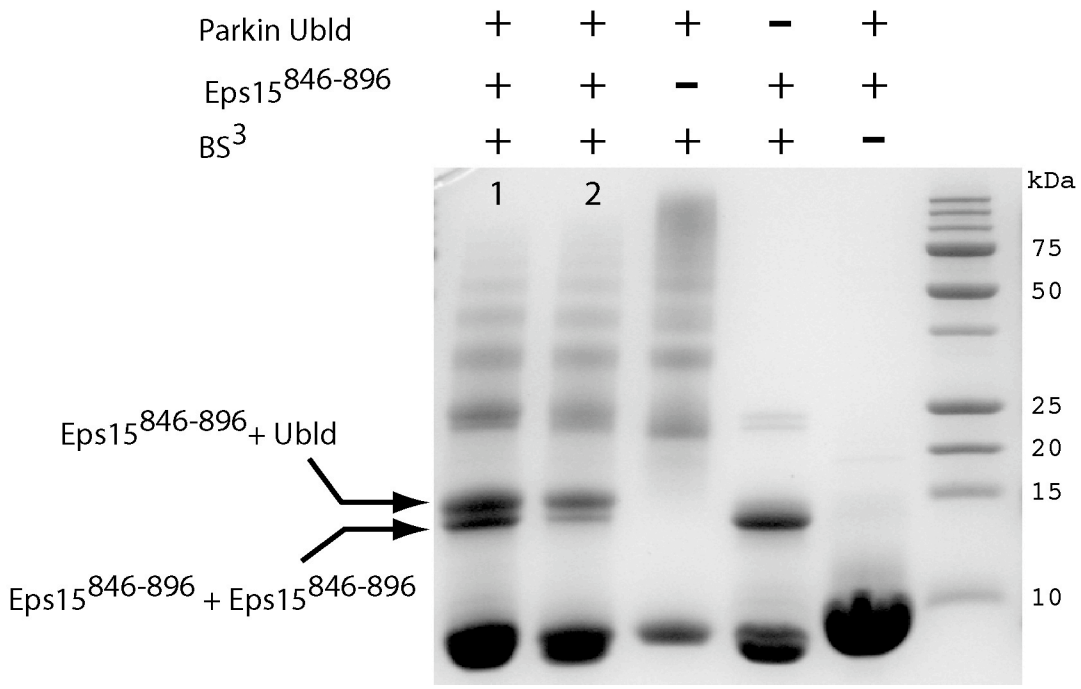
Safadi and Shaw Supplementary Figure 2.



Supplementary Figure S2. The stoichiometry of the Eps15$^{846-896}$ binding to the parkin Ubld is 1:1. Samples of Eps15$^{846-896}$ and the parkin Ubld were prepared in 10 mM KH_2PO_4, 1 mM TCEP, 150 mM NaCl at pH 7.0. The crosslinker BS^3 was also prepared in the same buffer. All proteins were prepared to a final concentration of 60 μM and the crosslinker to a 10-fold excess. Following cross-linking the samples were fractionated on a tricine gel and then stained with Coomassie dye. Lane 1 has a higher concentration of Eps15$^{846-896}$ than lane 2. The arrows indicate the proteins that make up that molecular weight band. Some higher molecular weight bands are observed as non-specific cross-linking of the parkin Ubld, and likely contains protein aggregates.