PROJECT

Supervisor:	Prof. Paola Panina
Title:	Discovery of novel druggable targets in progressive multiple sclerosis
Curriculum:	Neuroscienze e Neurologia Sperimentale

Link to the personal page of the University or relevant hospital site website: http://www.unisr.it/k-teacher/panina-paola/

Description of the Project (max 3,000 characters including spaces)

Background/gap of knowledge

Axonal damage and neurodegeneration are key hallmarks of multiple sclerosis (MS), particularly in its progressive form (PMS). Despite numerous clinical trials, effective treatments for PMS are still lacking with inconsistent results in clinical trials, highlighting a significant unmet clinical need. This project aims to identify novel, druggable targets for progressive MS by validating the neuroprotective effects of repurposed drug candidates previously identified through the multi-stage, multi-cellular phenotypic screening conducted in the BRAVEinMS project. Among these compounds, we focused on bavisant, a specific antagonist of the histamine receptor H3 (HRH3) that promoted neuroprotection/myelination in rodent and human 2D and 3D in vitro assays, and in experimental models of multiple sclerosis. The set of pharmacological data gathered to date by the BRAVEinMS network indicates that bavisant and related repurposed Hrh3 antagonists might represent promising drug candidates for clinical trials in patients with progressive forms of MS.

To achieve this, in vitro functional assays will be performed using neuronal cultures and 3D human induced pluripotent stem cell (hiPSC)-derived neural spheroids. These models will be used to assess the neuroprotective properties of candidate drugs under neurotoxic and neuroinflammatory conditions. Promising compounds were evaluated in vivo using preclinical animal models, with the ultimate goal of translating effective candidates into clinical applications.

The project will employ several key approaches: (i) functional assays to validate the neuroprotective effects of compounds in hiPSC-derived neurons and 3D spheroids; (ii) gene editing techniques to confirm drug targets and elucidate mechanisms of action of HRH3 antagonist); and (iii) in vivo evaluation of neuroprotective efficacy in experimental autoimmune

encephalomyelitis (EAE) and in animal model of progressive MS-like focal lesions by OxPC deposition (oxidized phosphatidylcholines).

The student will play an active role in developing and optimizing cellular assays to investigate drug-induced neuroprotection in human neuronal systems, including measurements of neuronal viability, dendritic morphology, and oxidative stress. Additionally, they will contribute to uncovering the context-specific mechanisms of action of selected compounds using CRISPR/Cas9-mediated gene editing. Molecular and biochemical targets will be further validated in hiPSC-derived 3D brain spheroids through single-cell RNA sequencing (scRNA-seq). Finally, the student will assess the therapeutic potential of selected compounds in vivo using EAE and OxPC models of MS.

Rationale and hypothesis

Multiple sclerosis (MS) is characterized by a complex interplay between neuroinflammation and neurodegeneration, with the latter representing the primary pathological driver of disability in progressive forms of the disease. Consequently, in addition to immunomodulatory therapies, there is a pressing need for neuroprotective strategies aimed at limiting neurodegeneration and slowing disability progression. In this context, validating repurposed drugs with neuroprotective and remyelinating potential may facilitate the identification of novel, druggable targets for PMS.

The project is based on the hypothesis that by directly providing neuroprotection (or indirectly by fostering remyelination) innovative neuroprotective/remyelination add-on therapies could be beneficial in progressive MS and CNS repair. Repurposing of existing drugs and identification of novel pathways involved in injury and repair will translate into future early-phase MS clinical trials.

Objectives and specific aims

The main objective of this project is to validate the neuroprotective properties of the novel repurposed drug candidate in 2D/3D hiPSC-derived neuronal in vitro models and to identify the potential targets/mechanism of action of the compound. The specific aims are:

1. to identify the target through which selected compound mediate neuroprotective/promyelinating effects in vitro

2. to define the role of HRH3 in (re)-myelination and neuroprotection, using loss-of-function studies in in vitro models.

3. to evaluate the efficacy of the drug candidate in vivo preclinical models and its potential relevance to neuroprotection in PMS.

Expected outcomes

We expect to identify therapeutic strategies through drug-repurposing to identify safe-in-human compounds favoring neuroprotection and remyelination in experimental models of MS and ultimately in the clinic. Since the focus is on HRH3 antagonism, we expect to further identify the mechanism of action of this drug as a neuroprotectant and pro-remyelinating therapy in progressive MS models.

Skills that the student should acquire (max. 600 characters including spaces):

- Pose a research question
- examine the range of modes of inquiry
- identify the appropriate research mode and procedure, define a sample/population
- identify a data collection strategy; analyze and interpret the data; draw conclusions from the data
- acquire experimental skills in the context of in vitro cellular models and organoids, and in vivo animal models
- write research reports/papers
- present a scientific project to an internal and external audience

References (max. 15)

1. Chataway J, De Angelis F et al., (2020), Efficacy of three neuroprotective drugs in secondary progressive multiple sclerosis (MS-SMART): a phase 2b, multiarm, double-blind, randomised placebo-controlled trialThe Lancet. Neurology, 19, 3, 3

2. Morris J, Soman K, Akbas R et al. (2023), The scalable precision medicine open knowledge engine (SPOKE): a massive knowledge graph of biomedical information. Bioinformatics, 39(2)

3. Pamies D, Barreras P, Block K, Makri G, Kumar A, Wiersma D, Smirnova L, Zang C, Bressler J, Christian KM, et al (2017) A human brain microphysiological system derived from induced pluripotent stem cells to study neurological diseases and toxicity. ALTEX 34: 362–376

4. Miller SD & Karpus WJ (2007) Experimental autoimmune encephalomyelitis in the mouse. Curr Protoc Immunol Chapter 15: Unit 15.1

5. Jean-Pierre Julien & Jasna Kriz (2006) Transgenic mouse models of amyotrophic lateral sclerosis, Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease, 1762: Issues 11–12, 1013-1024

6. Nadjet Gacem, Svetlana Bezukladova, et al. (2026) In silico screening and preclinical validation identify bavisant as a therapeutic candidate for multiple sclerosis. Sci Transl Med 18(833):eads0633. doi: 10.1126/scitranslmed.ads0633.

7. Yu et al. (2025) "Oxidized phosphatidylcholines deposition drives chronic neurodegeneration in a mouse model of progressive multiple sclerosis via IL-1β signaling" Nature Neuroscience; PMID 41326786