

Turning up the heat on L-type Ca^{2+} channels promotes neuronal firing and seizure activity

Andrew P Braun

Department of Physiology and Pharmacology; Faculty of Medicine; University of Calgary; Calgary, AB Canada

It is well recognized clinically that fever in young children (< 6 y of age) may lead to seizure activity in a small, but significant percentage of these individuals, which may have negative consequences for the developing brain and progressive cognitive function. In rodent models, exposure of acute brain slices to hyperthermic temperatures (i.e., 38–41°C) is reported to evoke membrane depolarization and increased neuronal firing, although the underlying molecular/cellular events responsible for these phenomena are not fully understood. Elevated temperature may alter membrane excitability by influencing individual ion channels within a given neuron, or alter the behavior and connectivity of neurons and glia that operate within a local network. In the present study, Radzicki and colleagues have examined the possibility that modest increases in tissue/body temperature (up to 40.5°C) may enhance the activity of voltage-gated Ca^{2+} channels, which could then promote spontaneous firing of individual neurons and greater network discharge. The results of this work indicate that fever-like temperatures positively and reversibly influence the gating properties of L-type Ca^{2+} channels, and that the L-type blocker nimodipine reduces both temperature-induced increases in spontaneous neuronal firing and the incidence/duration of discharge activity in a whole animal model of febrile seizure.

Experimentally, the authors utilized whole cell voltage clamp, along with extracellular field recordings, to monitor

voltage-dependent Ca^{2+} current activity and neuronal membrane depolarization/firing in acute hippocampal brain slices prepared from male rats at postnatal day 18. In whole animal studies, behavioral seizure activity and ictal discharges induced by endotoxin (i.e., lipopolysaccharide) injection plus moderate hyperthermia (i.e., body temperature up to 41.5 °C) were monitored in P14 animals.

In acutely prepared hippocampal slices, the authors found that pyramidal neurons subjected to ramp-style increases in ambient bath temperature (i.e., from 33 to 40.5 °C over a 5–6 min period) exhibited a modest membrane depolarization and a significant increase in their rate of spontaneous action potential firing, which was reversible upon cooling. The increase in electrical firing activity typically occurred at a threshold temperature of ~38 °C, and was still prominent in the presence of pharmacologic blockers of fast synaptic transmission (i.e., kynurenic acid and picrotoxin), indicating that the increase was due, in part, to changes in the intrinsic excitability of the neuron, and not strictly dependent upon stimulatory signals from neighboring cells. A qualitatively similar effect was also recorded from slices prepared from other brain areas (i.e., entorhinal cortex and occipital cortex), indicating that the observed temperature-induced firing activity was not unique to hippocampal neurons.

Using pharmacologic agents to eliminate voltage-gated Na^+ and K^+ channel currents, the authors observed that elevated temperature increased the activity of a cadmium- and dihydropyridine (nimodipine)-sensitive,

Keywords: neuron, fever, hyperthermia, hippocampus, $\text{Ca}_v1.2$, nimodipine

Submitted: 07/18/13

Accepted: 07/22/13

<http://dx.doi.org/10.4161/chan.25956>

Correspondence to: Andrew P. Braun;
Email: abraun@ucalgary.ca

Commentary to: Radzicki D, Yau HJ, Pollema-Mays SL, Mlsna L, Cho K, Koh S, et al. Temperature-sensitive $\text{Ca}_v1.2$ calcium channels support intrinsic firing of pyramidal neurons and provide a target for the treatment of febrile seizures. *J Neurosci* 2013; 33:9920-31; PMID:23761887; <http://dx.doi.org/10.1523/JNEUROSCI.5482-12.2013>

