Supplemental Figure 2. No specific Lys of Ub is essential for the stimulation of proteolysis by S5a. Troponin I was ubiquitinated by MuRF1 and UbcH5 with WT or various Ub mutants where a single Lys was exchanged with Arg in the presence of 26S proteasomes. Addition of S5a (500 nM) at the outset enhanced hydrolysis of Troponin I, while Troponin I ubiquitinated without S5a was not degraded. Lane 1: Ubiquitination without 26S proteasomes and S5a; Lane 2: Ubiquitination and degradation with 26S proteasomes in the absence of S5a; Lane 3: Ubiquitination and degradation with 26S proteasomes in the presence of S5a.