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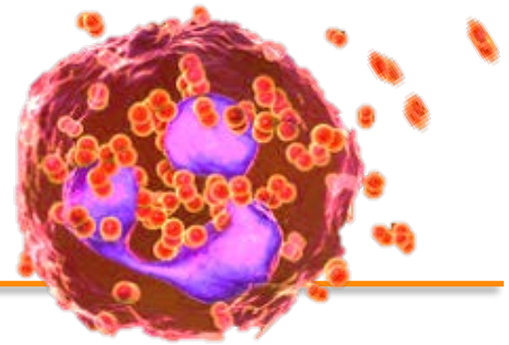
Hellenic Foundation for Research & Innovation

Description of the funded research project

1st Call for H.F.R.I. Research Projects to Support Faculty Members & Researchers and Procure High-Value Research Equipment

Enhancing the beneficial functions of CNS macrophages to promote remyelination as a prototypic therapeutic strategy for the treatment of progressive multiple sclerosis



Principal Investigator: Lesley Probert, PhD

Reader-friendly title: Brain Repair by Macrophages

Scientific Area: Life Sciences

Host Institution:
Hellenic Pasteur Institute, Athens, Greece

Collaborating Institutions:
Imperial College, London, UK
INmune Bio, La Jolla, CA, USA

Budget: **179.957,12 €**

Duration: **36 months (28/02/2020-27/02/2023)**

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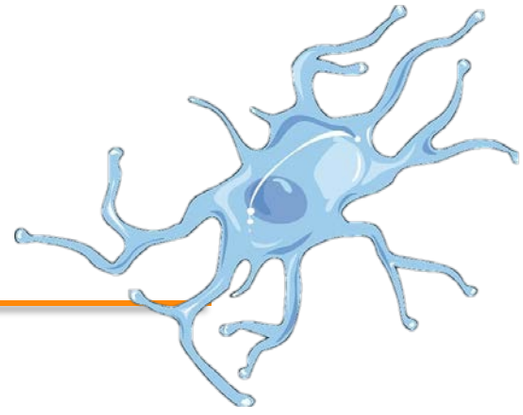
MacRepair



Multiple sclerosis (MS) is a chronic immune-mediated disease of the central nervous system (CNS) causing demyelination and neurodegeneration. In most patients, disease starts with relapsing-remitting symptoms (RRMS) due to autoimmune attack of the CNS and can be effectively treated with immuno-regulatory drugs. With increasing age and for unknown reasons, disease transits into a progressive form (PMS), where chronic “slow-burning” inflammation sets-up in the CNS white and grey matter and becomes resistant to the available drugs. Remyelination, a primary mechanism of neuroprotection and repair in the CNS, largely fails in MS and there is a desperate need for innovative approaches to restore lost myelin as a treatment for PMS.

MacRepair is based on two major recent advances in our understanding of CNS demyelination:
First, CNS macrophages named microglia, are critically involved in phagocytosing and clearing myelin debris from lesions, which in turn stimulates remyelination by oligodendrocyte precursor cells (OPC).
Second, soluble TNF, a master pro-inflammatory cytokine, inhibits remyelination by inhibiting the phagocytosis of myelin debris by CNS macrophages, and is therefore a candidate therapeutic target for PMS.

In MacRepair we will combine cutting-edge technologies in conditional gene targeting in mice, disease modelling, novel brain-penetrating biologics and single cell RNA sequencing of brain cells to address the following objectives:

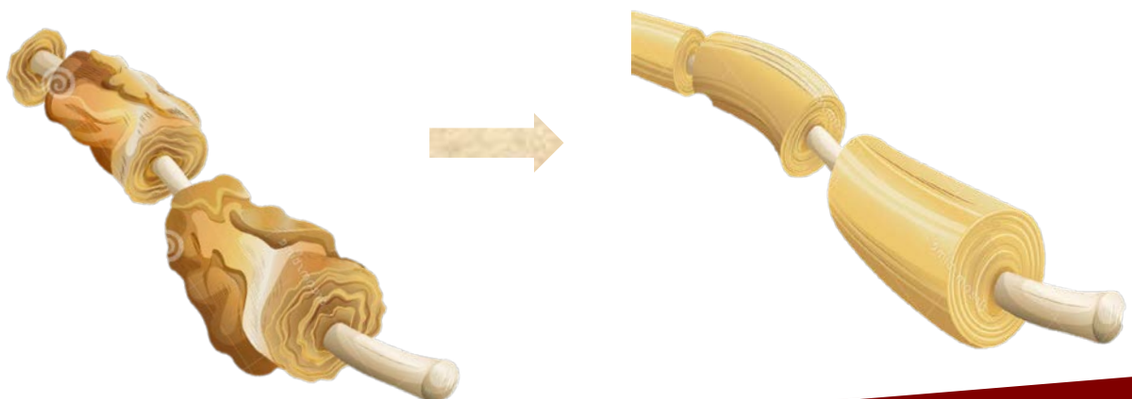
- Define the functional contribution of different macrophage populations (infiltrating macrophages versus microglia) to CNS demyelination & remyelination.
- Identify the main molecular mechanisms that switch CNS macrophages between beneficial pro-repair & deleterious inflammatory functions in the demyelinating lesions.
- Investigate the macrophage autonomous role of proinflammatory TNF/TNF receptor 1 signalling in remyelination failure in MS.
- Identify primary therapeutic targets in CNS macrophages for promoting remyelination as a treatment for PMS.

