Scharinger et al. BMC Pharmacology and Toxicology 2012, 13(Suppl 1):A50
http://www.biomedcentral.com/2050-6511/13/S1/A50

BMC Pharmacology & Toxicology

MEETING ABSTRACT Open Access

A mouse model to study the C-terminal regulation of $Ca_V1.3$ L-type calcium channels

Anja Scharinger[1], Florian Hechenblaikner[1], Gabriella Bock[1], Mathias Gebhart[1], Kai Schönig[2], Dusan Bartsch[2], Anupam Sah[1], Nicolas Singewald[1], Martina J Sinnegger-Brauns[1], Jörg Striessnig[1*]

From 18th Scientific Symposium of the Austrian Pharmacological Society (APHAR). Joint meeting with the Croatian, Serbian and Slovenian Pharmacological Societies.
Graz, Austria. 20-21 September 2012

Background

$Ca_V1.3$ voltage-gated L-type Ca^{2+} channels (LTCCs) play an important role for hearing, cardiac pacemaking and neuronal excitability. The C-terminus of $Ca_V1.3$ tightly controls channel gating by an intramolecular protein interaction involving two putative α-helices (termed PCRD, DCRD), which form a C-terminal modulatory mechanism (CTM) only in full-length $Ca_V1.3$ variants. In short ($Ca_V1.3_{42A}$ and $Ca_V1.3_{43S}$) $Ca_V1.3$ α1 subunit splice variants CTM is absent which leads to profound changes in channel gating: activation occurs at more negative voltages and Ca^{2+}-dependent inactivation (CDI) is faster.

Methods

We quantified $Ca_V1.3$ splice variants by qPCR analysis and transcript scanning, using different mouse tissues. To assess the physiological relevance of CTM, we generated a mutant mouse strain in which CTM function is disrupted by an HA-tag ($Ca_V1.3$-DCRD$^{HA/HA}$ mice). We used anti-HA antibodies to detect the expression of the HA-tagged full length channel by Western blot analysis.

Results

The short variants $Ca_V1.3_{42A}$ (highest relative abundance in substantia nigra (SN) and ventral tegmental area (VTA)) and $Ca_V1.3_{43S}$ are both less abundant in mouse brain indicating that the full length form $Ca_V1.3_L$ comprises the most abundant form (about 50% of all transcripts). In mouse heart short transcripts are rare and $Ca_V1.3_L$ represents about 90% of all known transcripts. $Ca_V1.3$-DCRD$^{HA/HA}$ mice contain a homozygous interruption of the CTM by disrupting the DCRD helix with an HA-tag. We show that this induces "short" gating properties in this mutant full-length variant. Homozygous mice are viable and display no gross anatomical and functional abnormalities. Expression of the HA-tagged full-length channel could be detected in mouse whole brain membrane preparations. Heterozygous mice show no overt differences in locomotor activity during daytime.

Conclusions

We have successfully generated a mouse model which will enable us to study the physiological role of CTM function *in vivo*. It mimics the (permanent) pharmacological inhibition of CTM function and will thus allow predictions about its potential as a new drug target. Furthermore, the HA-tagged α1 subunit will provide a tool to specifically determine the expression of $Ca_V1.3_L$ channels with anti-HA antibodies in mouse tissues.

Acknowledgements
Supported by the Austrian Science Fund project SFB f44, the EC project MRTN-CT-2006-35367 ("CavNet"), FWF 20670 and the University of Innsbruck.

Author details
[1]Department of Pharmacology and Toxicology, Institute of Pharmacy, and Center for Molecular Biosciences Innsbruck, University of Innsbruck, 6020 Innsbuck, Austria. [2]Department of Molecular Biology, Central Institute of Mental Health, University of Heidelberg, 68159 Mannheim, Germany.

Published: 17 September 2012

* Correspondence: joerg.striessnig@uibk.ac.at
[1]Department of Pharmacology and Toxicology, Institute of Pharmacy, and Center for Molecular Biosciences Innsbruck, University of Innsbruck, 6020 Innsbuck, Austria
Full list of author information is available at the end of the article

doi:10.1186/2050-6511-13-S1-A50
Cite this article as: Scharinger *et al.*: A mouse model to study the C-terminal regulation of $Ca_V1.3$ L-type calcium channels. *BMC Pharmacology and Toxicology* 2012 **13**(Suppl 1):A50.