Focus of the PhD-Project (Stephan Philipp / Adolfo Cavalié)

Modulation of TRPM3 ion channels by accessory subunits

TRPM3 proteins build Ca^{2+} permeable cation channels [1,3] activated by steroids [4,5,7] and sensitive to increased temperatures [8]. TRPM3 channels are expressed in pancreatic ß-cells as well as neurons of the dorsal root ganglion, where they act as mediators of insulin release [4] or nociceptors of noxious heat, respectively [8]. In addition TRPM3 channels are also expressed in brain, eye, pituitary gland and kidney, where they probably play different roles. The functional diversity of TRPM3 channels is enabled by a variety of TRPM3 isoformes which arise by alternative splicing and which display remarkably different biophysical properties [1,2]. Furthermore modulation of channel function may occur by accessory subunits. So far we found that TRPM3 binds TRPM1 but not other members of the TRPM family of ion channels [7]. It also appears that in the brain TRPM3 proteins reside within the nano-environment of voltage gated calcium channel proteins (VGCC) such as the ß subunits [6].

The focus of the project is the characterization of TRPM3 channels and their modulation by interacting proteins in cell culture systems (HEK, MDCK2, Ins1) as well as primary cells obtained from mice. As controls, cells of the same kind but from TRPM3 knock-out mice are available as well as HOM(e) made antibodies directed against channel proteins including TRPM3.

Methods:

PCR, qPCR; Northern,- Westernblot, reportergene assay; Ca^{2+}-Imaging; fluorescence associated cell sorting (FACS); immunoprecipitation; pull-down assay; peptide spot assays; transgenic mice, yeast-two-hybrids screens.

Selected publications:

1. Oberwinkler,J., Lis,A., Giehl,K.M., Flockerzi,V., Philipp,S.E.. (2005) Alternative Splicing Switches the Divalent Cation Selectivity of TRPM3 Channels. J. Biol. Chem. 280, 22540-22548.
2. Lis,A., Wissenbach,U, Philipp,S.E. (2005) Transcriptional regulation and processing increase the functional variability of TRPM channels. Naunyn-Schmiedeberg´s Arch.Pharmacol. 371, 315-324.

3. Oberwinkler,J., Philipp,S.E. (2007) TRPM3. In Flockerzi,V. and Nilius B (eds): Handbook of Experimental Pharmacology, Vol. 179 Transient Receptor Potential (TRP) Channels. Springer, Heidelberg.
4. Wagner,T.F.J., Loch,S. Lambert,S., Straub,I., Mannebach,S., Düfer,M., Lis,A. Flockerzi,V., Philipp,S.E., Oberwinkler,J. (2008) Transient receptor potential M3 channels are ionotropic steroid receptors in pancreatic ß-cells. Nat. Cell Biol. 12, 1421-1430.
5. Wagner,T.F., Drews,A., Loch,S., Mohr,F., Philipp,S.E., Lambert,S., Oberwinkler,J. (2010) TRPM3 channels provide a regulated influx pathway for zinc in pancreatic beta cells. Pflügers Arch. Eur. J. Physiol. 460, 755-765.
6. Muller,C.S., Haupt,A., Bildl,W., Schindler,J., Knaus,H,G., Meissner,M., Rammner,B., Striessnig,J., Flockerzi,V., Fakler,B., Schulte,U. (2010) Quantitative proteomics of the Cav2 channel nano-environments in the mammalian brain. Proc Natl Acad Sci U S A, 107, 14950-14957.
7. Lambert,S., Drews,A., Rizun,O., Wagner,T.F., Lis,A., Mannebach,S. Plant,S., Portz,M., Meissner,M., Philipp,S.E., Oberwinkler,J. (2011) Transient receptor potential melastatin 1 (TRPM1) is an ion conducting plasma membrane channel inhibited by zinc ions. J. Biol. Chem. 286, 12221-12233.
8. Vriens,J., Owsianik,G., Hofmann,T., Philipp,S.E., Stab,J., Chen,X., Benoit,M., Xue,F., Janssens,A., Kerselaers,S., Oberwinkler,J., Vennekens,R., Gudermann,T., Nilius,B., Voets,T. (2011) TRPM3 is a nociceptor channel involved in the detection of noxious heat. Neuron 70, 1-13.