Molecular Insights into Phospholipase Cε Regulation

Phospholipase Cε (PLCε) is essential for normal cellular function in the cardiovascular system, where it generates second messengers that increase intracellular calcium and activate downstream kinases. PLCε has modest activity under basal conditions, but is stimulated following the activation of GPCRs, through direct interactions with heterotrimeric and small GTPases, in particular Rap1A. We have used small angle X-ray scattering, single particle electron microscopy, and biochemical assays to begin characterizing the structure and regulation of PLCε, alone and upon binding of Rap1A. We have shown that PLCε is conformationally heterogeneous in solution, with its PH and RA domains also contributing required for maximum basal activity and stability. The C-terminal RA domain has been previously identified as the Rap1A binding site. We propose the conformational flexibility of this RA domain is essential for Rap1A-dependent activation, and that the mechanism has an allosteric component. Taken together, these studies provide the insights into the molecular basis underlying basal and activated phosphatidylinositol hydrolysis by PLCε.