A de novo mutation in the Sodium-Activated Potassium channel KCNT2 changes channel function and causes epileptic encephalopathy

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Background

Early infantile epileptic encephalopathy (EIEE) is a key epilepsy target gene

K+ Channels: KCNT1 and KCNT2

- Sodium-activated potassium channels contribute to firing accommodation and maintenance excitability of neurons.
- Despite high sequence homology and structural similarities, KCNT1 and KCNT2 channels appear to have very different physiological and pathophysiological roles.

Phe240Leu is a de novo epilepsy target gene

- Most of the identified mutations are gain-of-function variants increasing peak SLACK currents 3-12 fold.
- Despite high sequence homology and structural similarities, KCNT1 and KCNT2 channels appear to have very different physiological and pathophysiological roles.

Expression

They have distinct, often overlapping, patterns of localization within the central and peripheral nervous system.

Regulation

De novo KCNT2 mutation in an EOEE patient

- Substitution of Phe240 with polar and some non-polar residues induced constitutive channel activation.
- Hydrophobic interactions between Phe240 and residues in SS stabilize Slo2.1 channels in a closed state.

Hypothesis

Phe240Leu causes constitutive activity and ‘change-in-function’ of rSlick and ISlick in Xenopus Oocytes

Phe240Leu alters (Cl) sensitivity and K+ selectivity of rSlick channels in CHO Cells

Clinical presentation of the epileptic encephalopathy patient

- From 3 months, he had multiple daily seizures lasting up to 8 minutes.
- Currently at 4 years, he has multiple daily seizures that have remained resistant to UKISS trial, a ketogenic diet and the following anti-epileptic medications:
  - ethosuxamide, pyridoxal phosphate.
- At 4 months, he developed daily epileptic spasms lasting up to 8 minutes.
- Despite high sequence homology and structural similarities, KCNT1 and KCNT2 channels appear to have very different physiological and pathophysiological roles.

SUMMARY

- Early-onset epileptic encephalopathies (EIEEs) are a debilitating spectrum of disorders associated with cognitive impairments.
- We present the first clinical report of a KCNT2 mutation in an EIEE patient; The de novo heterozygous variant Phe240Leu Slick was identified by exome sequencing and confirmed by Sanger sequencing.
- Phe240Leu rSlick and hSlick channels were electrophysiologically characterized in CHO cells and Xenopus laevis oocytes, respectively, to reveal three significant alterations to channel function.
- Phe240Leu channels displayed constitutive activity.
- [Cl] sensitivity of WT channels was reversed in Phe240Leu channels.
- K+ selectivity WT channels were made non-selective by Phe240Leu.
- Further, rSlick channels induced membrane hyperexcitability when expressed in primary neurons, resembling the cellular seizure phenotype.
- Our results confirm that Phe240Leu is a ‘change-in-function’ KCNT2 mutation, the first description of altered selectivity in KNa channels, and indeed, K+ channels.
- Overall, the results establish pathogenicity of the Phe240Leu KCNT2 mutation in the reported EIEE patient.

Contributors

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