Structure – function relationship of the calcium channel TRPV5

Scientific context

Ion channels are involved in many cellular processes and defects in ion channel functioning are implicated in the pathogenesis of various disorders (together termed 'channelopathies'). The transient receptor potential (TRP) family is one of the largest classes of ion channels that are widely expressed from yeast to human. Based on the amino acid sequence homology, the TRP family is further subdivided and this project will focus on a specific member of the TRPV (vanilloid) subfamily – TRPV5. Within the TRP family, TRPV5 exhibits a unique calcium (Ca^{2+})-selectivity (>100 times over monovalent cations) and it forms the apical gate for transepithelial Ca^{2+} reabsorption in the kidney. It is therefore regarded as the gatekeeper of the body's Ca^{2+} balance.

Project background

A structural map of TRPV5 would provide new insights into the unique Ca^{2+}-selective permeability of the channel and shed new light on the mechanisms that induce the conformational changes needed for opening and closing of the channel pore. A better understanding of the inner workings of TRPV5 will serve to further our understanding of TRPV5-mediated Ca^{2+} transport in the kidney. Recently, single-particle cryo-electon microscopy (cryo-EM) has revitalized the field of structural biology, providing new opportunities to determine the structures of membranous proteins to near-atomic resolution. Using this technique, the first high-resolution 3D structure of a TRP channel (TRPV1, at 3.4 ångström) was published. We hypothesize that the structure of TRPV5 may be resolved using the same technique in order to provide detailed understanding in the molecular structure, gating mechanisms and activation mechanisms of TRP channels.

Aims and Research Questions

In collaboration with Prof. Yifan Cheng at the University of California, San Francisco (UCSF), we have elucidated the 3D protein structure of TRPV5 by single-particle cryo-EM. Such a 3D-structure map offers new possibilities to study the structure-function relationship of the ion channel. Within this project we will address the following topics:

- Identification of key amino acids in the pore region that are involved in ion channel gating
- What signals the structural rearrangement of the channel pore?
- Optimization of the TRPV5 protein expression and purification
- Interaction of TRPV5 with the Ca^{2+}-sensing protein calmodulin

What will you do?

We offer the possibility to perform exciting research in a professional, multicultural and highly motivating working environment with about 30 colleagues in a well-equipped department. You will be part of the Ca^{2+} research team in which you will be responsible for your own research question. Under the supervision of a postdoctoral researcher, you will learn a broad range of techniques, such as:

- molecular cloning
- cell culture
- western blotting
- fluorescence microscopy
- protein expression and purification
- electrophysiological analysis

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