Supplementary figure 1.

A  VPS35  mCherry-ATG9A  overlay

B  TBC1D5  LC3  Atg9A  overlay

DMSO  EBS (4h)  KL-0063794 (6h)

C  TBC1D5  myc-VPS29  LC3  overlay

DMSO  KL-0063794 (6h)
**Fig S1 | TBC1D5 and retromer associate with ATG9.** (A) Co-localization of endogenous VPS35 with ATG9A in U2OS cells stably expressing mCherry-ATG9 during autophagy induced by treatment with mTOR inhibitor KU0063794 for 6h. 2D plots represent signal intensities for both fluorophores across the marked vesicle (red line- VPS35, green line- ATG9A) (B) U2OS cells treated with EBSS for 4h or KU0063794 for 6h have been fixed and immune-labeled with anti-TBC1D5, anti-ATG9A and anti-LC3 antibody. Magnified pictures represent the regions of co-localization of endogenous TBC1D5, ATG9 and LC3, and 2D plots represent signal intensities for each fluorophore across the marked vesicle (blue line-TBC1D5, red line- LC3, green line- ATG9A) (C) U2OS cells, transiently transfected with myc-VPS29 have been treated for 6h with KU0063794, subsequently fixed and immune-labeled with anti-TBC1D5 and anti-LC3 antibodies. Magnified regions show that LC3 and TBC1D5 positive vesicles do not co-stain for myc-VPS29.