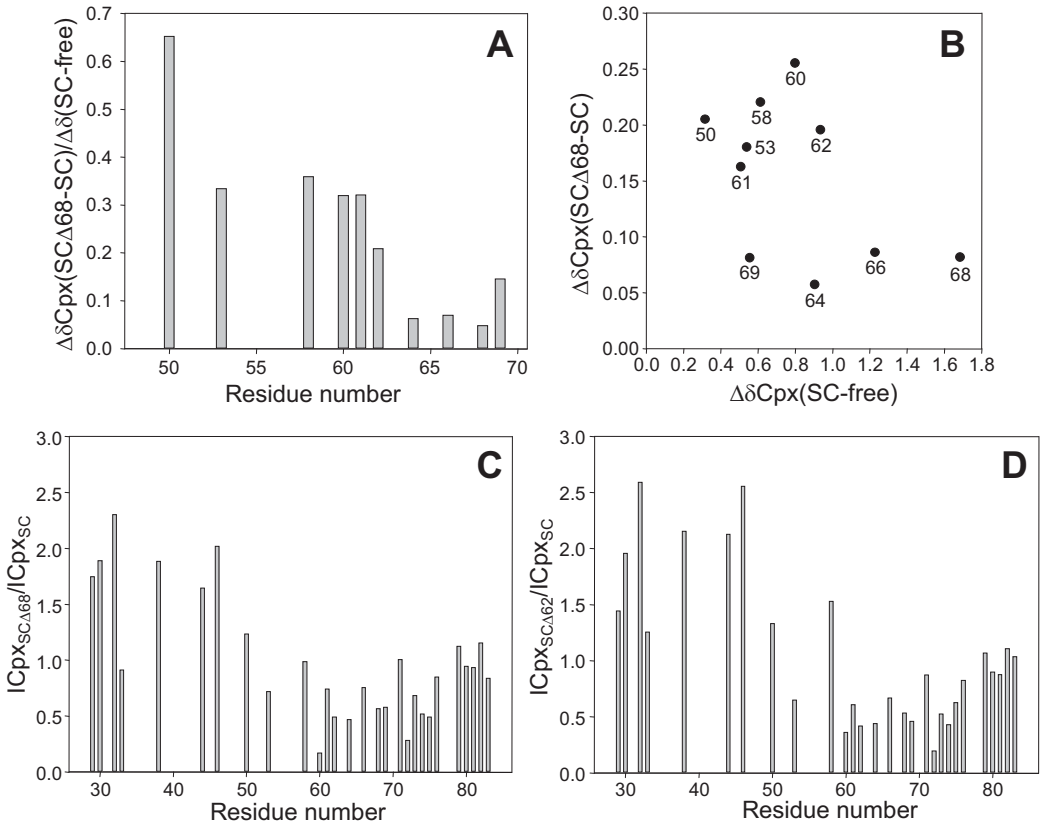

Figure 2 – figure supplement 1. Additional analysis of the interaction between ^{2}H,^{15}N-labeled CpxI fragments and synaptobrevin-truncated SNARE complexes.

A. Chemical shift changes in the CpxI central helix of SNARE complex-bound ^{2}H,^{15}N-CpxI(26-83) caused by truncation of synaptobrevin to residue 68, normalized by the changes caused by binding of ^{2}H,^{15}N-CpxI(26-83) to the SC. The chemical shift changes were calculated as $\Delta\delta = [(\Delta\delta HN)^2 + (0.17*\Delta\delta N)^2]^{1/2}$, where $\Delta\delta$HN and $\Delta\delta$N are the differences in HN and N chemical shifts, respectively, between the spectra being compared. For $\Delta\delta$Cpx(SCΔ68-SC), we compared ^{1}H-^{15}N TROSY-HSQC spectra of ^{2}H,^{15}N-CpxI(26-83) bound to SC and bound to SCΔ68. For $\Delta\delta$Cpx(SC-free), we compared ^{1}H-^{15}N TROSY-HSQC spectra of ^{2}H,^{15}N-CpxI(26-83) free and bound to SC. **B.** Plot of $\Delta\delta$Cpx(SCΔ68-SC) versus $\Delta\delta$Cpx(SC-free). **C,D.** Ratio between the intensities of cross-peaks of ^{1}H-^{15}N TROSY-HSQC spectra of ^{2}H,^{15}N-CpxI(26-83) bound to SCΔ68 (**C**) or SCΔ62 (**D**) vs those observed for ^{2}H,^{15}N-CpxI(26-83) bound to SC. To correct for small differences in protein concentrations, the cross-peaks intensities measured for each spectra were normalized with a correction factor derived by averaging the cross-peak intensities of the five C-terminal residues (residues 79-83), which were practically unaffected by the synaptobrevin C-terminal truncations. In all the plots shown in **A-D**, comparisons between chemical shifts or cross-peak intensities were made only for cross-peaks that could be identified in all the relevant spectra based on the assignments available for free and SNARE complex-bound Cpx(26-83) (Figures 2A,B) (Pabst et al., 2000; Chen et al., 2002) and the progressive movements caused by truncations in the SNARE complex (see Figure 2C).