# Spatiotemporal control of endocytosis - from mechanisms to synaptic function

**Volker Haucke**

Leibniz Institut für Molekulare Pharmakologie (FMP) Berlin
Robert-Roessle-Straße 10, 13125 Berlin, Germany

Clathrin/ AP-2-mediated endocytosis (CME) serves crucial roles in cell physiology including the regulation of cell signalling, development, or neurotransmission, and serves as an entry port for pathogenic bacteria and viruses. How CME is controlled in space and time is unknown.

In my talk I will summarize our recent findings regarding the role of PI conversion from phosphatidylinositol-4,5-bisphosphate [$PI(4,5)P_2$]-positive early to phosphatidylinositol-3,4-bisphosphate [$PI(3,4)P_2$]-positive late endocytic intermediates en route to endosomes. We recently demonstrated that timed formation of $PI(3,4)P_2$ by class II phosphatidylinositol 3-kinase C2α (PI3K C2α) spatiotemporally controls CME by enabling enrichment of the BAR domain protein SNX9 at late-stage endocytic intermediates prior to fission. Combined mathematical modelling, super-resolution imaging, and genetic manipulations provide a mechanistic framework for how PI conversion from $PI(4,5)P_2$ to $PI(3,4)P_2$ and subsequent assembly of SNX9-containing scaffolds spatiotemporally control CME en route to endosomes.

In the second half of my presentation I will discuss our most recent data regarding the mechanism of synaptic vesicle recycling in the brain and in auditory inner hair cells. We find depletion of clathrin or conditional knockout of AP-2 results in defects in synaptic vesicle reformation and an accumulation of endosomes, indicating that clathrin/ AP-2 mediate synaptic vesicle reformation mainly form internal endosomal structures rather than the plasma membrane. Presynaptic membrane retrieval and endosome formation largely occur by clathrin-independent endocytosis via dynamin 1/3 and endophilin. These results together with theoretical modelling provide a conceptual framework for how synapses capitalize on clathrin-independent endocytosis and clathrin/ AP-2-mediated vesicle reformation from endosomes to maintain excitability over a broad range of stimulation frequencies.