

Figure S1

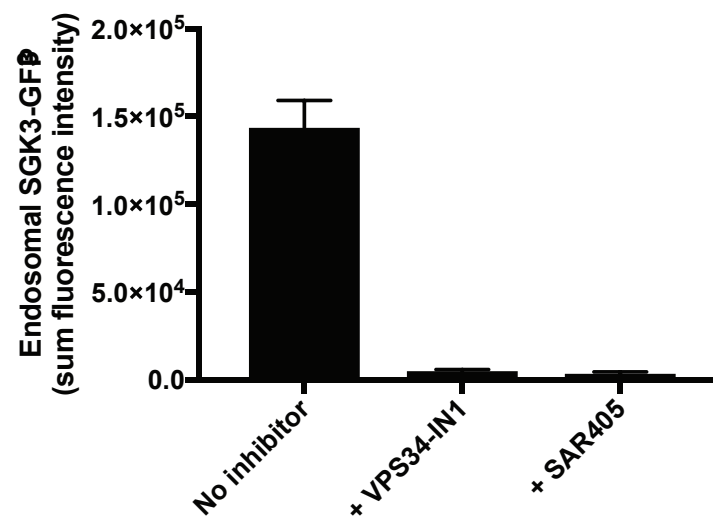
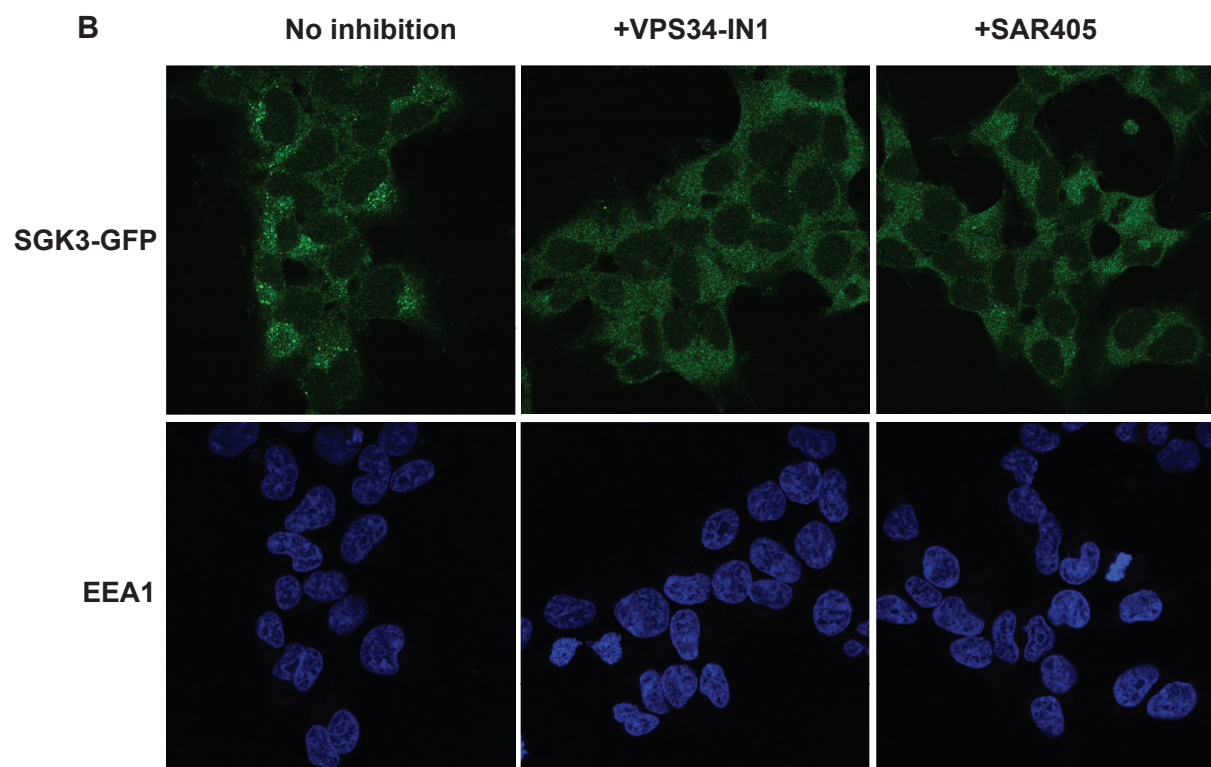
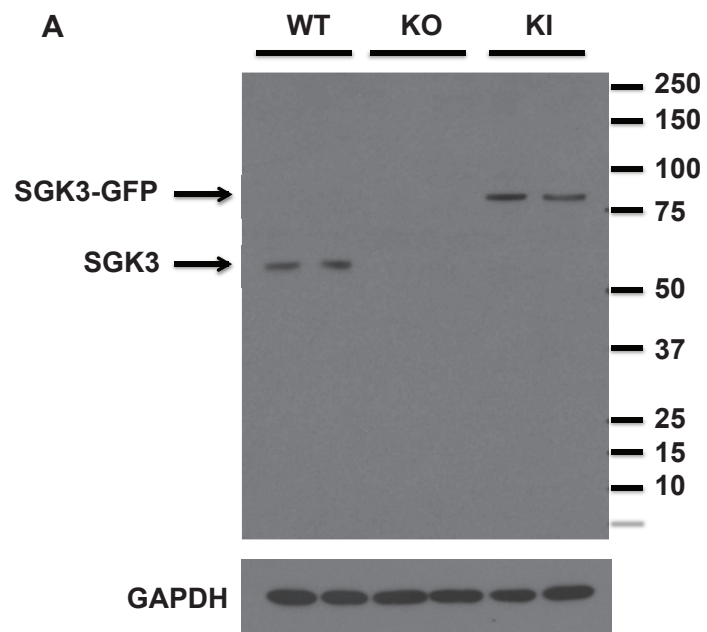


Figure S1 – Generation of SGK3 knock out and GFP-SGK3 knock in cell lines and the impact of hVPS34 inhibition on GFP-SGK3 localisation. (A) Wild type (WT), SGK3 knock-out (KO) and GFP-GFP-SGK3 knock-in (KI) HEK293 cells were lysed and cell lysates subjected to immunoblot analysis with the indicated antibodies. **(B)** GFP-SGK3 knock-in HEK293 cells cultured in 10% (v/v) serum were treated with or without 1 μ M VPS34-IN1 or 1 μ M SAR405 for 1 hour prior to fixation with 4% (v/v) paraformaldehyde. GFP distribution in cells was visualized using chicken anti-GFP primary and anti-chicken Alexa Fluor-488 to enhance the GFP signal (upper panel). DAPI was used to stain nuclei (lower panel). Histogram represents quantitation of GFP-SGK3 on endosomal structures using sum of intensity of fluorescent signal from staining of GFP \pm S.E.M. following the various treatments.