Joseph Lyons
Department of Molecular Biology and Genetics, Aarhus University, Denmark

Preparation of lipid flippases for cryo-EM studies.

P4-ATPases are lipid flippases that drive the ATP-dependent inward translocation (flipping) of lipids within the membrane. They are members of the P-type ATPase superfamily and largely function as binary complexes with an auxiliary protein from the CDC50 family. Select lipid flippases are auto-regulated by conserved motifs in their termini and require activation by an external stimulus, i.e., binding of an activating lipid or regulatory protein, and/or specific phosphorylation of the termini. To elucidate the auto-regulation and transport mechanisms, we determined the structures of yeast and mammalian lipid flippases by cryo-electron microscopy.

In this talk, I will give an overview of the tour de force we embarked on, from expression through purification to cryo-EM imaging of these lipid flippases.