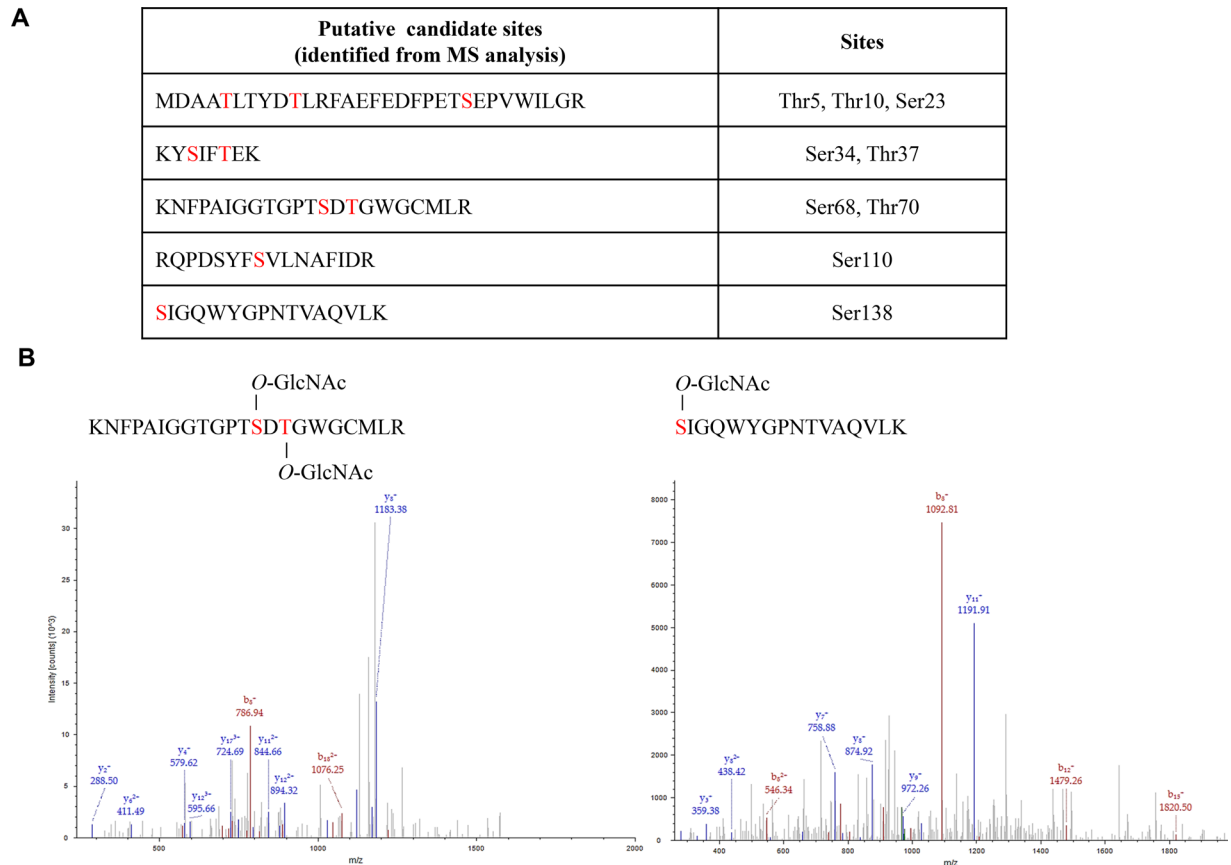
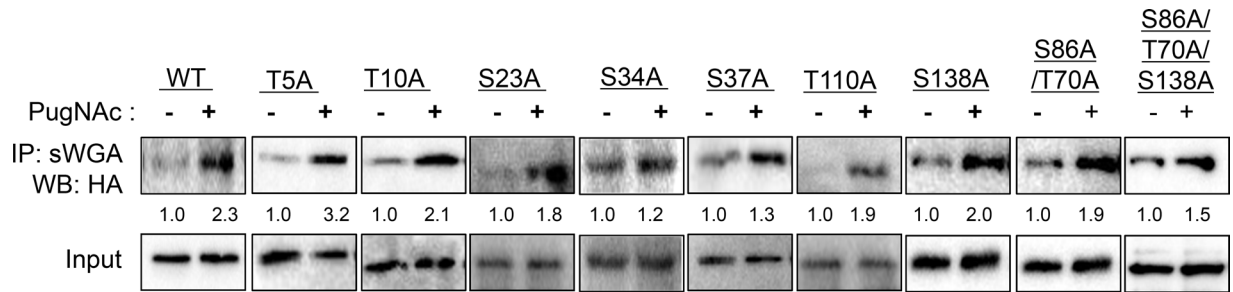


## O-GlcNAcylation of ATG4B positively regulates autophagy by increasing its hydroxylase activity

### SUPPLEMENTARY FIGURES



**Supplementary Figure S1: Mass spectrometric analysis of ATG4B protein.** **A.** List of putative candidate peptides of O-GlcNAcylation of ATG4B from mass spectrometric analysis. **B.** MS spectrum for the ATG4B fragmented peptide. SY5Y/HA-ATG4B cells were treated with PugNAc (100  $\mu$ M) for 24 h. HA-tagged ATG4B protein was purified by immunoprecipitation and subjected into mass spectrometric analysis described in Material and Method.



**Supplementary Figure S2: Treatment with PugNAc, an OGA inhibitor activates autophagy.** Wild type ATG4B (WT) and its mutants were overexpressed in SH-SY5Y cells, and the cells were treated with PugNAc (100  $\mu$ M) for 24h. *O*-GlcNAcylated proteins in the cells were pull-down with sWGA-agarose antibody, and the precipitated sWGA-complex was analyzed with Western blotting with anti-HA antibody.