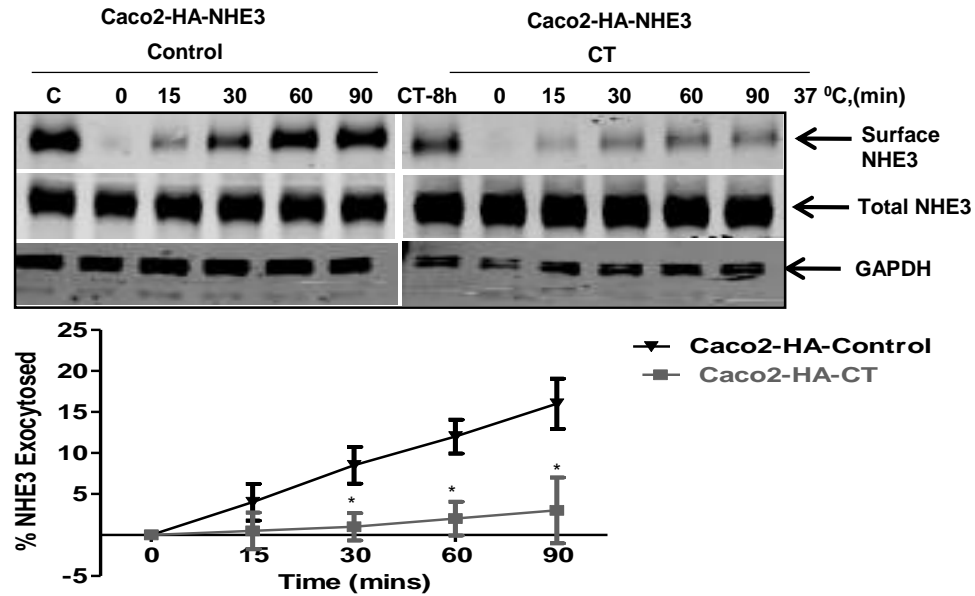


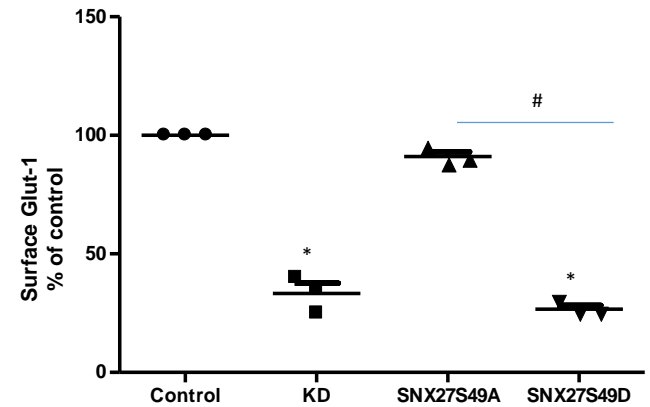
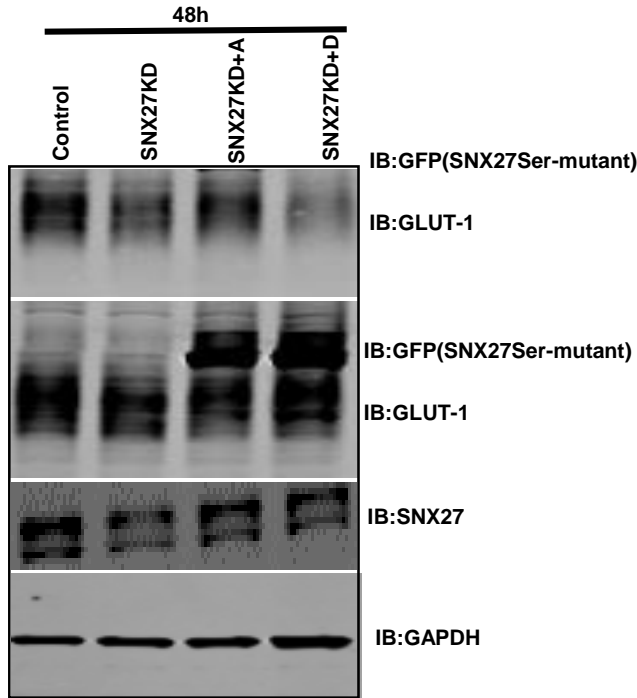
Fig. S1



**Cholera toxin inhibits exocytosis of NHE3 in intestinal epithelial cells.**

Immunoblot and densitometry analysis of untreated control and CT treated (100ng/0.5ml-8 h) confluent Caco-2/bbe-HANHE3 cells. Results are mean  $\pm$ SD of three experiments. \* $p < 0.05$  values are comparison between control and CT-treated cells.

