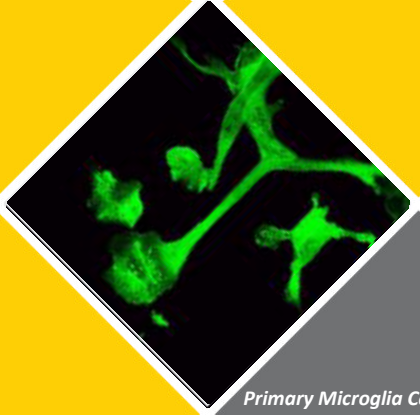
Autoimmune Diseases

MULTIPLE SCLEROSIS



Primary Microglia Cells Challenged and Quantitatively Monitored for their Inflammatory Profile

Background

One of the many open questions in multiple sclerosis research is whether inflammation in the CNS is initiated by an autoimmune attack, triggered by unidentified environmental factors, or represents a response to axonal degeneration and myelin degradation secondary to processes that are intrinsic to the CNS. Lesions characterized by microglial activation and hypoxia-like characteristics, as well as cortical lesions and the slowly progressive chronic phase of the disease, are likely driven by activated myeloid cells.

However, at present it is not clear what keeps the microglial cells activated. It is possible that the T cells found throughout the CNS of patients with multiple sclerosis provide constant stimuli, i.e. by pro-inflammatory cytokines, which activate microglia.

Pathology Model

In order to recreate an in vitro pathological scenario mimicking MS condition, microglia cells will be exposed to detrimental challenges known to activate microglia in the neuroinflammatory scenarios leading to MS (i.e. hypoxia and LPS exposure) and functional parameters will be quantitatively evaluated.

Moreover, the effect of the CLIENT's compound in the modulation of the detrimental scenario will be quantitatively monitored in a dose response fashion.

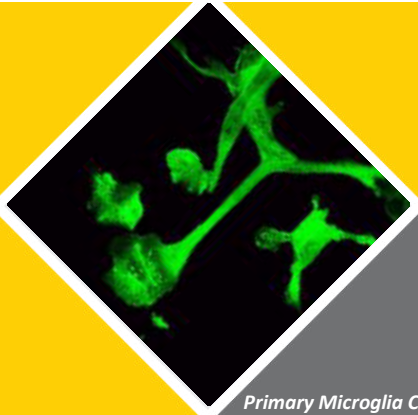
Readouts

The following parameters will be quantitatively evaluated:

Biochemical Characterization

- Cell viability and toxicity: (i.e. MTT assay)
- Cell migration: (i.e. chemotactic chamber)
- Total ROS production: (i.e. DCF-DA fluorescent assay)
- Phagocytic activity: (i.e. Vybrant Assay)
- Mitochondrial damage: (i.e. HCS Mitochondrial Health assay)
- Cytokine production by inflammatory panel on multiplex ELISA: (i.e. IL-1α , IL-1β, IL-2, IL-4, IL-6, IL-8, IL-10, VEGF, TNF-α, IFN-γ, EGF, MCP-1)
- NO production: (i.e. Griess assay)

MULTIPLE SCLEROSIS



Primary Microglia Cells Challenged and Quantitatively Monitored for their Inflammatory Profile

Functional Characterization

- Membrane permeability (i.e. Yo-Pro1 uptake)
- Microvesicle shedding