CSC-RUB PhD Project Proposal

Title: Mechanisms of Proteostasis Dysregulation in Neurodegenerative Diseases

Sector of research: Molecular and Cellular Neuroscience

Degree awarded: PhD in Neuroscience

Keywords: proteostasis, ubiquitin-proteasome system, autophagy, protein quality control, biomolecular condensates, stress granules, Amyotrophic Lateral Sclerosis, Parkinson's Disease, Huntington's Disease, super-resolution microscopy

Supervisor of PhD project:

Prof. Dr. Konstanze F. Winklhofer
Department of Molecular Cell Biology, Institute of Biochemistry and Pathobiochemistry
Ruhr University Bochum, Universitätsstrasse 150, 44801 Bochum
konstanze.winklhofer@rub.de
https://twitter.com/winklhofer_lab

Research focus of supervisor:

The major aim of our research is to uncover molecular mechanisms underlying neurodegenerative diseases, which is a prerequisite to develop causative therapeutic strategies for these disorders. Specifically, we are studying the role of the proteostasis network in preventing protein aggregation and in promoting degradation of misfolded proteins linked to neurodegeneration. Another focus of our work is the role of mitochondria as key organelles in orchestrating cellular signaling, interorganellar communication, and in regulating viability and bioenergetics. We have a longstanding expertise in cell biology, molecular biology, protein biochemistry, and advanced cellular imaging, including super-resolution microscopy and live cell imaging.

Publications:

van Well EM, Bader V, Patra M, Sánchez-Vicente A, Meschede J, Furthmann N, Schnack C, Blusch A, Longworth J, Petrasch-Parwez E, Mori K, Arzberger T, Trümbach D, Angersbach L, Showkat C, Sehr DA, Berlemann LA, Goldmann P, Clement AM, Behl C, Woerner AC, Saft C, Wurst W, Haass C, Ellrichmann G, Gold R, Dittmar G, Hipp MS, Hartl FU, Tatzelt J, **Winklhofer KF** (2019). A protein quality control pathway regulated by linear ubiquitination. **EMBO J** 38(9): e100730.

Duscha A, Gisevius B, Hirschberg S, Yissachar N, Stangl GI, Eilers E, Bader V, Haase S, Kaisler J, David C, Schneider R, Troisi R, Zent D, Hegelmaier T, Dokalis N, Gerstein S, Del Mare-Roumani S, Amidror S, Staszewski O, Poschmann G, Stühler K, Hirche F, Balogh A, Kempa S, Träger P, Zaiss MM, Holm JB, Massa MG, Nielsen HB, Faissner A, Lukas C, Gatermann SG, Scholz M, Przuntek H, Prinz M, Forslund SK, **Winklhofer KF**, Müller DN,

Linker RA, Gold R, Haghikia A (2020) Propionic Acid Shapes the Multiple Sclerosis Disease Course by an Immunomodulatory Mechanism. **Cell**, 2020 Mar 19;180(6):1067-1080.e16.

Woerner AC, Frottin F, Hornburg D, Feng LR, Meissner F, Patra M, Tatzelt J, Mann M, **Winklhofer KF**, Hartl FU, Hipp MS (2016). Cytoplasmic protein aggregates interfere with nucleo-cytoplasmic transport of protein and RNA. **Science** 351(6269): 173-6.

Müller-Rischart AK, Pilsl A, Beaudette P, Hadian K, Deinlein A, Funke M, Patra M, Motori E, Schweimer C, Kuhn PH, Hrelia S, Lichtenthaler SF, Wurst W, Trümbach D, Langer T, Krappmann D, Dittmar G, Tatzelt, J, **Winklhofer KF** (2013). The E3 ligase parkin maintains mitochondrial integrity by increasing linear ubiquitination of NEMO. **Mol Cell** 49(5): 908-21.

Henn IH, Bouman L, Schlehe JS, Schlierf A, Schramm JE, Wegener E, Nakaso K, Culmsee C, Berninger B, Krappmann D, Tatzelt J, **Winklhofer KF** (2007). Parkin mediates neuroprotection through activation of IKK/NF-kB signaling. **J Neurosci** 27: 1868-1878.

Summary of research plan

Background:

The fidelity of cellular machineries implicated in the removal of damaged and dysfunctional proteins, such as the ubiquitin-proteasome system (UPS) or the autophagy-lysosome pathway, plays a crucial role in maintaining protein homeostasis (proteostasis). A decrease in the efficiency of these systems paralleled by an increase in the abundance of misfolded proteins occurs during aging and is a common denominator of neurodegenerative diseases. Ubiquitination is an essential player in proteostasis regulation but also in cellular signaling in response to various stress conditions. Both cellular degradation systems, the proteasome and autophagy, employ ubiquitin for selection and targeting of substrates to the degradative machineries. We recently found that linear ubiquitination, characterized by an unconventional mode of ubiquitin linkage, plays an important role in proteostasis regulation and prevents the accumulation of protein aggregates found in neurodegenerative diseases (van Well, EMBO Journal, 2019).

Study objective:

The aims of the project are

- to identify E3 ubiquitin ligases and deubiquitinases implicated in remodeling the protein aggregate surface with ubiquitin
- to study how modification of misfolded proteins by ubiquitination affects protein aggregation and degradation via autophagy or the proteosome
- to explore the role of the ubiquitin-like modifier SUMO in proteostasis regulation.

Expected Results:

Based on our previous data, we hypothesize that the specific functions of ubiquitination are mediated by differences in ubiquitin chain architecture. We expect that in the context of this project, components of the ubiquitination machinery are identified that are implicated in protein quality control and can be exploited as therapeutic targets in translational

approaches. At least one research article and one review article should be published based on this project.

Methods:

Advanced cellular imaging (super-resolution microscopy, live cell imaging), state-of-the-art techniques in cell biology, biochemistry, molecular biology, such as CRISPR/Cas genome editing, immunoprecipitation, immunoblotting, immunocytochemistry, immunohistochemistry, reporter gene assays, mass spectrometry.

Candidate Requirements:

We are seeking a highly motivated, enthusiastic candidate with good communication skills, fluency in English and the ability for teamwork. Expertise in primary neuronal cell culture is welcome.

Motivation for CSC application (max 250 words):

We offer a stimulating international research environment with numerous national and international collaborations, integration in research centers (RESOLV Cluster of Excellence, DFG Research Unit 2848, Parkin Consortium of the Michael J. Fox Foundation, International Max Planck Research School for Living Matter), and admission to the Ruhr University Research School for interdisciplinary skills development and mentoring. Laboratory work is supplemented by seminars, summer schools, elective workshops, career development training and participation in national and international conferences.