inward Ca^{2+} current, which was evident at the membrane holding potential of -65 mV. To account for this increased Ca^{2+} current activity at hyperpolarized potentials, the authors observed that elevation of bath temperature from 35 to 39–40°C produced a modest leftward shift of ~ 8 mV in the Ca^{2+} current activation curve, along with a rightward shift of ~ 10 mV in the voltage-dependent inactivation curve. Taken together, these two effects would be expected to substantially increase voltage-gated Ca^{2+} channel availability/opening at the typical neuronal resting membrane potential of -65 mV. Although RT-PCR analysis of neurons in the CA1 hippocampal region revealed prominent expression of mRNA encoding both dihydropyridine-sensitive Cav1.2 and Cav1.3 isoforms, recordings of temperature-sensitive Ca^{2+} current activity in hippocampal slices from Cav1.3 knockout mice did not reveal any noticeable differences in current amplitude compared with control mice, suggesting that Cav1.3 channels may play only a minor role in the observed temperature-sensitive responses. What is not clear from these studies, however, is whether low voltage-activated, Cav3.x (i.e., T-type) Ca^{2+} channels play any role in this response. Although nimodipine has been used as a selective inhibitor of L-type channels, this drug is also reported to block Cav3.2 channels with an IC_{50} value of 5–10 μM .¹ Such data raise the possibility that T-type channels, which may be active at -65 mV, may also contribute to the observed temperature-sensitive Ca^{2+} current.

Members of the transient receptor potential (TRP) family of cation channels, particularly the TRPV isoforms, display temperature-sensitive gating, such that

their open probability increases with elevated ambient temperature (i.e., Q_{10} values for this effect range from 10–20). TRPV channels are abundant in peripheral sensory afferent fibers and typically mediate sensory perception of both warmth and noxious heat. In hippocampal slices, pharmacologic blockade of TRPV channels did not decrease the magnitude of inward background current and membrane depolarization evoked by elevated temperature, suggesting that other ionic conductances must be responsible for these effects. Experimentally, substitution of extracellular Na^+ by impermeable choline revealed the presence of a temperature-induced inward current at the resting membrane potential. This additional conductance, likely mediated by a Ca^{2+} - and voltage-insensitive Na^+ leak channel (i.e., NALCN) appears to be functionally important for depolarizing neurons to a threshold potential (i.e., -65 to -60 mV) that would encourage the opening of voltage-gated, L-type Ca^{2+} channels at the elevated temperature. Once activated, nimodipine-sensitive L-type channels would cause additional membrane depolarization and promote action potential firing via activation of fast, voltage-gated Na^+ channels. Importantly, this excitatory process would not be expected to occur at normal tissue temperatures of 36–37°C, as the enhanced gating properties of L-type Ca^{2+} channel required to drive intrinsic neuronal firing are not exhibited at tissue temperatures $< 39^\circ\text{C}$.

The observed inhibition of temperature-induced, neuronal firing by 0.5–3 μM nimodipine in hippocampal slices suggested that similar low doses may also prevent temperature-induced, seizure-like

activity in vivo. Using a combination of endotoxin (i.e., lipopolysaccharide) injection and hyperthermia to induce febrile seizures in immature rats, the authors noted that low dose nimodipine treatment significantly reduced the proportion of animals exhibiting generalized tonic-clonic (i.e., behavioral) seizure activity from 80 to 20%, and further decreased the duration of ictal episodes nearly 10-fold, as determined by EEG recordings.

Collectively, these results suggest that hyperthermia/fever-evoked increases in the spontaneous firing activity of CNS neurons is due in large part to excitatory shifts in the activation and inactivation behavior of neuronal L-type Ca^{2+} channel gating, leading to greater activity of these channels at more hyperpolarized membrane potentials. Moreover, pharmacologic blockade of these channels in vivo appears to significantly reduce both the incidence and duration of fever-associated seizure activity in an immature rat model. Based on the scope of these findings, the authors suggest that L-type Ca^{2+} channel blockade may represent a novel therapeutic approach to treat febrile seizure in young children, in whom classic anti-epileptic drugs, such as blockers of fast, voltage-gated Na^+ channels, may be contraindicated.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

References

1. Furukawa T, Nukada T, Namiki Y, Miyashita Y, Hatsuno K, Ueno Y, et al. Five different profiles of dihydropyridines in blocking T-type Ca^{2+} channel subtypes (Ca(v)3.1 (α_{1G}), Ca(v)3.2 (α_{1H}), and Ca(v)3.3 (α_{1I})) expressed in *Xenopus* oocytes. *Eur J Pharmacol* 2009; 613:100-7; PMID:19401195; <http://dx.doi.org/10.1016/j.ejphar.2009.04.036>