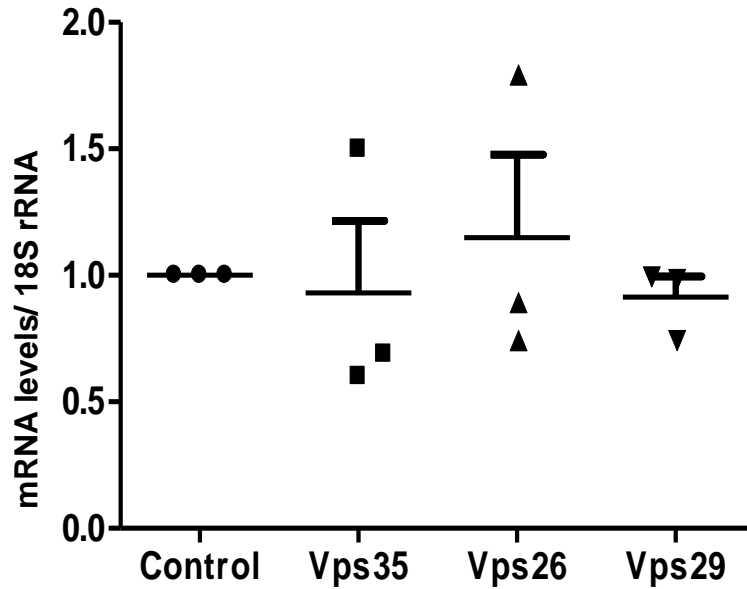
Fig. S2



**Surface expression of Glut-1 in Ser49D containing cells is similar to SNX27KD cells.**

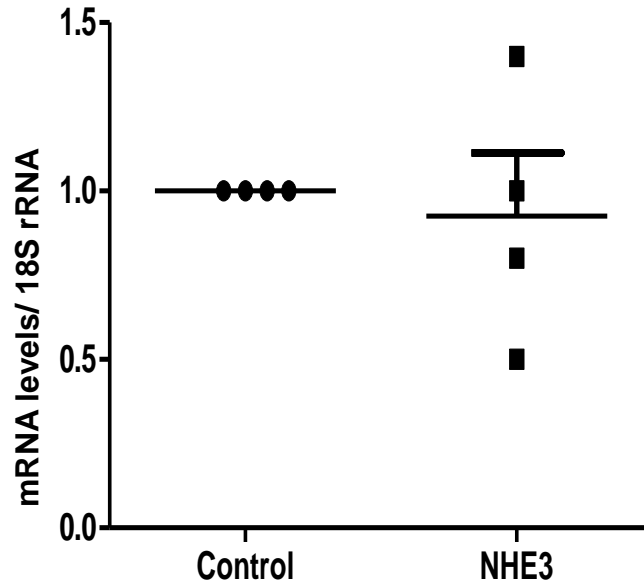
Control and SNX27shRNA containing HeLa cells were transfected with Ser49-A/D mutants and surface biotinylated 48h post transfection. Biotinylated proteins were captured from lysates with streptavidin beads at indicated time points after biotinylation and subjected to quantitative western blotting. The plots represent the mean of 3 independent experiments. Error bar indicates the SE of three experiments. \* $p < 0.05$  values are comparison between control, # $p < 0.05$  values are comparison between A mutant.

Fig. S3



**mRNA expression of core retromer protein is not affected by CT.** mRNA levels of Vps35, Vps26 and Vps29 were not affected by CT (48h) compared to control, as measured by qRT-PCR. Results are shown as mean values  $\pm$ SEM, n=4, p=NS compared to control.

