of this study introduce the possibility that NEU molecular interactions with sialic acid might be more effective than the traditional sodium-channel blocker AEDs, such as phenytoin and carbamazepine. Interestingly, the traditional AEDs are thought to block seizures by acting on sodium-channel inactivation, thus causing a use-dependent block that reduces high-frequency repetitive firing without greatly altering the threshold for sodium-mediated action potentials or low-frequency repetitive firing. The rationale underlying use of traditional sodium-channel blocker AEDs is that seizures involve particularly large depolarizations with abnormal high-frequency firing of action potentials—one that is different from sodium-channel function during normal behavior. Thus, drugs that act on this high-frequency firing mechanism should theoretically be less apt to have nonspecific effects on other normal neural activity. Accordingly, will elevating sodium-current threshold with NEU actually block seizures without having effects on other functions of these neural circuits or on normal behaviors? Intuitively, it seems likely that drugs that preferentially block high-frequency, repetitive firing versus ones that raise threshold for action potentials would more selectively block seizures without affecting behavior, but this assumption has not yet been tested.

A key concept in the development of potential new AEDs is to study behavioral toxicity early in the screening process as the agent is assessed for efficacy in suppressing seizures. Dose-dependent toxicity is obviously an important issue in screening of any AED (2,3), and a drug with substantial toxicity at doses that suppress seizures is unacceptable. Thus, the dose-response between establishing antiseizure actions is only meaningful in relationship to the negative effects on normal behaviors (e.g., cognitive tests). Thus, these studies raise the issue of whether potential new AEDs should not only block mechanisms that are known to be active during seizures, but also not affect those that are likely to be active during normal brain function. The question is whether new AEDs should target the threshold for voltage-gated sodium current (and thus, action potential threshold) as a mechanism to block seizures, since virtually all neurophysiological mechanisms underlying normal behaviors engage sodium-mediated action potentials. Future research undoubtedly will need to target this and related questions.

The Isaev et al. report is best viewed as an initial study on sialic acid and NEU, with intriguing observations that deserve further investigation. All of the technical and conceptual issues aside, the study reminds the neuroscience community about the importance of glycoproteins in the modulation of neuronal and network excitability. For investigators interested in developing treatments for epilepsy, sialic acid might be a new target to explore. It is very unlikely that NEU, itself, could be an agent, as it would be difficult to administer over the long term, and it is unclear how it could be delivered to the epileptogenic zone or seizure focus. However, simple molecules that are activated when orally administered and that interact with sialic acid in some way to reduce neuronal excitability might be worth evaluating. Whether this approach would have the same therapeutic effect when applied globally rather than through focal infusion also will need to be evaluated. For the moment, the study provides reason to consider indirect approaches to the modulation of neuronal excitability, rather than the full frontal assault of direct channel agonists or antagonists. Ultimately, it may be that a combination of several indirect approaches will yield greater benefits in selectively suppressing seizures in epilepsy.

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References

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