P028 Structural and functional analyses of ALIX interactions with host CHMP4 and retrovirus Gag proteins

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The ESCRT pathway facilitates membrane remodelling events in a series of diverse biological processes including enveloped virus budding, multivesicular body formation, and cytokinesis. To promote retrovirus budding, the ESCRT pathway-associated protein ALIX functions as an adaptor protein that links the viral Gag protein with the CHMP4 subunits of the ESCRT-III complex. Specifically, the V domain of ALIX binds short peptide YPX_nL motifs (late domains) within the HIV-1 and EIAV Gag proteins, while the Bro1 domain of ALIX binds C-terminal sites on the three human CHMP4 proteins (CHMP4A-C). Here, we report crystal structures of $ALIX_{Bro1}$ in complex with its CHMP4 binding sites, which reveal that the CHMP4A-C termini form amphipathic helices that bind in similar sites across the conserved concave surface of $ALIX_{Bro1}$ and present a unique pattern of hydrophobic residues that allows ALIX to differentiate between CHMP4 and other ESCRT-III proteins. We also report the crystal structures of ALIX in complex with YPX_nL late domains from HIV-1 and EIAV. These structures reveal how ALIX connects retroviral Gag proteins to the downstream ESCRT-III machinery to facilitate retrovirus budding.