Biochemical Characterization and Imaging of Arc: Insights into Neurodegeneration and Alzheimer's Development

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Alzheimer's Disease is is characterized by loss of synapses in the cerebral cortex and accumulation of extracellular Beta-Amyloid Peptides (neuritic plaques). Assemblies of Beta-Amyloid are produced by the sequential cleavage of amyloid precursor protein (APP) by Beta-secretase (BACE1) and Y-secretase. Arc, a ~50 kDa IEG protein, has been shown to possibly interact with Presenilin1 (PS1) to regulate Y-secretase trafficking and confer activity dependence. Existing research on Arc has mostly focused on its role in endocytic machinery, where it regulates neuroplasticity by promoting endocytosis of AMPA-type glutamate receptors (AMPARs). Last year, I identified how Arc's self-association and interaction with Dynamin 2 modulates endocytosis of AMPARs, yet the mechanisms as to how exactly these proteins interact remained unclear. In this project, I show how the associations occur directly on the plasma membrane, where Arc self-associates into higher-order structures to act as a scaffold for interactions with PS1. For this study, bacterially expressed protein, GST-Arc was characterized for its potential interaction on cell membranes. Unilaminar Vesicles acted as Cell Membranes, as Fluorescence Correlation Spectroscopy (FCS) studies were done to quantify Arc's interactions with lipids. Furthermore, HEK cells were transfected with plasmids for EGFP-Arc to identify expression and association on cell surface and within the cytosol using Number and Brightness (N&B) method. My findings support the model of inactivated Arc in the cytosol being directed to the plasma-membrane where activation and self-association leads to scaffolding, allowing Arc to interact with PS1 ultimately leading to activity-dependent generation of assemblies of Beta-Amyloid.

Awards Won: