Bernadett Bacsa

Gottfried Schatz Research Center (Medical Physics and Biophysics), Medical University of Graz

Project Title:

The role of TRPC6 channels in allergic airway inflammation

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Transient Receptor Potential Canonical (TRPC) channels (TRPC1-7), are nonselective Ca^{2+} permeable cation channels. While primarily studied in cardiovascular and nervous systems, increasing evidence suggests that these channels are important modulators of homeostatic and pathological immune responses. Among TRPC channels, TRPC6 particularly is highly expressed in human and mouse lungs and plays an important role in airway inflammation[1]. Our recent data show that TRPC3/6- signaling pathways can be activated in immune cells using TRPC3/6-specific pharmacological tools[2] and that TRPC6 is highly expressed in human eosinophils.

Hypothesis and Objectives:

We hypothesize that TRPC6 channels have (patho)physiological roles in eosinophilic airway inflammation. In this project, we will investigate the role of TRPC6 channels in peripheral blood leukocytes from allergic and non-allergic asthmatics as compared to healthy controls. We will further evaluate the value of the TRPC6 channel as a therapeutic target in an *in vivo* mouse model of house dust mite-induced allergic lung inflammation.

Methodology:

Responses of eosinophils to TRPC6-specific modulators will be investigated using functional assays, including Ca^{2+} influx, chemotaxis, shape change, and apoptosis. The cellular expression of TRPC6 will be analyzed in human and murine lung tissue using *in situ* hybridization and immunofluorescence. In a translational approach, a well-established house dust mite–induced mouse model[3] will be used to assess the role of TRPC6 in disease pathogenesis through lung function testing and flow-cytometric immunophenotyping of lung tissue and bronchoalveolar lavage fluid (BALF). Cytokines and lipid mediators in serum, lung tissue, and BALF will be quantified by ELISA and mass spectrometry. In parallel, TRPC6 expression and functional activity will be compared in eosinophils from asthmatic patients and healthy controls. Finally, the cellular localization of TRPC6 and its downstream signaling pathways will be examined using Ca^{2+} imaging and immunofluorescence, employing novel TRPC3/6 photopharmacological inhibitors in murine bone marrow–derived and human eosinophils.

References:

1. R. L. Corteling et al., Am J Resp Cell Mol 30, 145-154 (2004)
2. B. Bacsa et al., Front Immun 11, 613194 (2020)
3. Teppan et al., Front Immun 15, 1408772 (2024)

Thomas Baernthaler

Otto Loewi Research Center (Pharmacology), Medical University of Graz

Project Title:

TRPC1 in idiopathic pulmonary fibrosis

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Idiopathic pulmonary fibrosis (IPF) is a chronic progressive disease with a median survival of only 3-4 years, significantly shorter than many types of cancer. It is characterized by exaggerated extracellular matrix (ECM) deposition in the lungs, loss of alveolar tissue and disrupted gas exchange. Currently, there are only two therapeutic options for the treatment of IPF, pirfenidone and nintedanib. While these drugs slow disease progression and might prolong survival, they are neither curative, nor do they entirely stop loss of lung function. Furthermore, a significant proportion of patients discontinue anti-fibrotic therapy due to side effects. Therefore, other therapeutic options are urgently needed.
Transient Receptor Potential Canonical Channel Isoform (TRPC)1 is a non specifc ion channel located on plasma mebranes. TRPC1 has been shown to mediate myofibroblast transdifferetiation and fibroblast proliferation, two hallmarks of pulmonary fibrosis. We found that TRPC1 shows highest expression of all TRP channels in fibroblasts from IPF patients.

Hypothesis and Objectives:

Due to our preliminary data, we know that TRPC1 shows highest expression of all TRP channels in fibroblasts from IPF patients and thus hypothesize that it may be a novel therapeutic target in IPF. We want to assess whether (I) TRPC1 modulation has therapeutic potential in PF of different etiologies (II) which mechanisms and cells mediate these effects and (III) whether TRPC1 modulation is able to revert fibrosis.

Methodology:

We will use bleomycin and TGF-beta transgenic mouse models of PF to test effects of TRPC1 modulation and KD and assess fibrosis using hydroxyproline test, Ashcroft scoring, and lung function test. Fibroblasts and alveolar epithelial cells will be used for cell culture experiments, followed by ELISA, qPCR and WB to detect DES effects on for example ECM production, lipofibroblast differentiation or survival associated pathways. We will investigate proliferation/apoptosis via EDU/Ki67 and AnnexinV/PI/activated caspase3 staining respectively, cell counts and caspase 3/7 activity. Precision cut lung slices (PCLS) from PF patients of different etiology and healthy controls will be treated with DES. Readouts will include safety and pharmacokinetics (mouse models), respective pathways/affected cells (RNASeq, sc/sn-RNASeq) including validation and morphometry

(microscopy/immunofluorescence).

References:

Rajesh, R; Mooslechner, AA; Schweighofer, H; Pahernik, S; Lanz, I; Atallah, R; Platzer, W; Aigner, C; Benazzo, A; Angiari, S; Marsh, L; Kwapiszewska, G; Heinemann, A; Bärnthaler, T; Succinate aggravates pulmonary fibrosis through the succinate/SUCNR1 axis. Am J Physiol Lung Cell Mol Physiol. 2025;

Shuni Li, Lihui Qu, Lifen Zhou, Na Zhan, Linmei Liu, Yuquan Ling, Qingzi Chen, Wuping Lai, Nan Lin, Jianhua Li,; Biomass fuels related-PM2.5 promotes lung fibroblast-myofibroblast transition through PI3K/AKT/TRPC1 pathway, Ecotoxicology and Environmental Safety,2024,

Kenichi Ikeda, Toshiaki Nakajima, Yumiko Yamamoto, Nami Takano, Tomofumi Tanaka, Hironobu Kikuchi, Gaku Oguri, Toshihiro Morita, Fumitaka Nakamura, Issei Komuro, Roles of transient receptor potential canonical (TRPC) channels and reverse-mode Na+/Ca2+ exchanger on cell proliferation in human cardiac fibroblasts: Effects of transforming growth factor β1,; Cell Calcium, 2013,

