Kinetic Characterization of Phospholipase C Gamma 2 Using Mass Spectrometry

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Neurodegeneration is an increasing health crisis in the United States, with Alzheimer's Disease (AD) currently being the sixth leading cause of death for people older than sixty-five. There are no drugs that reverse or cure AD, and the four FDA-approved drugs for AD only help to alleviate symptoms. My project seeks to address the unmet need for a drug that treats AD by researching a novel AD enzyme target, phospholipase C gamma 2 (PLCG2). PLCG2 is an enzyme expressed in microglial cells in the brain, and PLCG2 catalyzes the transformation of phosphatidylinositol 4,5-bisphosphate (PIP2) into inositol 1,4,5-trisphosphate (IP3) and diacylglycerol (DAG), which activates microglia to phagocytose beta-amyloid, a protein linked to AD pathology. We hypothesize that a small-molecule activator of PLCG2 could be therapeutically beneficial for people with AD by stimulating phagocytosis of beta-amyloid by microglia. In this study, I purified PLCG2 for kinetic studies using desorption-ionization mass spectrometry (DESI-MS) in collaboration with Dr. Graham Cooks lab. Kinetic experiments found that catalysis of PI(4,5)P2 of various lipid tail lengths shows substrate cleavage efficiency based on lipid size. Furthermore, this method can track PLCG2 activity in response to adding small molecules—ATP inhibition was quantified by mass spectrometry, and future high-throughput screening using DESI-MS will be completed to find small-molecule activators of PLCG2. Kinetic characterization of PLCG2 will build the foundation to perform high-throughput screening for activating compounds as the first step in the drug development pipeline for developing PLCG2 as a potential therapeutic for AD.