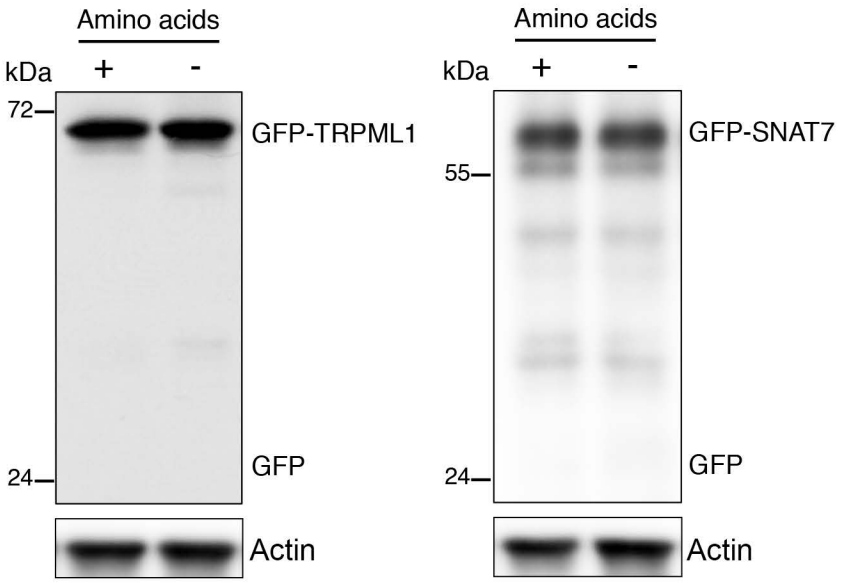
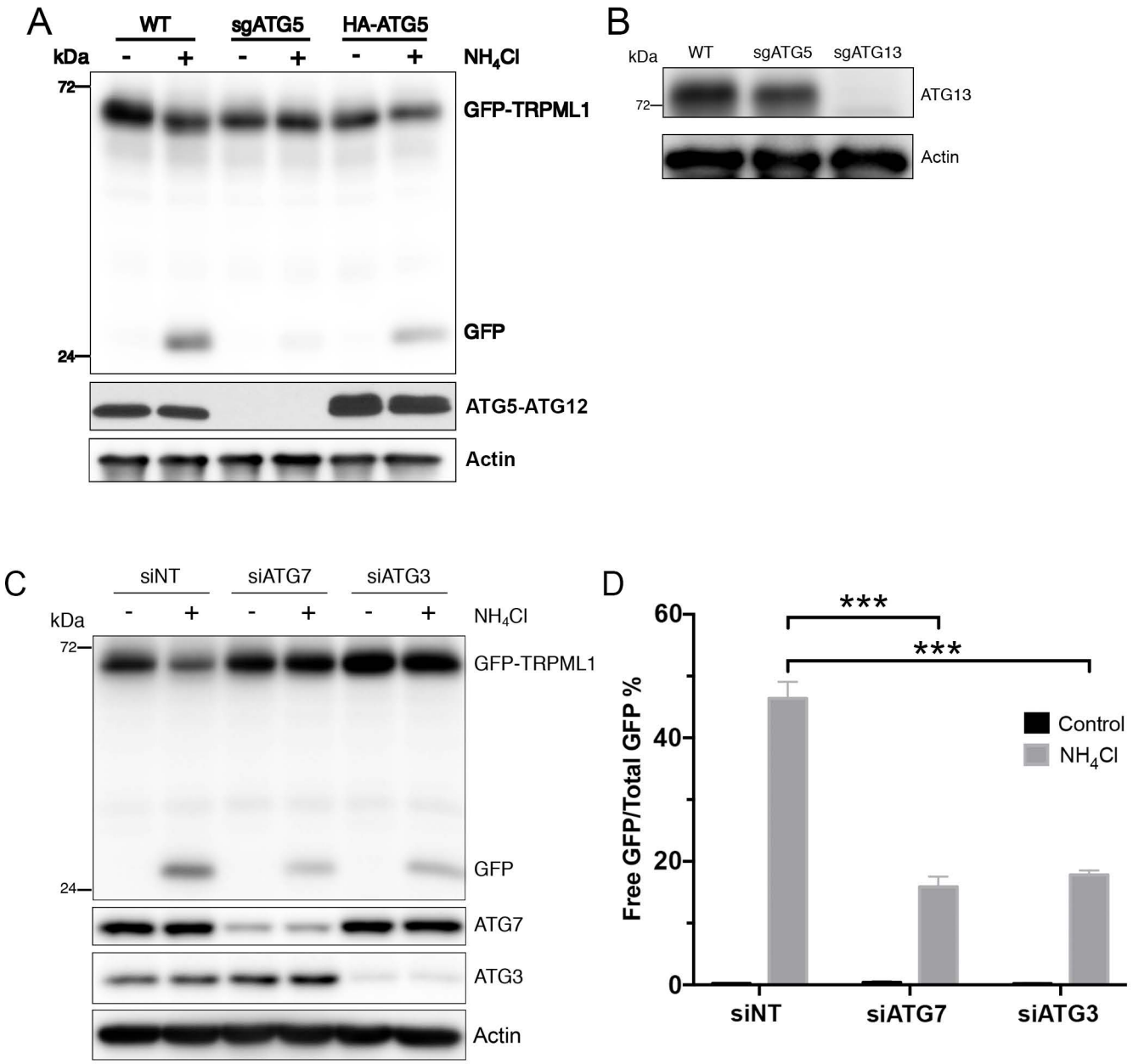
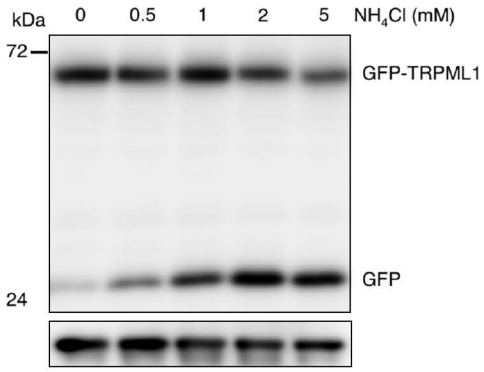


