

Sample	Low-magnification two-dimensional cryoEM for docking			High-magnification cryoET for membrane morphology			
	Docked (percentage of all vesicles)	Undocked (percentage of all vesicles)	Total vesicles counted	Primed (percentage of docked vesicles)	Contact (percentage of docked vesicles)	Extended (percentage of docked vesicles)	Total docking sites
Time course 1min (Figure 3)	441(62.4%)	265(37.6%)	706	2(2.8%)	52(73.2%)	17(24.0%)	71
Time course 5min (Figure 3)	366(78.8%)	98(21.2%)	464	27(30.0%)	51(56.7%)	12(13.3%)	90
Time course 60min (Figure 3)	444(88.2%)	59(11.8%)	503	46(38.3%)	60(50.0%)	14(11.7%)	120
-Syt1	28 (7.8%)	334 (92.2%)	362	2(18.2%)	6(54.5%)	3(27.3%)	11
SUV:SUV (figure S4A)	Unable to assign	Unable to assign	Unable to assign	Unable to assign	Unable to assign	Unable to assign	Unable to assign
vSNARE GUVs: tSNARE SUV (Figure S4B)	257(62.2%)	156 (37.8%)	413	2(3.4%)	54(93.1%)	2(3.4%)	58

Table S3. Effect of incubation time and initial membrane curvature on vesicle docking and membrane morphology (raw data for figures 3 and S4).

For each reaction, two randomly selected grid squares were imaged at low-magnification and the number of SUVs in close proximity to a GUV (docked) or not (undocked) were counted (see supplementary methods). At a higher magnification, 5-16 tomograms were collected for each sample and the observed docking sites were classified as a “protrusion”, a “contact” or an “extended” membrane-membrane contact site (see supplementary methods for definitions). The first three rows of this table contain raw data used for Figure 3. This quantification was performed blind. The last two rows contain raw data for Figure S4. For the SUV:SUV fusion experiment, it was not possible to assign bona-fide docking sites in either the low-magnification pictures or in high-magnification tomograms due to the similarity of the vesicle sizes.