SUPPLEMENTAL DATA

Supplementary Figure S1. Backbone assignment of the LD in VAMP7(1-118) and VAMP7(1-180). *A*. ¹H-¹³C strips of CBCA(CO)NH and HNCACB spectra are shown for the N-H chemical shifts corresponding to the first seven N-terminal amino acids of the VAMP7(1-118) construct. 14 strips are arranged in pairs (HN(CO)CACB on the left and HNCACB on the right) that are sequentially aligned. Each pair of strips was taken at the ¹⁵N plane corresponding to the amide ¹⁵N chemical shift of the residues indicated below the strips in red boxes. Black lines illustrate the connectivity between sequential C β (upper lines) and C α (lower lines), that produced the assignment of the resonances for the VAMP7a(1-7) region. *B*. ¹H-¹³C strips of HN(CO)CA and HNCA spectra are shown for the N-H chemical shifts corresponding to the first seven N-terminal amino acids of VAMP7(1-180). 14 strips are arranged in pairs (HN(CO)CA on the left and HNCA on the right) that are sequentially aligned. Each pair of strips was taken at the ¹⁵N plane corresponding to the amide ¹⁵N chemical shift of the residues indicated below the strips of HN(CO)CA and HNCA on the right) that are sequentially aligned. Each pair of strips was taken at the ¹⁵N plane corresponding to the amide ¹⁵N chemical shift of the residues indicated below the strips in red boxes. Black lines illustrate the corresponding to the amide ¹⁵N chemical shift of the residues indicated below the strips in red boxes. Black lines illustrate the connectivity between sequential C α .

<u>Supplementary Figure S2.</u> LD dimerization. *A*. Overlay of the ¹H, ¹⁵N-HSQC spectra of 18 μ M (black) and 450 μ M (magenta) VAMP7(1-118). For the 18 uM spectrum (black), the HSQC experiment was set to select only for NH resonances. Top left corner: summary of ¹H and ¹⁵N combined chemical shift changes in the VAMP7 LD induced by LD dimerization. The combined ¹H and ¹⁵N chemical shift changes are defined as $\Delta ppm = [(\Delta \delta_{HN})^2 + (\Delta \delta_N \times \alpha_N)^2]^{1/2}$, where $\Delta \delta_{HN}$ and $\Delta \delta_N$ are chemical shift differences of amide proton and nitrogen chemical shifts of the VAMP7 LD in the two different concentrations. The scaling factor (α_N) used to normalize the ¹H and ¹⁵N chemical shifts is 0.17. *B*. Elution profile of SEC-MALS of the isolated LD. Molar mass (red profile) and absorbance at 280 nm (black) (0.5 ml/min).





Supplementary figure S2