



Supplementary Figure. (A) HeLa cells were co-transfected to express both FLAG-tagged PX-BAR from SNX9 and myc-tagged PX-BAR from SNX18. After incubation with appropriate antibodies, the cells were visualized by epifluorescence. The boxed area is magnified below. Scale bar is 10 μ m. Note the high degree of co-localization on the same tubes, which in higher magnification reveals a patchy distribution of the two proteins. (B) Endogenous SNX18 and Arf1 were visualized by immunofluorescence in HeLa cells. The magnifications of the boxed area show SNX18 and Arf1 separately and the merge image reveals minimal overlap between the stains. Scale bar is 10 μ m. (C) HeLa cells were stained for endogenous SNX18 and clathrin. Insets show magnification of the boxed sections. SNX18 and clathrin positive puncta do not co-localize. Scale bars are 10 μ m. (D) Pull-down experiment with GST and GST-fusion proteins with the LC-domains of SNX18 and SNX9. GST-proteins bound to beads were incubated with clathrin purified from K562 cytosol (Lundmark and Carlsson, 2003). Bound clathrin was analyzed by immunoblotting and densitometry analysis revealed that 20% of input bound to SNX9-LC, whereas no significant binding of clathrin could be detected for SNX18-LC and the GST control. The Coomassie-stained gel shows the GST-proteins. Bands below GST-LC are breakdown products of the fusion proteins.