Disrupted $Ca_V1.2$ Selectivity Causes Overlapping Long QT and Brugada Syndrome Phenotypes in *CACNA1C*-E1115K iPS Cell Model

Short title: Pathological mechanisms in *CACNA1C*-E1115K iPS cell model

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ABSTRACT

Background

A missense mutation, in the α1c-subunit of voltage-gated L-type Ca^{2+} channel (LTCC)-coding *CACNA1C*-E1115K, located in the Ca^{2+} selectivity site, causes a variety of arrhythmogenic phenotypes.

Objective

We aimed to investigate the electrophysiological features and pathophysiological mechanisms of *CACNA1C*-E1115K in patient-specific induced pluripotent stem cell (iPSC)-derived cardiomyocytes (CMs).

Methods

We generated iPSCs from a patient carrying heterozygous *CACNA1C*-E1115K with overlapping phenotypes of long QT syndrome, Brugada syndrome, and mild cardiac dysfunction. Electrophysiological properties were investigated utilizing iPSC-CMs. We used iPSCs from a healthy subject and an isogenic iPSC line corrected using CRISPR-Cas9-mediated gene editing as controls. The mathematical E1115K-CM model was developed using a human ventricular cell model.

Results

Patch-clamp analysis revealed that E1115K-iPSC-CMs exhibited reduced peak Ca^{2+} current density and impaired Ca^{2+} selectivity, with an increased permeability to monovalent cations. Consequently, E1115K-iPSC-CMs showed decreased action potential plateau amplitude ($APA_{plateau}$), longer action

potential duration (APD), and a higher frequency of early afterdepolarization compared with controls. In optical recordings examining the anti-arrhythmic drug effect, late Na^+ channel current (I_{NaL}) inhibitors (mexiletine, GS-458967) shortened APDs specifically in E1115K-iPSC-CMs. AP-clamp using a voltage command obtained from E1115K-iPSC-CMs with lower $APA_{plateau}$ and longer APD confirmed the upregulation of I_{NaL}. An *in silico* study recapitulated the *in vitro* electrophysiological properties.

Conclusions

Our iPSC-based analysis in *CACNA1C*-E1115K with disrupted Cav1.2 selectivity demonstrated that the aberrant currents through the mutant channels carried by monovalent cations resulted in specific action potential changes, which increased endogenous I_{NaL}; thereby synergistically contributing to the arrhythmogenic phenotype.

Keywords:

L-type Ca^{2+} channel; induced pluripotent stem cell; late Na^+ channel current; long QT syndrome; Brugada syndrome; gene editing; arrhythmia; ion selectivity

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