“Elucidating the role of cysteine modifications of proteins at the synapse: How alpha-synuclein pre-formed fibrils alter dendritic spine density”

Alpha-synuclein (α-syn) is a 14kDa protein found in neurons that is implicated in multiple neurodegenerative diseases. Under pathophysiological conditions α-syn aggregates to form fibrils, a toxic precursor of the insoluble Lewy bodies that deposit in these neurons. It has recently been reported that mitochondrial stress in the form of increased reactive oxygen and nitrogen species is a direct consequence of α-syn accumulation on the mitochondrial membrane. I propose to determine the oxidative changes to proteins evoked by this stress and their impact on synaptic dysfunction in synucleinopathies. I hypothesize that oxidative modifications of synaptic proteins underlie the synaptic dysfunction that results from α-syn misfolding and accumulation on mitochondrial membranes. I will first examine the effects of cysteine oxidation on rat primary cortical neurons caused by exposure to pre-formed fibrils (PFFs). PFF exposed neurons will be imaged using a confocal microscope to determine the effect on the dendritic spines of the neurons. Next, I will determine the in vitro oxidative proteome evoked by α-syn aggregation. This will be done using biochemical techniques to assess if PFF exposure and subsequent α-syn accumulation causes an increase in cysteine modifications. The cysteine modified proteins will be determined by mass spectrometry and prioritized based on overall protein abundance and known function at the synapse. Finally, I will elucidate the role of cysteine modified proteins on spine morphology and function through overexpression of non-redox modifiable variants.