Rajesh, R; Atallah, R; Bärnthaler; Dysregulation of metabolic pathways in pulmonary fibrosis T, Pharmacology and Therapeutics 2023; 246: 108436

Sanja Curcic

Gottfried Schatz Research Center (Medical Physics and Biophysics), Medical University of Graz

Project Title:
Immune cell-specific targeting of TRPC3/6 channels in psoriasis

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Transient Receptor Potential Canonical (TRPC) channels are non-selective Ca^{2+}-permeable cation channels that play key roles in cellular Ca^{2+} signaling. While TRPC channels are best known for their functions in the cardiovascular and nervous systems, increasing evidence indicates that they also regulate immune cell activation, differentiation, and inflammatory signaling[1]. Psoriasis is a chronic immune-mediated inflammatory skin disease driven by complex interactions between innate and adaptive immune cells, including macrophages, T cells, B cells, dendritic cells, and neutrophils. Although altered expression of TRPC6 has been reported in psoriatic lesions[2], the functional relevance of TRPC3 and TRPC6 channels in immune cell regulation during psoriatic inflammation remains poorly understood. So far, only limited evidence suggests that pharmacological activation of TRPC6, such as with hyperforin[3], has beneficial effects; however, the underlying mechanisms remain poorly understood.

Hypothesis and Objectives:

We hypothesize that TRPC3 and TRPC6 channels critically regulate immune cell-specific Ca^{2+} signaling pathways that shape the development and severity of psoriatic inflammation. The objective of this project is to define the role of TRPC3/6-dependent Ca^{2+} signaling in immune cell-driven psoriasis and to identify the immune cell populations most strongly affected by modulation of these channels. Furthermore, we aim to determine whether targeted pharmacological manipulation of TRPC3/6 represents a viable therapeutic strategy for controlling psoriatic disease progression.

Methodology:

The PhD student will investigate the contribution of TRPC3 and TRPC6 channels to psoriatic inflammation using genetic mouse models and established pharmacological modulators. Preliminary data from our group already indicate altered immune cell composition in the spleen of TRPC3/6-deficient mice, with pronounced changes in macrophage, T cell, and B cell populations, supporting a systemic role of TRPC3/6 in immune regulation.

Psoriatic inflammation will be induced using the imiquimod (IMQ)-induced mouse model, a well-established *in vivo* model that mimics key pathological features of human psoriasis. Disease severity

will be assessed using a PASI-like scoring system based on erythema, scaling, and skin thickening, complemented by histological analysis of epidermal thickness, hyperkeratosis, and epidermal differentiation.

Infiltrating immune cell populations in the skin—including macrophages, T cells, B cells, antigen-presenting cells, plasmacytoid dendritic cells, and neutrophils—will be characterized by immunohistochemistry and flow cytometry. Expression of inflammatory mediators, particularly components of the IL-23/IL-17 axis, will be quantified by qPCR in skin tissue. In parallel, immune cell composition and activation in spleen and lymph nodes will be analyzed to assess systemic immune alterations.

Based on these findings, immune cell subsets most strongly regulated by TRPC3/6 will be identified and subjected to targeted functional analyses, including Ca^{2+} influx measurements, activation marker expression, proliferation, and cytokine production. Finally, the therapeutic potential of established TRPC3/6 activators and inhibitors will be evaluated *in vivo* to assess their impact on psoriatic inflammation.

References:

1. Wenning AS et al. (2011) TRP expression pattern and the functional importance of TRPC3 in primary human T-cells Biochim Biophys Acta Mol Cell Res 1813(3): 412–423

2. Leuner K et al. (2011) Reduced TRPC channel expression in psoriatic keratinocytes is associated with impaired differentiation and enhanced proliferation PLoS One 6(2): e14716

3. Zhang S et al. (2021) Hyperforin ameliorates imiquimod-induced psoriasis-like murine skin inflammation by modulating IL-17A-producing γδ T cells Front Immunol 12: 635076

Benjamin Gottschalk

Gottfried Schatz Research Center (Molecular Biology and Biochemistry), Medical University of Graz

Project Title:

TRPML1 in Lysosome-Mitochondria Crosstalk during Cancer Energy Stress

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Cells adapt to metabolic stress by reorganizing their organelles, and some of the most critical decisions happen at nanoscale contact sites between mitochondria and lysosomes. These contact sites act as highly dynamic signaling hubs that coordinate ion fluxes, energy production, and organelle quality control. In cancer cells, where energy stress is common, understanding how these interactions are regulated at the level of individual organelles and even single molecules is a major open challenge. Our recent work shows that mitochondrial cristae junctions are not static structures: they can open to allow proton leakage, reshaping local membrane potential and metabolic output[1,2]. This process is tightly regulated by mitochondrial Ca^{2+} uptake through the MCU complex and its gatekeeper MICU1[3]. Intriguingly, TRPML1, a lysosomal Ca^{2+} channel best known for maintaining lysosomal ion homeostasis, appears to directly influence mitochondrial Ca^{2+} signaling specifically at lysosome–mitochondria contact sites[4]. How TRPML1-mediated ion fluxes reshape mitochondrial ultrastructure and function in stressed cancer cells is largely unknown and ideally suited for investigation using single-cell and super-resolution microscopy.

Hypothesis and Objectives:

We hypothesize that metabolic stress enhances lysosome–mitochondria coupling in cancer cells, and that TRPML1-dependent ion signaling at these contact sites actively remodels mitochondrial structure, membrane potential gradients, and bioenergetic performance. These processes are expected to be highly heterogeneous and dynamic at the single-cell and sub-organelle level.

Methodology:

During the first phase of the project, the student will establish live-cell imaging approaches using genetically encoded biosensors for Ca^{2+}, pH, K^{+}, and reactive oxygen species targeted to lysosomes and defined mitochondrial subcompartments. By combining these tools with pharmacological activation of TRPML1, the student will characterize how ion fluxes and metabolic stress reshape lysosome–mitochondria communication in single cancer cells.

