

MURC 2019

Presentation ID: 257

Presentation Format: Poster Presentation

Presentation Title: Mutagenesis, Molecular Cloning and Transformation Efficiency of the Hyperpolarization-activated Cyclic Nucleotide-gated (HCN) channel

The Hyperpolarization-activated Cyclic Nucleotide-gated (HCN) is a membrane protein that is expressed by excitable cells such as those found in the central nervous system as well as autorhythmic cardiac cells of the sinoatrial node (SAN). These voltage dependent channels exhibit a unique trait as the inward flow of Na⁺ and the outward flow of K⁺ directs the membrane potential towards the action potential threshold which, in turn, activates Ca²⁺ channels and the firing of the next action potential. This study consisted of a site directed mutagenesis where a single mutation was made from serine to phenylalanine on the 100th amino acid residue of the human HCN1 (hHCN1) channel via Polymerase Chain Reaction (PCR). Subsequently, the plasmid with the mutant hHCN1 was transformed into XL10-Gold cells and plated to facilitate bacterial growth. The plasmid was purified through a Mini-prep and Sanger sequencing was done to confirm the mutation. This mutation has been documented as a genetic characteristic of individuals with epilepsy. The excitation of neurons is fundamentally managed by HCN channels, therefore mutations may result in abnormal protein structure or bioelectric activity. In future investigations, the wildtype and mutant channel will be tagged with GFP as a fluorescent protein and then expressed in several mammalian cell lines such as Xenopus oocytes and Chinese hamster ovary cells to determine the transfection efficiency and protein localization by confocal microscopic imaging. The effects of this mutation on protein structure will also be investigated through computer modelling and cryoEM.

Themes:

Check (highlight) the most applicable theme according to the abstract.

<input type="checkbox"/> Innovation and Technology	<input checked="" type="checkbox"/> Health and Wellness	<input type="checkbox"/> Culture and Society	<input type="checkbox"/> Sustainability and Conservation
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Comments:

As MURC is a generalist conference, I would recommend using less jargon. Your current abstract focuses on your methods but I would suggest including a brief background or reason for conducting your research before diving into the methods. What is the significance of your research?