

Abstract

Lysosomes are degradation hub of cells breaking down endocytic, phagocytic and autophagic cargo. Lysosomes also play crucial role in diverse cellular processes such as plasma membrane repair, antigen presentation, tumor invasion and metabolic signaling. Arl8b is a small GTP-binding protein (G) that predominantly resides on lysosomes and regulates both its anterograde motility and fusion with late endosomes by recruiting its downstream effectors- SKIP and HOPS complex, respectively. To further explore the role of Arl8b and its effectors, we have identified RUN domain-containing proteins, PLEKHM1 and Rabip4', as novel Arl8b interaction partners and elucidated the significance of these interactions. We show that both PLEKHM1 and Rabip4' bind to Arl8b through their respective N-terminal RUN domains, but are diverse in their localization and lysosome regulation. We deciphered that PLEKHM1 serves as a linker between Arl8b and late endosomal small G-protein Rab7, facilitating the recruitment of multisubunit tethering factor HOPS complex to vesicle contact sites and regulates degradation of endocytic and autophagic cargo. We also noted that PLEKHM1 and SKIP, both of which bind to Arl8b via their respective RUN domains compete for binding to Arl8b and position lysosomes in opposing manner. In the second project, we found that Rabip4' and Arl8b colocalized primarily on early endosomal structures and Arl8b depletion led to partial loss of Rabip4' from endosomal membranes. Rabip4' and Arl8b interaction is essential for maintaining endolysosomal morphology and regulates endocytic cargo degradation. Further, Rabip4' regulates maturation of Salmonella-containing vacuole in the host cells and Rabip4' depletion led to defective Salmonella replication in mammalian cells. Characterization of PLEKHM1 and Rabip4' as interaction partners of lysosomal small GTP-binding protein Arl8b expands our understanding of the molecular players regulating lysosome biology.