In the second phase, the project will leverage super-resolution and ultrafast confocal microscopy in living cells to investigate how TRPML1 regulates lysosomal acidification, mitophagy, and the dynamics

of mitochondrial cristae at contact sites. Structured illumination microscopy[1-3] will be used to resolve spatial gradients in mitochondrial membrane potential, allowing functional readouts to be directly linked to nanoscale organelle organization.

In the final phase, confocal super-resolution PAINT microscopy and single-molecule tracking will be established to probe mitochondrial ultrastructure with nanometer precision. This will enable a detailed analysis of local cristae organization and reveal inter- and intra-lysosomal heterogeneity in TRPML1 distribution, with a particular focus on mitochondria–lysosome interfaces.

Overall, this project offers an exciting opportunity for a student interested in single-cell biology, organelle dynamics, and cutting-edge microscopy to study how nanoscale ion signaling and organelle architecture jointly control cancer cell metabolism under energy stress.

References:

[1] Gottschalk, B; et al. 2024. Sci Rep 14, 14784 (2024). https://doi.org/10.1038/s41598-024-65595-z

[2] Gottschalk, B; et al. 2022. Commun Biol.; 5(1): 649 Doi: 10.1038/s42003-022-03606-3.

[3] Gottschalk, B; et al. 2019. Nat Commun.; 10(1):3732-3732 Doi: 10.1038/s41467-019-11692-x

[4] Burgstaller, et al. 2019. ACS Sens.; 4(4): 883-891. Doi: 10.1021/acssensors.8b01599

Lukas Groschner

Gottfried Schatz Research Center (Molecular Biology and Biochemistry), Medical University of Graz

Project Title:

TRP channels in cerebellum-like circuits

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

The *Trpc3* gene is associated with hereditary forms of ataxia. Mice with a gain-of-function mutation in this gene, which encodes the transient receptor potential channel TRPC3, feature a striking form of ataxia (1). Since the first report of the moonwalker phenotype, these mice have been teetering in circles. Sixteen years later, the pathophysiology of this channelopathy remains a mystery. Microscopic evidence hints at a developmental defect affecting the dendrites of cerebellar Purkinje cells (1), but the signaling pathways downstream of TRPC3 that control dendritogenesis are unknown. We propose to investigate this problem in *Drosophila melanogaster*, where developmental, structural, and functional studies can be carried out with ease. The *Drosophila* mushroom body bears remarkable similarity to the vertebrate cerebellum (2): Kenyon cell axons resemble the parallel fibers of granule cells; in place of Purkinje cells, they form synapses with mushroom body output neurons (MBONs). Like their mammalian counterparts, MBONs have elaborate dendrites and express *trp*, the eponymous homologue of the mouse *Trpc3* gene (3). The aim of the proposed project is to pinpoint the mechanisms by which TRP channels shape the growth and function of neurons.

Hypothesis and Objectives:

1) The Drosophila *trp* gene influences the morphology and excitability of MBONs. Our objective is to interfere with the expression and function of *trp* in MBONs at distinct developmental stages and assess neuronal structure and function.

2) TRP channels localize to distinct subcellular domains. The student will label TRP channels using an endogenous tag and visualize their subcellular distribution throughout development.

3) TRP channel activity changes over developmental stages and starts intracellular signalling cascades. Transcriptional, morphological and functional changes in response to acute TRP channel perturbation will be recorded using patch-seq experiments.

Methodology:

The student will use RNA interference and photopharmacology to control TRP channels in genetically defined MBONs and quantify neuronal structure and function using confocal microscopy and patch clamp electrophysiology *in vivo*. The generation of transgenic *Drosophila* lines will allow for the

recombinase-dependent labelling of endogenous TRP channels in defined cell types. Tagged channels will be visualized during dendritogenesis using super-resolution microscopy. The student is expected to combine the acquired skills with the transcriptomic profiling of single cells to identify TRP-dependent signaling pathways. This project will provide training in *Drosophila* genetics, confocal and super-resolution microscopy, molecular biology, and *in vivo* electrophysiology

References:

1. Becker *et al*. (2009) A point mutation in TRPC3 causes abnormal Purkinje cell development and cerebellar ataxia in moonwalker mice. *Proc Natl Acad Sci U S A* 106(16): 6706–11.
2. Groschner & Miesenböck (2019) Mechanisms of sensory discrimination: Insights from *Drosophila* olfaction. *Annu Rev Biophys* 48(1):209–29.
3. Davis *et al*. (2020) A genetic, genomic, and computational resource for exploring neural circuit function. *eLife* 9:e50901.

Jordi Heijman

Gottfried Schatz Research Center (Medical Physics and Biophysics), Medical University of Graz

Project Title:

Computational modeling of TRPC channels in the heart

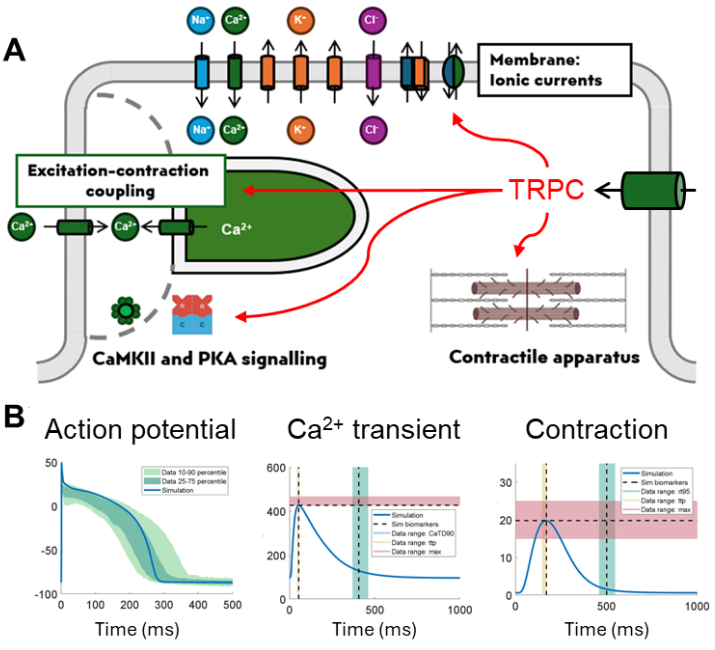
*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Cardiac remodelling following a pathologic insult to the heart, plays a central role in cardiovascular disease, with subsequent development of cardiac arrhythmias as a major mediator of morbidity and mortality. Emerging evidence suggests an important role for transient receptor potential (TRP) channels, in particular the TRPC subfamily, in both atrial and ventricular proarrhythmic remodeling. However, a quantitative understanding of the downstream signaling pathways and their relative contribution to arrhythmogenesis is lacking.