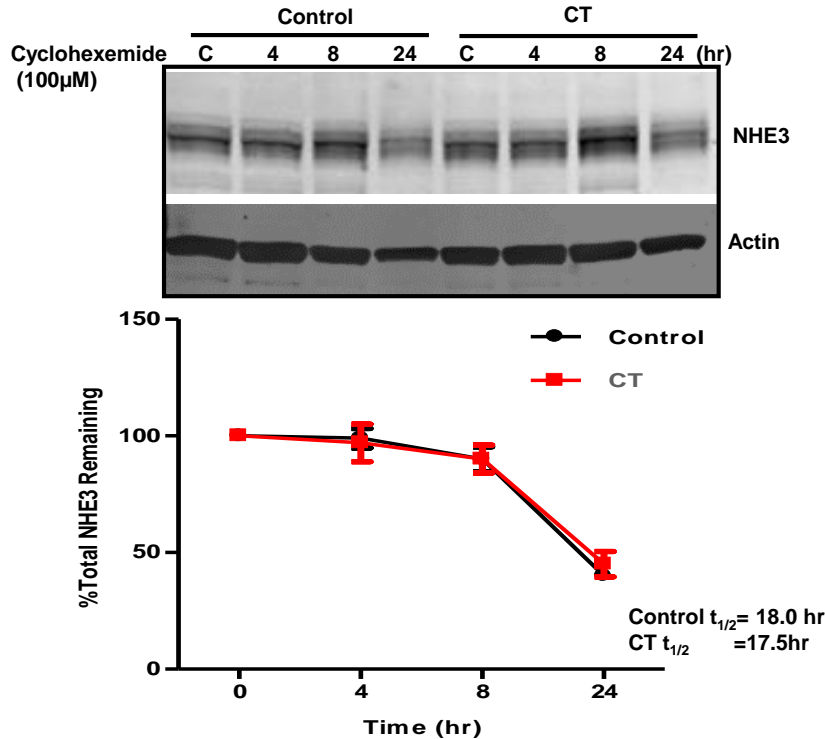
Fig. S4



**mRNA expression of NHE3 in response to R55.**

NHE3 mRNA levels were not affected by R55 treatment (48h) compared to control, as measured by qRT-PCR. Results are shown as mean values  $\pm$  SEM,  $n=4$ ,  $p=NS$  compared to control

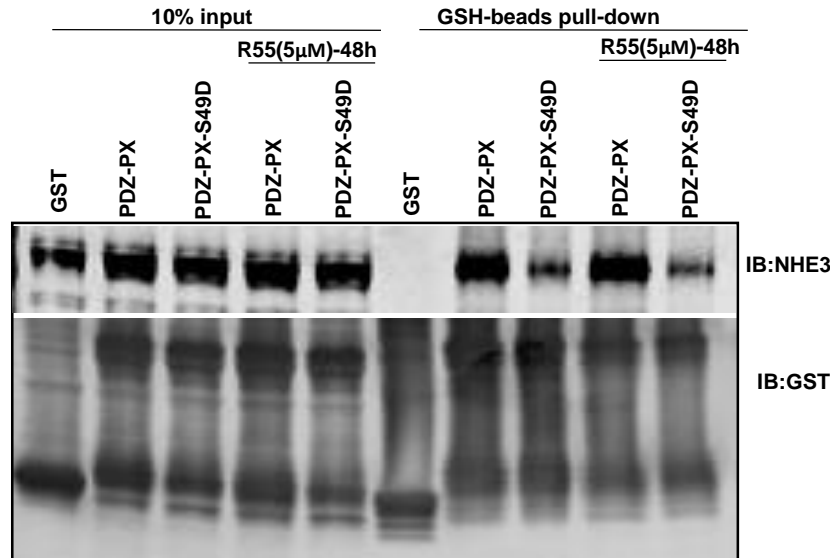
Fig. S5



### Half life of total NHE3 was not affected by CT.

Total NHE3 levels in control and CT treated cells were determined in the presence of cycloheximide (100µM) over the indicated periods. Densitometric analyses of Western blots were performed to calculate % of NHE3 remaining over indicated time periods. A representative blot for three independent experiments is shown. Results are mean  $\pm$  SEM for each time point. n=3. All points were NS compared to control.

Fig. S6



**Binding of NHE3 with SNX27Ser49D mutant was not affected by R55 .**

GST, GST-SNX27-PDZ-PX or GST-SNX27-PDZ-PX-Ser49 D fusion proteins were mixed with HEK-HA-NHE3 control or R55(5μM)-48h treated cell lysate and then subjected to pull-down with GSH resin. Samples were analyzed by Western blot (IB) with antibodies against NHE3 and GST. Experiment was repeated three times, and one representative result is shown .