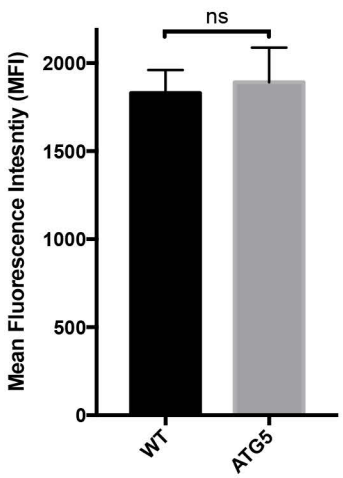
Supplemental Figure 1



Supplemental Figure 2



Supplemental Figure 3



Supplemental Figure 4

Supplemental Figure 1. Amino acid starvation does not induce the turnover of lysosomal transmembrane proteins, Related to Figure 1. Western blot shows GFP-TRPML1 (left) or GFP-SNAT7 (right) expression in cells cultured in full media or amino acid-free media for 24 hours. Actin loading controls are also shown.

Supplemental Figure 2. Expression of *ATG5* and *ATG13* in cells used in this study, Related to Figure 3. (A) Western blot shows ATG5-ATG12 conjugation in *WT* and *sgATG5* cells, and *sgATG5* cells with rescued expression of HA-tagged ATG5. Note that the turnover of GFP-TRPML1 induced by treatment with ammonium for eight hours is inhibited in *sgATG5* cells and rescued by expression of HA-ATG5. Actin loading controls are also shown. (B) Western blot shows knockout of *ATG13* generated by CRISPR/Cas9. Actin loading controls are also shown. (C) Blot show*s* GFP-TRPML1 cleavage induced by ammonium treatment in cells depleted of *ATG7 or ATG3* using siRNA. Actin loading controls are also shown. (D) Quantification of turnover of GFP-TRPML1 under the indicated conditions from the blot in part C. Data are from three independent experiments; error bars represent SEM. ***p<.001.

Supplemental Figure 3. Dose-dependent induction of GFP-TRPML1 turnover by treatment with ammonium for 18 hours at the indicated concentrations, Related to Figure 3.

Supplemental Figure 4. Lysosome function in *WT* and *sgATG5* cells, Related to Figure 4. Graph shows DQ-BSA fluorescence intensities quantified by flow cytometry from three independent experiments of *WT* and *sgATG5* cells cultured in full media.