Modulation of TRP channels by temperature in planar lipid bilayers

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Thermal Transient Receptor Potential (TRP) channels belong to the large family of TRP channels. They are an important class of receptors found widely distributed throughout the mammalian central and peripheral nervous systems. They have been shown to be directly activated by heat or cold in physiologically relevant temperature ranges, but are also activated by mechano-stimulation and various ligands. Understanding the mechanisms of temperature activation could lead to the discovery of novel compounds with differing effects on ligand activation and temperature activation for the treatment of pain and other disease states, with fewer side effects.

We have employed parallel planar lipid bilayer instrumentation (Orbit mini, Nanion) to study different reconstituted thermo-TRP channels, particularly the purified human TRP-A1, -V1, -V3 and -M8 channels. Planar lipid bilayers can be formed in the Orbit family systems by painting lipids in organic solvents over Micro Electrode Cavity Array (MECA) chips, a 2 by 2 array of circular micro-cavities in a highly inert polymer. The reconstitution of TRP channels is achieved by adding the purified proteins directly to the bilayers.

In this study, we demonstrate the activation of the different TRP channels by cold and heat in a fully controlled environment using artificial membranes together with a Peltier element integrated in the Orbit mini setup to actively cool and heat the system to the desired temperatures (±1˚C). Furthermore, we compared our Q10 data from purified proteins to TRP channels expressed in HEK cells (Millipore, Chantest) using an automated patch clamp platform (Patchliner, Nanion) with temperature control.

Our results support the hypothesis that temperature sensitivity is located within the channels and does not require any second messengers, including calcium or accessory proteins.