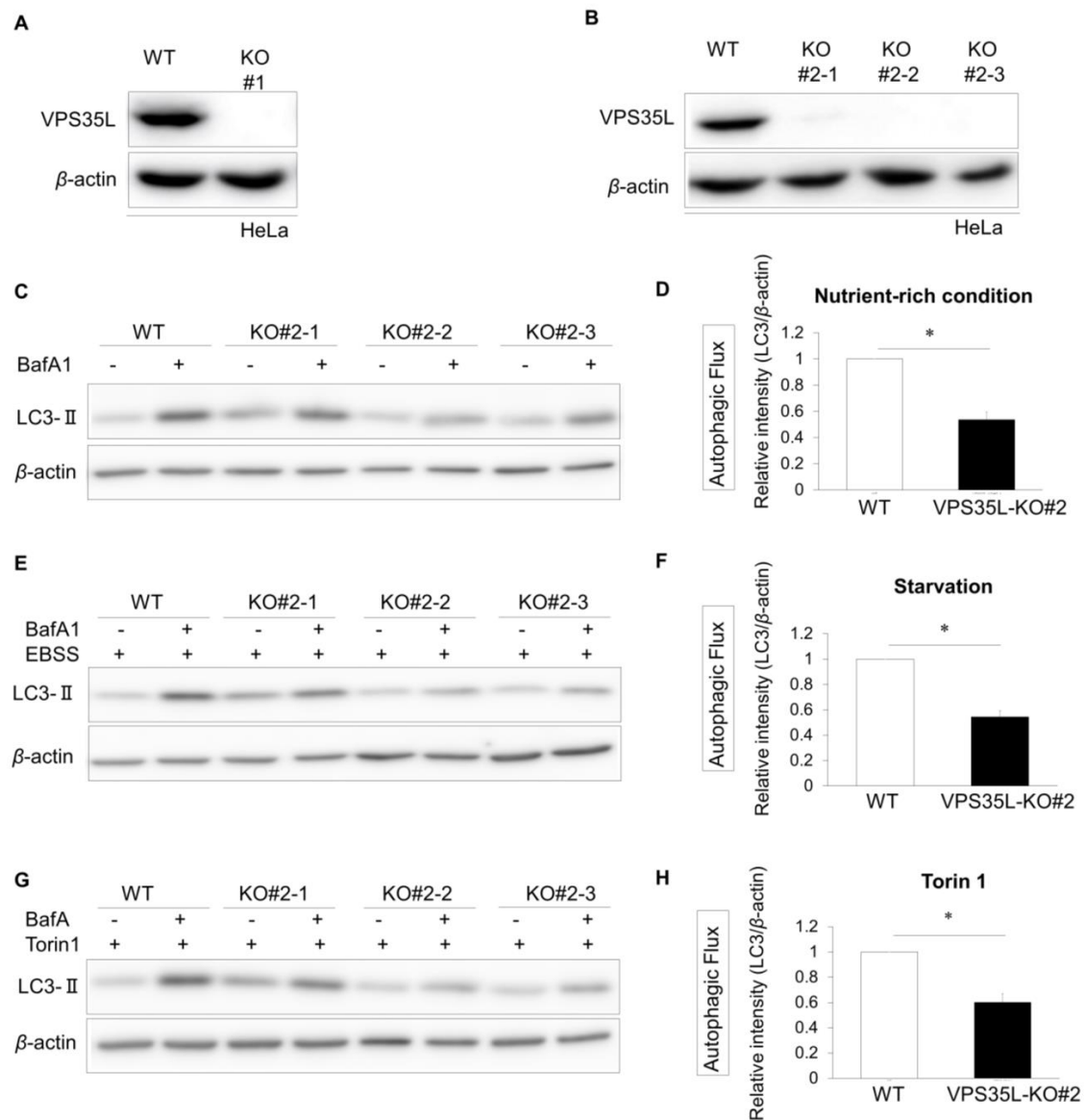


Supplementary Figure 2

**Suppl. Figure 2. Autophagic functions in VPS35L knockout cells.**

(A, B) Western blot analysis of VPS35L in VPS35L-KO cells. VPS35L protein was undetectable in knockout clones. (C)(E)(G) Representative immunoblots with anti-LC3 antibodies. The amount of LC3-II was quantitated as an indicator of autophagic vacuoles. The difference in LC3-II intensities between the presence and absence of BafA1 represent autophagic degradation activities (flux) during the assay period (2h). Cells were incubated in regular medium, EBSS (starvation condition), or 250nM Torin 1 with or without 125nM BafA1. (D)(F)(H) The LC3-II protein expression was quantified from three independent sets of immunoblots and normalized with β -actin. Significant decrease of autophagy flux was detected in the VPS35L-knockout cells under nutrient-rich condition (A, B), after starvation (C, D), and after treatment with Torin 1 (E, F). Means and s.e.m. are shown; * $P < 0.05$.