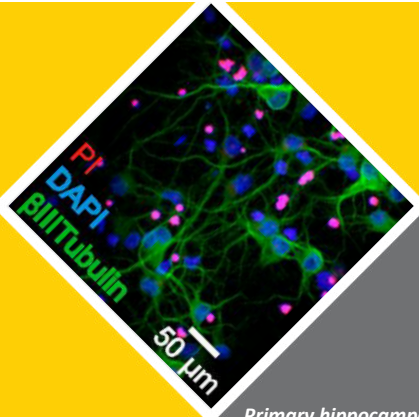
Brain Diseases

ALZHEIMER'S DISEASE



Primary hippocampal neurons in culture

Background

Alzheimer's disease (AD) is the most common cause of dementia among ageing population. However, the mechanisms regulating synaptic dysfunction in AD are not fully understood, and require an in depth analysis of the crosstalk mechanisms ongoing during an inflammatory event, between the neuronal and non-neuronal cells present in the microenvironment, playing a crucial role in disease onset.

Pathology Model

In order to evaluate the client's compound modulatory activity of the neuroinflammatory events leading to neurodegeneration in AD, hippocampal neurons will be exposed to either Abeta oligomers, or to glial Abeta- primed glial medium. Both the direct effects on hippocampal neurons as well as the microglial-mediated effects will be taken into consideration.

Readouts

The following parameters will be analyzed:

Step 1 - Direct Neuronal Damage

Primary neuronal cultures isolated from Sprague Dowley rats will be exposed to Abeta oligomers in the presence / absence of CLIENT's. The following parameters will be analyzed:

1. Morphological Characterization
    - Qualitative evaluation of neuronal cytoskeletal disruption
    - Quantitative evaluation of dendritic branching
    - Quantitative evaluation of neurite elongation modulation
2. Biochemical Characterization
    - Quantitative evaluation of neuronal cell death (i.e. PI/DAPI/Calcein AM)
    - Quantitative evaluation of caspase activation (i.e. 3 or 8 or 9)
    - Quantitative evaluation of DNA degradation (i.e. TUNEL staining)
3. Analysis of Oxidative Stress
    - Quantitative evaluation of total ROS production
    - Quantitative evaluation of NO production

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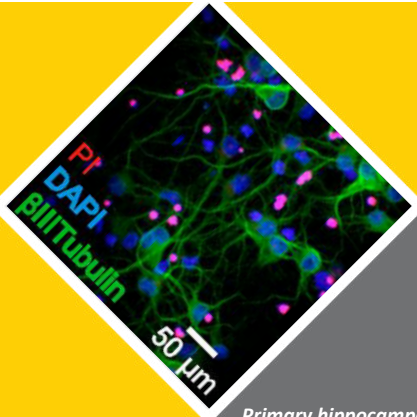

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Step 2 - Glial Pro-Inflammatory Phenotype

Microglia cells (i.e. BV2) will be challenged with Abeta oligomers in the presence/absence of CLIENT's compound. The following parameters will be quantitatively monitored on glial cells:

1. Biochemical characterization
    - Quantitative evaluation of metabolic activity
    - Inflammatory cytokine production: (i.e. IL1 beta, TNFalpha, IL6)
    - Total ROS production
    - Quantification of NO production

2. Functional characterization
    - Quantitative evaluation of phagocytic potential
    - Quantitative evaluation of intracellular calcium dynamics
    - Quantitative evaluation of membrane permeability
    - Quantitative evaluation of microvesicle shedding