Hypothesis and Objectives:

Mechanistic computational models of cardiac electrophysiology help to integrate experimental data and enable the identification of nodal points that may constitute promising therapeutic targets. However, none of the currently available models integrate TRPC channels and their downstream effects on Ca^{2+} handling.[1] We aim to develop novel state-of-the-art computational tools (freely available in open-source format) to simulate the effects of TRPC-channel remodeling and their modulation by photo-pharmacological tools, and to use these models to identify novel therapeutic targets.



Methodology:

The PhD student will be trained in computational modeling of cardiac cellular electrophysiology and will develop Markov-model formulations of TRPC channel gating with/without different pharmacological modulators based on existing and newly collected experimental data. The TRPC ion-channel models will be integrated into detailed models of the human atrial

and ventricular cardiomyocyte recently developed by the PI (Figure).[2] Models will be calibrated based on cardiomyocyte contractility and Ca^{2+}-cycling data obtained by live-cell imaging as part of the TRPC.at project (Holzer) and used to assess the contribution of TRPC to cellular arrhythmogenesis at baseline and in the presence of disease-related TRPC remodeling. The same approach will be used to develop a novel model of the human cardiac fibroblast, integrating key fibroblast ion channels and Ca^{2+}-handling properties based on published data from long-term collaborators of the PI.[3] Alterations in fibroblast Ca^{2+}-handling will be considered a key indicator of the pro- or anti-fibrotic consequences of TRPC-channel modulation. Sensitivity analyses employing large populations of virtual fibroblasts and cardiomyocytes with variations in model parameters will identify the parameters associated with proarrhythmic phenotypes with or without TRPC remodeling. Together, these analyses will provide novel quantitative insights into the role of TRPC in proarrhythmic cardiac remodeling.

References:

1. Trayanova NA, Lyon A, Shade J, Heijman J (2024). Computational modeling of cardiac electrophysiology and arrhythmogenesis: toward clinical translation. Physiol Rev.;104(3):1265-1333. doi: 10.1152/physrev.00017.2023.
2. Heijman J et al. (2023) Enhanced Ca2+-Dependent SK-Channel Gating and Membrane Trafficking in Human Atrial Fibrillation. Circ Res;132(9):e116-e133. doi: 10.1161/CIRCRESAHA.122.321858.
3. Wu CT et al. (2014) Disease and region-related cardiac fibroblast potassium current variations and potential functional significance. Cardiovasc Res.;102(3):487-96. doi: 10.1093/cvr/cvu055

Senka Holzer (PI) / Oleksandra Tiapko (Co-PI)

Department of Internal Medicine, Division of Cardiology and Gottfried Schatz Research Center (Medical Physics and Biophysics), Medical University of Graz

Project Title:

Roles of TRPC3 and TRPC6 channels in cardiac remodeling

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Cardiac remodelling encompasses molecular, cellular and gene expression changes following pathologic insults to the heart, impacting cardiac function and patients' prognosis. In cardiomyocytes, angiotensin II (ATII) signalling directly regulates TRPC3 and TRPC6 channels, and their genetic deletion or pharmacological inhibition protects the heart from ischemia-reperfusion injury and pathological hypertrophy.[1] RNA-seq analysis of heart tissue from mice lacking all TRPCs links TRPC genes to contractility and inflammatory pathways.[2] The central question of how the cardiomyocyte translates altered TRPC-mediated signalling into deleterious genetic programs that leave it dysfunctional remains unresolved.

Hypothesis and Objectives:

We hypothesize that chronic activation of the ATII-TRPC3/6 axis disrupts subcellular Ca^{2+} homeostasis, triggering Ca^{2+}-mediated hypertrophic and inflammatory transcription in cardiomyocytes. To test this, we will 1) assess TRPC3/6 expression and activity in cardiomyocytes following chronic ATII exposure, 2) analyze TRPC3/6 levels in human hypertrophied and failing hearts, 3) investigate ATII-TRPC3/6-mediated dysregulation of contractility and Ca^{2+} cycling across cytoplasmic, nuclear, and mitochondrial compartments, and 4) examine how altered Ca^{2+} signalling impacts hypertrophic and inflammatory pathways, leading to *in vivo* cardiac dysfunction.

Methodology:

The PhD student will assess TRPC3/6 expression and subcellular localization in WT, TRPC hepta-KO, TRPC1/2/4/5/6/7 KO and TRPC1/2/3/4/5/7 KO mice challenged with continuous ATII administration, and in hypertrophic or failing human myocardium by qPCR, western blotting of subcellular fractions and immunocytochemistry. TRPC channels activity will be investigated by patch-clamping using pharmacological modulators (activators: GSK1702934A, BI-2; inhibitors: Pyr10). Functional roles of TRPC3/6 in cardiomyocyte contractility and Ca^{2+} cycling will be assessed by live-cell widefield cellular and confocal subcellular Ca^{2+} imaging of cardiomyocytes isolated from corresponding groups and following same interventions.[3-5] Data on *in vivo* cardiac function of control and ATII-challenged mice

will be assessed by transthoracic echocardiography.[3] The effects of altered ATII-TRPC3/6 axis on cellular transcriptome will be addressed by RNAseq and confirmatory protein analyses of cardiomyocyte isolated from corresponding TRPC mice.

References:

1. Seo K. et al. Combined TRPC3 and TRPC6 blockade by selective small-molecule or genetic deletion inhibits pathological cardiac hypertrophy. Proc Natl Acad Sci USA. (2014).
doi:10.1073/pnas.1308963111.

