Channels 8:5, 389–390; September/October 2014; © 2014 Taylor & Francis Group, LLC

Cortical KCNQ2/3 channels; insights from knockout mice

Heun Soh[†], Zachary Niday[†], and Anastasios V Tzingounis*

Department of Physiology and Neurobiology; University of Connecticut; Storrs, CT USA

[†]These authors contributed equally to this work.

KCNQ2 and KCNQ3 potassium channels are critical for proper brain function, as mutations in either *Kcnq2* or *Kcnq3* have been associated with multiple pediatric epilepsy disorders.[1] In most neurons, KCNQ2 and KCNQ3 channels are found as heteromers (KCNQ2/3) mediating the M-current, a non-inactivating potassium conductance that activates as neurons approach action potential threshold.[2] Additionally, KCNQ2/3 channels underlie the medium afterhyperpolarization (mAHP) and, depending on cell type, contribute to the slow afterhyperpolarization, thus preventing runaway activity following trains of action potentials.[2,3] Despite great progress in our understanding of the physiological, biophysical and trafficking properties of KCNQ2/3 heteromers,[2] the lack of selective KCNQ2 and KCNQ3 channel blockers and the fact that constitutive *Kcnq2*-knockout mice die soon after birth has made it challenging to explore the precise role of individual KCNQ2 and KCNQ3 channels in neuronal excitability. In a new publication,[4] Soh et al address this knowledge gap by crossing newly developed *Kcnq2*- and *Kcnq3*-floxed mice to the *Emx1-ires-Cre* driver line, which express cre recombinase in ~90% of cortical neurons starting at ~E10.5. Using these mouse models, the authors demonstrate the essential role for KCNQ2 in controlling neuronal excitability.

Previous studies showed that knockin mice expressing loss-of-function mutations to KCNQ2 and KCNQ3 channels mimicking those observed in human epilepsy could lead to seizures and death.[5] Soh et al extended these findings by showing that deletion of *Kcnq2*—but not *Kcnq3*—from cortical pyramidal neurons is sufficient for the development of aberrant EEG activity and leads to death by the third week of life. Electrophysiological recordings from pyramidal neurons in the CA1 subregion of the hippocampus showed that in the absence of KCNQ2 channels the M-current was reduced by ~85%, whereas the mAHP was reduced by ~50–60%. The reduction to the mAHP also led to prolonged afterdepolarization likely leading to the higher number and frequency of action potentials observed in *Kcnq2*-null neurons. Unlike *Kcnq2*-null neurons, *Kcnq3*-null neurons had near-normal excitability and mAHP even though the M-current was reduced by approximately 50%.

The authors also found that loss of KCNQ2 channels resulted in a greater decrease in expression of the remaining cortical KCNQ channels, KCNQ3 and KCNQ5, while *Kcnq3* deletion only resulted in small reductions in KCNQ2 and KCNQ5 protein levels. Further pharmacological analysis suggested that, in the absence of KCNQ3, KCNQ2 channels are still functional, possibly compensating for the loss of KCNQ3 channels. Therefore, the observed severe phenotype of *Kcnq2* conditional-knockout mice might simply be due to the global loss of cortical KCNQ channels, thereby decreasing the likelihood of compensation by other KCNQ channels.

While the Soh et al.[4] study critically expands our understanding of KCNQ2/3 channel function in pyramidal neurons, it also introduces several new questions. For instance, KCNQ2/3 channels are highly expressed in the distal region of the axon initial segment (AIS) where they co-localize with Nav1.6, a voltage-gated sodium channel (VGSC) also implicated in pediatric epilepsies and the initiation of action

Keywords: Epilepsy, KCNQ2, KCNQ3, M-current, **medium afterhyperpolarization, epilepsy**

*Correspondence to: Anastasios V Tzingounis; Email: anastasios.tzingounis@uconn.edu

Submitted: 07/10/2014

Accepted: 07/11/2014

http://dx.doi.org/10.4161/19336950.2014.948755

Soh H, Pant R, Loturco JJ, Tzingounis AV. Conditional deletions of epilepsy-associated KCNQ2 and KCNQ3 channels from cerebral cortex cause differential effects on neuronal excitability. J Neurosci 2014; 34(15):5311-21; http://dx.doi.org/10.1523/JNEUROSCI.3919-13.2014

potentials.[6,7] Does loss of KCNQ2/3 channels lead to higher activity of Nav1.6?

