

Title: A novel phosphatidylinositol-4,5-bisphosphates binding site on the N-terminal region of the Melastatin receptor

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Transient receptor potential melastatin 1 (TRPM1) channel belongs to the superfamily of ion channels that respond to various physiological stimuli like chemosensation, thermosensation and mechanosensation. TRP channels have six transmembrane domains with a pore region between the fifth and the sixth segment. Cytosolic N-/C-tails are responsible for regulation of TRPs, which carry binding sites for signal molecules. TRPM1 channel is ubiquitously expressed in most eukaryotic cells and is involved in many cellular processes like transduction of sensory signals and regulation of Ca²⁺ and Mg²⁺ homeostasis. Mutations in TRPM1 gene effect on cell cycle in human skin cells. It seems that a loss of TRPM1 in human melanocytes correlates with increased aggressiveness in melanoma and the homozygous loss of TRPM1 in bipolar cells in retina could be a possible explanation for congenital stationary night blindness in humans.

We studied possible interactions between phosphatidylinositol-4,5-bisphosphate (PIP2) and the N-terminus (NT) of TRPM1. Using bioinformatic approach we identified PIP2 binding site in A451-N566 region. This domain contains several basic amino acids which can interact with anionic phospholipids. Alanine substitution mutagenesis screening revealed the crucial amino acids for these interactions. The equilibrium dissociation constants were estimated using surface plasmon resonance measurement. We identified PIP2-binding site and found mutations that decreased the affinity of TRPM1-NT/PIP2 interaction. Based on this results we concluded that basic residues play crucial role in TRP channels binding to PIP2. Moreover, we have provided the structural insight to TRPM1-NT/PIP2 interaction using computer ligand docking.

Supported by Grants GAUK 238214 and GACR P304/12/G069.

Biography

Michaela Jirku has graduated in 2011 at the University of Chemistry and Technology, Prague, field of clinical biochemistry. Now she has studied PhD in biochemistry for 4 years. She is a co-author of 5 papers in reputed journals.