2. Formoso K. et al. RNA-seq analysis reveals TRPC genes to impact an unexpected number of metabolic and regulatory pathways. Sci Rep. (2020).
doi:10.1038/s41598-020-61177-x.

3. Ljubojevic-Holzer S. et al. CaMKIIdeltaC Drives Early Adaptive Ca^{2+} Change and Late Eccentric Cardiac Hypertrophy. Circ Res. (2020).
doi: 10.1161/CIRCRESAHA.120.316947.

4. Ljubojevic-Holzer S. et al. Loss of autophagy protein ATG5 impairs cardiac capacity in mice and humans through diminishing mitochondrial abundance and disrupting Ca2+ cycling. Cardiovasc Res. (2022).
doi: 10.1093/cvr/cvab112.

5. Voglhuber J. et al. Functional remodelling of perinuclear mitochondria alters nucleoplasmic Ca^{2+} signalling in heart failure. Philos Trans R Soc Lond B Biol Sci. (2022).
doi: 10.1098/rstb.2021.0320.

Corina Madreiter-Sokolowski

Gottfried Schatz Research Center (Molecular Biology and Biochemistry), Medical University of Graz

Project Title:

ROS Signaling and TRP Channel Activity in Aging – From C. elegans to Mammalian Models

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Reactive oxygen species (ROS) are central regulators of cellular signaling but become increasingly dysregulated during aging, contributing to functional decline across tissues. In parallel, transient receptor potential (TRP) channels constitute a highly conserved superfamily of cation channels that control ion homeostasis, excitability, and stress responses. Importantly, several TRP family members can respond to ROS, suggesting that TRP channels may act as redox-sensitive signal hubs that translate oxidative stress into downstream physiological programs. This is particularly relevant in aging, where elevated ROS levels coincide with progressive deterioration of subcellular integrity, including changes in mitochondrial structure, redox balance, and stress resilience.

Hypothesis and Objectives:

We hypothesize that age-associated ROS elevation modifies the activity of redox-sensitive TRP channels, thereby perturbing ion homeostasis and stress-response signaling and ultimately contributing to reduced organismal fitness and longevity. To test this hypothesis, this Ph.D. project will utilize Caenorhabditis elegans (C. elegans) and mammalian cellular aging models to molecularly and functionally characterize ROS-sensitive TRP channels during aging and to identify the downstream signaling cascades that connect ROS-TRP interactions to stress resistance and aging pathways.

Methodology:

The candidate will combine live-cell fluorescence microscopy using organelle-targeted biosensors to define TRP channel activity and its modulation by ROS, complemented by molecular biology approaches to resolve downstream pathway engagement. In C. elegans, these mechanistic insights will be linked to organismal phenotypes, including locomotion, stress resilience, and lifespan. At the same time, key mechanisms will be tested for conservation in mammalian aging models.

References:

Please familiarize yourself with our publications via PubMed Madreiter-Sokolowski.

Roland Malli (PI)/ Benjamin Gottschalk (Co-PI)

Center for Medical Research (Core Facility Bioimaging) and Gottfried Schatz Research Center (Molecular Biology and Biochemistry), Medical University of Graz

Project Title:

Decoding Lysosome–Mitochondria Crosstalk in TRPML1-Mediated Ferroptosis

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Ferroptosis, an iron-dependent form of cell death, has emerged as a central process in cancer, neurodegeneration, and aging. While traditionally viewed as a cytosolic event, recent evidence points to lysosomes as hotspots for iron-driven oxidative damage. These organelles reduce Fe^{3+} to Fe^{2+}, produce hydrogen peroxide, and create a local environment ideal for radical chemistry. The lysosomal channel TRPML1 controls Fe^{2+} and Ca^{2+} release, shaping mitochondrial ion supply and ferroptotic signaling. Yet, exactly how lysosomes and mitochondria communicate during ferroptosis remains a mystery. This project is part of TRPCat, where PhD students explore ion channels from the TRP family to uncover how ion signaling orchestrates cellular pathways. In this project, you will combine cutting-edge bioimaging and biosensor technologies to decipher the hidden dynamics of ferroptosis.

Hypothesis and Objectives:

We hypothesize that TRPML1-mediated release of Fe^{2+} and Ca^{2+} from lysosomes controls ferroptotic signaling through direct lysosome–mitochondria crosstalk. Interrupting this communication may reveal the hidden triggers of iron-dependent cell death and uncover organelle-specific mechanisms that remain largely unexplored. To address this, the project will: *i*) Develop novel biosensors and assays to track Fe^{2+}, Ca^{2+}, and oxidative events at the subcellular level in real time, offering unprecedented insight into ferroptosis dynamics. *ii*) Visualize and dissect lysosome–mitochondria communication, mapping TRPML1 activity and iron flow between organelles during ferroptotic signaling. *iii*) Reveal the mechanisms governing iron-dependent cell death, showing how lysosomal iron handling shapes mitochondrial responses and ferroptotic outcomes. *iv*) Translate these insights into experimental strategies to modulate ferroptosis, providing potential therapeutic avenues and innovative tools for the wider research community.

Methodology:

The project combines advanced molecular biology, live-cell imaging, and biosensor development[1-3]. You will employ high- and super-resolution fluorescence microscopy to capture signalling activities in real time and to visualize cellular organelles and their dynamics. Analysis of lysosome–mitochondria

contacts and TRPML1 activity will be central, enabling a detailed mechanistic understanding of ferroptotic signaling from the subcellular to the molecular level. By integrating these approaches, you will develop novel experimental tools and uncover how lysosomes orchestrate life-or-death decisions in the cell.