Project originality

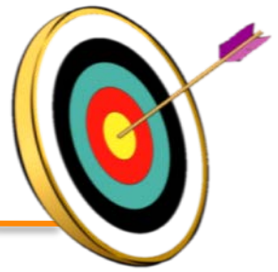
Current treatments for MS can prevent relapses, which are caused by infiltration of immune system cells into the brain and spinal cord tissues, but are not effective in preventing neuron damage that starts from the very beginning of disease and accumulates over time leading to irreversible neurological deficits and brain atrophy. Thus, identifying the cellular and molecular mechanisms of the primary CNS repair mechanism, named remyelination, is key to the development of novel neuroprotective and reparative therapies for use during the entire course of this disease.

Our recent discovery that a novel selective inhibitor of the pro-inflammatory cytokine soluble TNF (solTNF) penetrates the blood-brain barrier and profoundly promotes remyelination in an experimental demyelination model by increasing beneficial macrophage function demonstrates the potent inhibitory effect of neuroinflammation on the remyelination process. **In MacRepair we will test the core hypothesis that CNS macrophages (including microglia and infiltrating macrophages) are central players in determining whether demyelinated axons will become remyelinated and therefore protected, or not during the course of a demyelinating disease.** We propose that solTNF, produced locally in the inflamed CNS, is directly responsible for switching CNS macrophages away from a beneficial pro-repair phenotype towards a delererious, pro-inflammatory form, via its receptor TNFR1, and that inhibitors of solTNF/TNFR1 signaling are therefore promising anti-inflammatory and neuroprotection therapies for use in all types of MS.

The project will develop novel strains of conditionally gene-targeted mice for TNF receptors for investigating the differential contributions of microglia and peripheral macrophages to demyelination and remyelination; will transfer expertise for gene expression analysis of single cell nuclei captured from brain lesions into the Greek lab; and will use novel proprietary brain-penetrating biologics to provide proof-of-principle that increasing beneficial pro-repair macrophages is sufficient to promote remyelination, thereby opening an interesting new field in drug design for the treatment of MS.



Expected results & Research Project Impact



There is no effective therapy for PMS. As of 2020 there are fourteen approved therapies for treating relapses in RRMS, but only one of these (ocrelizumab) shows any effect in PMS and this is not yet approved for use in Greece. None of the current therapies stop the transition of disease from RRMS to PMS. Remyelination is the primary mechanism that would prevent or reverse PMS by mediating neuroprotection and myelin repair, but there are still no drugs to kick-start or enhance this important CNS repair process when it fails.

Strategies for promoting remyelination are the holy grail of research aiming to develop effective therapies for PMS as well as for classical neurodegenerative diseases, trauma and stroke. In **MacRepair** we focus on defining the roles of CNS macrophage populations in this process, and particularly on identifying the molecular mechanisms that control the generation of beneficial pro-repair macrophages over deleterious pro-inflammatory macrophages in MS lesions. We expect, (1) to validate the potent inhibitory effect of soluble TNF/TNFR1 signaling on CNS remyelination and to prove that selective inhibition of soluble TNF using a novel brain-penetrating inhibitor enhances the generation of pro-repair macrophages, promotes remyelination and is therefore a promising therapeutic approach for PMS, (2) to define the individual roles of peripheral macrophages and CNS microglia in remyelination, (3) to identify and validate additional molecular targets for promoting pro-repair macrophages and remyelination.

The socioeconomic impact of MS in the West is high. MS is a lifelong disease affecting ~2.3 million people worldwide starting mostly in young adulthood. The majority (85%) of affected individuals develop a slow and irreversible progressive disease (PMS) characterized by increasing loss of mobility and cognitive impairment leading to major societal impacts with a high economic and personal burden. The discovery of drugable targets that would promote CNS remyelination as a therapeutic approach for PMS and possibly also other degenerative CNS conditions is expected to have high impact in the field of human health.

The importance of this funding

- **Innovative research:** MacRepair is expected to establish the role of different CNS macrophage populations in remyelination, to define the main pathways responsible for switching macrophages/microglia between pro-inflammatory and pro-repair functions, and thereby to identify novel candidate therapeutic targets for PMS, which is still untreatable.

- **Young scientists**: The project is multidisciplinary and engages two talented young scientists, one biologist and one computer engineer/bioinformatician, who have registered for PhD degrees based on their projects in MacRepair. The research will train them in state-of-the-art methodologies and allow them to collaborate with international experts, to transfer acquired expertise to Greece, and to communicate research ideas and results at international conferences and in refereed publications.

- **International collaboration:** We have interactive external collaborators who are leaders in the fields of drug engineering and development (INmuneBio, La Jolla, USA) and of single nuclei RNA sequencing and analysis of gene expression (Imperial College Faculty of Medicine, London, UK).

- **Transfer of technology:** The project foresees 1-2 visits by researchers from HPI to Imperial College London, where they will collaborate hands-on in methodologies for gene expression profiling of single macrophage nuclei captured from demyelinating lesions in mice, and analysis using state-of-the-art computational facilities. This know-how will be transferred back to the host Institute in Greece.

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COMMUNICATION

185 Syggrou Ave. & 2 Sardeon St. 2
171 21, N. Smyrni, Greece
+30 210 64 12 410, 420
communication@elidek.gr
www.elidek.gr