One could envision that the overlap of KCNQ2/3 channels with sodium channels might limit their persistent activity at subthreshold membrane potentials, leading to increased excitability and prolonged afterdepolarization.[6,7] Increased persistent sodium current might also lead to greater contribution to the mAHP following trains of action potentials. As the mAHP hyperpolarizes the membrane potential it will also transiently close the persistent sodium current leading to an amplified hyperpolarization, as the closing of inward sodium current is functionally similar to the opening of a potassium channel. A greater contribution of sodium channels to the mAHP might lead to action potential synchronization. The involvement of VGSCs to the mAHP might explain the discrepancy between the large M-current reduction (measured in the presence of TTX, a VGSC blocker) and the persistence of the mAHP in *Kcnq2*- and *Kcnq3*-null neurons. Additionally, as KCNQ2 channels bind to CK2,[8] a kinase required for the proper localization of VGSC to the AIS, KCNQ2 loss might also change the local concentration of CK2, leading to mislocalization of VGSCs in the AIS and a change in action potential properties. Future work combining *Kcnq2, Kcnq3* and sodium channel knockout mice might clarify these questions.

The aberrant cortical activity measured with EEG recordings in *Kcnq2*-null neurons suggests a hyperexcitable cortical network. Therefore, does loss of KCNQ2 channels also lead to increased spontaneous and/or mini EPSC activity? Does it also homeostatically affect IPSC activity? As cortical interneurons also express KCNQ2/3 channels, does conditional KCNQ2/3 deletion simultaneously from both interneurons and pyramidal neurons exacerbate or alleviate cortical hyperexcitability? This is an important issue, as the role of KCNQ2/3 channels in interneuron physiology has been largely unexplored compared to the plethora of studies on KCNQ2/3 channels in pyramidal and sensory neurons. Lastly, what leads to the premature death of *Kcnq2* conditional knockout mice—bradycardia, respiratory failure, or both?

In summary, the availability of *Kcnq2*- and *Kcnq3*-floxed mice will be invaluable for genetically defining the role of KCNQ2/3 channels throughout the nervous system and could also contribute to a better understanding of *Kcnq2* and *Kcnq3* epilepsy-related disorders.

References

1. Scott RC, et al. Nat Med 2012; 18:1624-5; PMID: 23135515; http://dx.doi.org/10.1038/nm.2987
2. Cooper EC. Semin Cell Dev Biol 2011; 22:185-92; PMID:20940059; http://dx.doi.org/10.1016/j.semcdb.2010.10.001
3. Tzingounis AV, et al. Proc Natl Acad Sci 2008; 105:19974-9; PMID:19060215; http://dx.doi.org/10.1073/pnas.0810535105
4. Soh H, et al. J Neurosci 2014; 34:5311-21; PMID:24719109; http://dx.doi.org/10.1523/JNEUROSCI.3919-13.2014
5. Singh NA, et al. J Physiol 2008 586:3405-23; PMID:18483067; http://dx.doi.org/10.1113/jphysiol.2008.154971
6. Battefeld A, et al. J Neurosci 2014 34:3719-32; PMID:24599470; http://dx.doi.org/10.1523/JNEUROSCI.4206-13.2014
7. Hu W, et alY. Nat Neurosci 2009; 12:996-1002; PMID:19633666; http://dx.doi.org/10.1038/nn.2359
8. Kang S, et al. J Biol Chem 2014; 289:11536-44; PMID:24627475; http://dx.doi.org/10.1074/jbc.M113.528497