References:

[1] Akyol, A; et al *ACS Sens. 2025*; 10(8):5854-5861 Doi: 10.1021/acssensors.5c01165

[2] Pilic J et al *Mol Cell*. 2024; 84(14): 2732-2746.e5. Doi: 10.1016/j.molcel.2024.06.009

[3] Erdoğan, YC; et.al. *ACS Sens*. 2024; 9(9):4680-4689 Doi: 10.1021/acssensors.4c01058

Nagaraj Chandran

Department of Internal Medicine (Division of Respiratory Medicine, Lung Research Cluster), Medical University of Graz

Project Title:

Role of TRPC- BK channel axis in Regulating Calcium Influx and Functional Polarization of Macrophages

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Calcium (Ca^{2+}) signaling plays a central role in orchestrating macrophage polarization between pro-inflammatory (M1) and reparative (M2) phenotypes, especially during lung injury. Sustained Ca^{2+} influx, predominantly mediated by Transient Receptor Potential Canonical (TRPC) channels, is essential for initiating and maintaining M1 polarization through pathways such as STAT1 and NF-κB1. The activity of these Ca^{2+}-permeable channels is tightly regulated by the membrane potential (V_m), which in turn is governed by K^+ conductance. Among potassium channels, the large-conductance Ca^{2+}-activated K^+ (BK) channel has emerged as a crucial modulator of macrophage electrophysiology2. By promoting K^+ efflux, BK channels help maintain a hyperpolarized V_m, thereby limiting the electrochemical drive for TRPC-mediated Ca^{2+} influx. Loss or inhibition of BK channels leads to membrane depolarization, amplifying Ca^{2+} entry via TRPC channels and skewing macrophage polarization toward a sustained pro-inflammatory M1 phenotype.

Hypothesis and Objectives:

We hypothesize that this TRPC–BK channel feedback axis is a key regulator of macrophage Ca^{2+} homeostasis during lung injury. Disruption of BK channel function elevates basal Ca^{2+} levels and blunts stimulus-evoked Ca^{2+} responses, promoting sustained inflammation and tissue damage. This integrated approach will reveal whether a BK–TRPC axis controls macrophage Ca^{2+} homeostasis and polarization in lung injury, and whether targeting this axis can shift macrophage responses toward resolution. This could provide a novel ion-channel-based strategy to treat inflammatory and fibrotic lung disease. The PhD student will investigate the contribution of TRP channels to Ca^{2+} homeostasis and macrophage polarization, and extend these findings to pulmonary vascular cells to explore cross-tissue relevance using photoswitchable TRPC modulators.

Methodology:

Bone marrow–derived macrophages (BMDMs) from both murine models (wild-type and BK knockout) will be analyzed functionally by patch-clamp electrophysiology and live-cell Ca^{2+} imaging to measure V_m and TRPC-mediated Ca^{2+} influx across M0, M1 and M2 states. Pharmacological TRPC inhibitors and

siRNA knockdown will employed and downstream readouts will include polarization markers, transcriptomics, and metabolic profiling. BMDMs from WT or $BK^{-/-}$ mice co-cultured with endothelial cells to measure cytokine transfer, barrier integrity (TEER), and TRPC activity. These studies will determine the feasibility of selective, light-controlled TRPC modulation as a therapeutic strategy to fine-tune macrophage-driven inflammation in lung injury. BK KO mice to intratracheal LPS (acute lung injury) and, in parallel, to bleomycin (lung injury induced fibrosis model) to evaluate both early inflammatory and late reparative/fibrotic outcome. Bronchoalveolar lavage, lung histology, immune cell profiling and cytokine quantification will be used to assess injury severity and resolution. Bioinformatic analysis of RNA-seq data and multi-omics integration will be employed to identify ion channel–regulated inflammatory and reparative programs

References:

1.Viviane Nascimento Da Conceicao, iScience,2021,24 (11),
2.Chen, Y.Cells 2024, 13(4), 322;

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Project Title:

Modulation of TRPC1/3/6 via photoactivable drugs for the regulation of pulmonary vascular tone

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Pulmonary vascular diseases characterized by progressive vascular remodeling of small pulmonary arteries, lead to elevated pulmonary vascular resistance and right heart failure. Endothelial dysfunction and excessive proliferation of smooth muscle cells, both associated with altered calcium (Ca^{2+}) homeostasis are key players in these processes. TRPC channels are critical regulators of intracellular Ca^{2+} levels and are implicated in vascular remodeling[1,2]. Understanding their role could uncover novel therapeutic targets for selective modulation of pulmonary vascular tone.

Hypothesis and Objectives:

Our preliminary data indicate compartment-specific expression of TRPC1/3/6 channels in pulmonary arteries from healthy donors and patients with severe pulmonary vascular disease. We hypothesize that dysregulated activity of these TRPC channels alters Ca2+ homeostasis, contributing to vascular remodeling and elevated pulmonary vascular tone. To test this, we will: i) investigate TRPC1/3/6 function in pulmonary vascular cells using photoswitchable modulators; ii) assess the impact of TRPC-mediated Ca^{2+} signaling on vasoreactivity and vascular integrity and iii) evaluate light-controlled TRPC modulation as a therapeutic strategy.

Methodology:

This PhD project explores the role of TRPC channels in pulmonary vascular cells using both murine models and human donor lungs. The student will isolate endothelial and smooth muscle cells, and apply genetically encoded calcium indicators to monitor real-time Ca^{2+} dynamics in live cells and intact tissues. To precisely control calcium influx, photoswitchable TRPC modulators will be used in combination with advanced imaging techniques. Precision-cut lung slices (PCLS) will preserve the 3D lung architecture. A range of assays will assess TRPC-mediated effects on vasoreactivity (via small-vessel myography and ex vivo murine lung system), barrier integrity (TEER), cell proliferation, and gene expression. High-resolution confocal and super-resolution microscopy will support structural and spatial analysis of TRPC channels. Machine learning–based image analysis will be developed to quantify Ca2+ dynamics in tissue remodeling. This project offers exceptional interdisciplinary training at the

interface of vascular biology, ion-channel physiology, advanced imaging, and bioinformatics-supported image analysis. The PhD candidate will gain hands-on experience in both mouse and human systems, ensuring strong translational relevance.

References:

1. Weissmann N et al. Classical transient receptor potential channel 6 (TRPC6) is essential for hypoxic pulmonary vasoconstriction and alveolar gas exchange Proc Natl Acad Sci USA. (2006). doi: 10.1073/pnas.0606728103;

2. Malczyk et al. Classical transient receptor potential channel 1 in hypoxia-induced pulmonary hypertension. Am J Respir Crit Care Med. . (2013). doi: 10.1164/rccm.201307-1252OC.

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Project Title:

Molecular modelling of human TRPC channels under (patho)physiological conditions

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Calcium ion channels mediate neurotransmission and cardiac function by fixing and modulating the calcium concentrations in the cytoplasm and cellular organelles and constitute one of the central pillars of cell signaling. Thus, the dysregulation of these calcium ion channels and related effectors is associated with different pathophysiological conditions in cardiopathies, pain, cancer, neurological disorders, and respiratory disorders, amongst others. The members of the classical transient receptor potential (TRPC) subgroup of the TRP superfamily are nonselective cation channels located at the human cell plasma membrane. In the last years, the structures of different TRP channels have been solved, which has stimulated their potential as therapeutic targets.1 However, a general understanding of how amino acid mutations on TRPC impact functionality, dynamics, and pathophysiology, or their binding to effectors, lipids, probes or potential drugs to these channels, remains elusive.

Hypothesis and Objectives:

In this project we will investigate the pathophysiology of TRPC3, and other related TRPC channels in the context of lipid membranes by means of advanced simulation techniques. Starting from the all-atom representation of the studied macromolecules, we will generate conformational ensembles to study the role of the network of residues on the dynamics of the channel as well as the degree of coupling of this nextwork between them. This information will be used to shed rationale on the role of each of the positions of these channels on specific diseases and to predict their impact in other physiological conditions. Moreover, we will design and search for binders and probes of different natures able to modulate TRPC function.

Methodology:

The PhD student will be trained in advanced molecular dynamics simulation techniques as well as in a variety of modeling approaches for biological systems. The PhD candidate will learn different de novo algorithms for the prediction of the structure of TRPC channels and will simulate the dynamics of them using from classical force-field molecular dynamics simulations, metadynamics, accelerated Gaussian molecular dynamics, to coarse-grained approaches, amongst others. All these data will be

used as a training dataset for the elucidation of a set of descriptors per residue useful to predict their role in the context of the pathophysiology of the channel:2 dynamics, connectivity with different regions of the channel, or stability, amongst others. Additionally, we will search for new peptide binders and probes able to modulate TRPC function.3 In all cases, the student will benefit from the close and direct collaboration with the experimental team, enabling an iterative validation and refinement of the computational data.

References:

1. Koivisto, AP., Belvisi, M.G., Gaudet, R. et al. Advances in TRP channel drug discovery: from target validation to clinical studies. Nat Rev Drug Discov 21, 41–59 (2022).

2. Mandl Š, Di Geronimo B, Alonso-Gil S, Grininger C, George G, Ferstl U, et al. A new view of missense mutations in α-mannosidosis using molecular dynamics conformational ensembles. Protein Science. 34, e70080 (2025)

3. Lichtenegger, M., Tiapko, O., Svobodova, B. et al. An optically controlled probe identifies lipid-gating fenestrations within the TRPC3 channel. Nat Chem Biol 14, 396–404 (2018)

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Project Title:

TRP channels in extracellular vesicles and their impact on tumor progression

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Dysregulation of calcium signaling has been increasingly recognized as characteristic of different cancer cell types, contributing to survival and the resistance of the tumors to conventional therapies. For example, coordinated Ca2+ oscillations resulted in rapid glioblastoma growth that make this tumor resistant to chemotherapy[1]. In addition, chemoresistance of breast cancers has correlated with higher TRPC5 expression in these tumor cells[2]. Interestingly, extracellular vesicles (EVs), which contain functional TRPC5 channels have been suggested as a basis for spreading of chemoresistance within a tumor entity and thereby contribute to disease progression[2].

Hypothesis and Objectives:

We will investigate the release of TRPC5 EVs from breast cancer tumor cells for the progression of the tumor using a vascularized in-vivo embryonic chicken tumor model, known as CAM assay[3]. Ex-ovo breast cancer tumor progression will be investigated for one week, and at the same time we will monitor Ca2+ and fluorescence imaging of the progressing tumor to determine the role of TRPC5 in chemoresistance development. Our objective is to monitor the formation and transfer of fluorescent labeled TRPC5 EVs and the Ca2+ signals associated with this process in the involved tumor cells with high spatiotemporal resolution. In addition, we will investigate Ca2+-dependent gene regulation programs, e.g. of NFAT in single tumor cells and on CAM tumors to determine tumor remodeling over days. We hypothesize that high-precision modulation of Ca2+ signaling by photopharmacological modulation of TRPC5 will manipulate EV transfer, progression of chemoresistance and tumor remodeling.

Methodology:

The PhD student will be trained in long-term in vivo tumor fluorescence microscopy using the embryonic chicken tumor assay (CAM assay) to monitor TRPC5 expressing EV transfer throughout the tumor. Breast cancer cells will be genetically engineered to express fluorescently labeled TRPC5, highly sensitive Ca2+ sensors, and a reporter vector to track NFAT-dependent transcription and then onplanted on CAM assay. Optical imaging will assess tumor proliferation, invasion, and vascularization. High-resolution imaging will capture TRPC5 EV release in single breast cancer cells and their transfer

between tumor cells. Photopharmacological manipulation of TRPC5 will reveal its role in EV formation, the contribution of Ca2+ signaling, and how the associated Ca2+ signals drive gene regulation and chemoresistance.

References:

1. Hausmann et al. (2022) Autonomous rhythmic activity in glioma networks drives brain tumour growth Nature 613(7942):179-186

2. Wang et al. (2016) Increasing circulating exosomes-carrying TRPC5 predicts chemoresistance in metastatic breast cancer patients Cancer Science 108 (2017) 448–454

3. Handl et al. (2024) Continuous iontronic chemotherapy reduces brain tumor growth in embryonic avian in vivo models J Control Release :369:668-683

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Project Title:

Redox-Sensitive TRP Channel Modulation and Photoregulation of Neuronal and Cardiac Excitability

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Living cells in the brain and the heart communicate through electrical signals. These signals determine whether neurons fire action potentials or whether heart cells beat in a stable rhythm. In many diseases, including chronic pain, neurodegeneration and cardiac arrhythmias, this electrical activity becomes unstable. A major driver of this instability is oxidative stress, in particular short-lived bursts of reactive oxygen species (ROS) such as hydrogen peroxide (H_2O_2) that arise during inflammation, metabolic stress or tissue damage.

Today it is becoming clear that ROS are not only harmful byproducts but also act as important signaling molecules that directly regulate ion channels in the cell membrane. Among the most important of these are TRP channels, which are present in both neurons and cardiomyocytes. When TRP channels are modulated by oxidative signals, they can push cells into a hyperexcitable state associated with pain or arrhythmias, or into a suppressed state associated with fatigue and failure of normal signaling. Despite their importance, it is still poorly understood how oxidative signals control TRP channels in a precise and time-dependent manner, and how this translates into changes in the electrical behavior of cells. One major reason is that it has so far been challenging to generate well-controlled oxidative signals at exactly the place where ion channels operate, namely the cell membrane.

Hypothesis and Objectives:

The central idea of this project is that short, localized oxidative signals act as switches that determine whether TRP channels are turned on or off, and thereby decide how excitable a neuron or a heart cell becomes. The objective of this PhD project is to understand how the timing and strength of membrane-proximal H_2O_2 signals control TRP channel function and how this, in turn, shapes electrical activity in neuronal and cardiac cells. By uncovering these relationships, the project aims to explain how oxidative stress is converted into altered firing patterns in nerves and disturbed rhythm in the heart.

Methodology:

The project will combine modern electrophysiology with light-responsive semiconductor materials to

study living cells at the level of single ion channels and action potentials. Neurons and cardiac cells will be grown in the laboratory and their electrical activity will be measured using patch-clamp techniques, which allow direct recording of ion channel currents and action potentials with high precision.

To control oxidative signaling, special light-sensitive materials will be used that generate small, localized amounts of hydrogen peroxide when illuminated. This makes it possible to trigger oxidative signals exactly at the cell membrane and to vary their timing and intensity in a highly controlled way. By applying these light-induced oxidative signals while recording electrical activity, the project will directly observe how TRP channels and cellular excitability respond in real time.

The resulting data will reveal how oxidative signals shift cells between normal activity, hypersensitivity and electrical instability. These experimental findings will then be used to build simplified computational models that predict how oxidative stress can lead to pain or arrhythmias, providing a bridge from fundamental cell biology to clinically relevant disease mechanisms.

References:

1. Schmidt, T. et al. Light stimulation of neurons on organic photocapacitors induces action potentials with millisecond precision. Advanced Materials Technologies, 2022, 2101159. doi:10.1002/admt.202101159

2. Jakešová, M. et al. Optoelectronic control of single cells using organic photocapacitors. Science Advances, 2019, 5(4): eaav5265. doi:10.1126/sciadv.aav5265

3. Savva, A. et al. Photo-chemical stimulation of neurons with organic semiconductors. Advanced Science, 2023. doi:10.1002/advs.202300473

4. Nowakowska, M. et al. Light-controlled electric stimulation with organic electrolytic photocapacitors achieves complex neuronal network activation. Advanced Healthcare Materials, 2024. doi:10.1002/adhm.202401303

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Project Title:
Tuning the hippocampal calcium signals — how TRPC3 shapes neuronal (patho)physiology

*This project is part of the PhD program **TRPC.at (TRP-dependent cation signaling)**. The program funds 15 positions with clinical/pre-clinical so-supervision (link).*

Background:

Transient receptor potential canonical 3 (TRPC3) is a Ca^{2+}-permeable ion channel that plays an important role in neuronal excitability and intracellular Ca^{2+} signaling. Gain-of-function mutations in TRPC3 cause cerebellar ataxia, as found in the *Moonwalker* mouse model[1], which exhibits excessive Ca^{2+} influx and severe motor coordination deficits. Beyond ataxia, dysregulated Ca^{2+} signaling in the brain has been implicated in neurodevelopmental disorders such as autism spectrum disorder (ASD), although the contribution of TRPC3 remains largely unexplored. Understanding how TRPC3 activity is regulated in neurons and how disease-associated variants alter neuronal function is therefore of high relevance for both basic neuroscience and neurological disease research.

Hypothesis and Objectives:

This project aims to determine how patient-derived gain- and loss-of-function variants of TRPC3 linked to ASD and movement disorders affect Ca^{2+} signaling and neuronal excitability in hippocampal neurons. Using wild-type and mutant TRPC3 constructs together with transgenic mouse models lacking endogenous TRPC channels or selectively expressing TRPC3 alone, we will dissect how TRPC3-mediated Ca^{2+} entry shapes intrinsic firing properties and downstream cellular responses. The central hypothesis is that disease-associated alterations in TRPC3 activity disrupt hippocampal neuronal homeostasis through aberrant Ca^{2+} influx, leading to altered excitability and maladaptive signaling.

Methodology:

The PhD student will gain extensive hands-on training in state-of-the-art electrophysiology and imaging techniques. Viral transduction will be used to express wild-type and patient-derived TRPC3 variants in dissociated hippocampal neurons and organotypic hippocampal slice cultures. Genetically encoded Ca^{2+} indicators will enable high-resolution imaging of TRPC3-dependent Ca^{2+} dynamics, while whole-cell patch-clamp recordings will assess neuronal excitability and TRPC3-mediated currents. Immunocytochemistry and fluorescence microscopy will be used to quantify channel expression and subcellular localization. In addition, photopharmacological tools developed in our laboratory will

provide precise temporal control of TRPC3 activity, allowing direct links between Ca^{2+} influx and functional neuronal outcomes to be established.

Together, these approaches will reveal how disease-associated TRPC3 signaling alters hippocampal physiology and uncover Ca^{2+}-dependent mechanisms underlying TRPC3-driven neuronal dysfunction. This interdisciplinary project offers comprehensive training in cellular neuroscience and provides mechanistic insight into ion channel dysfunction in neurodevelopmental and movement disorders.

References:

1. Becker, E. B. E. *et al*. A point mutation in TRPC3 causes abnormal Purkinje cell development and cerebellar ataxia in moonwalker mice. *Proc. Natl. Acad. Sci. U.S.A.* **106**, 6706–6711 (2009).