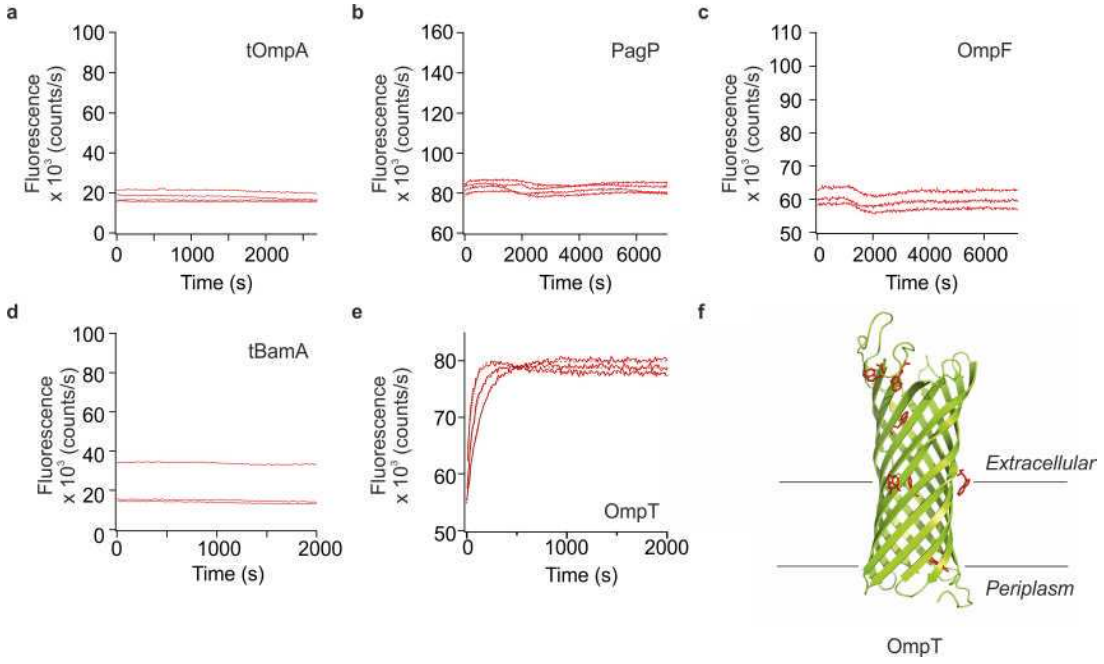
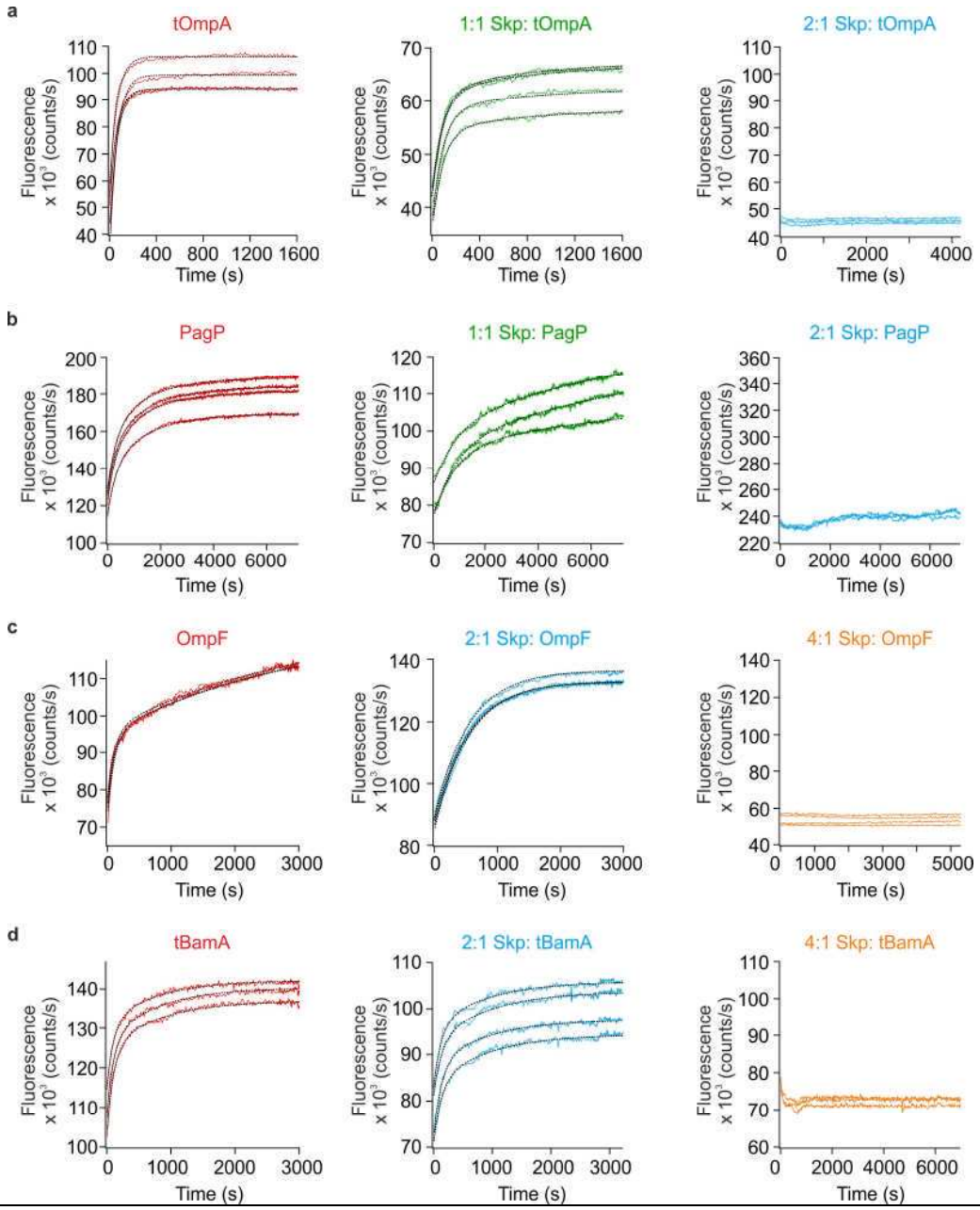


Supplementary Figure 1

OMP folding transients in the absence of lipids.

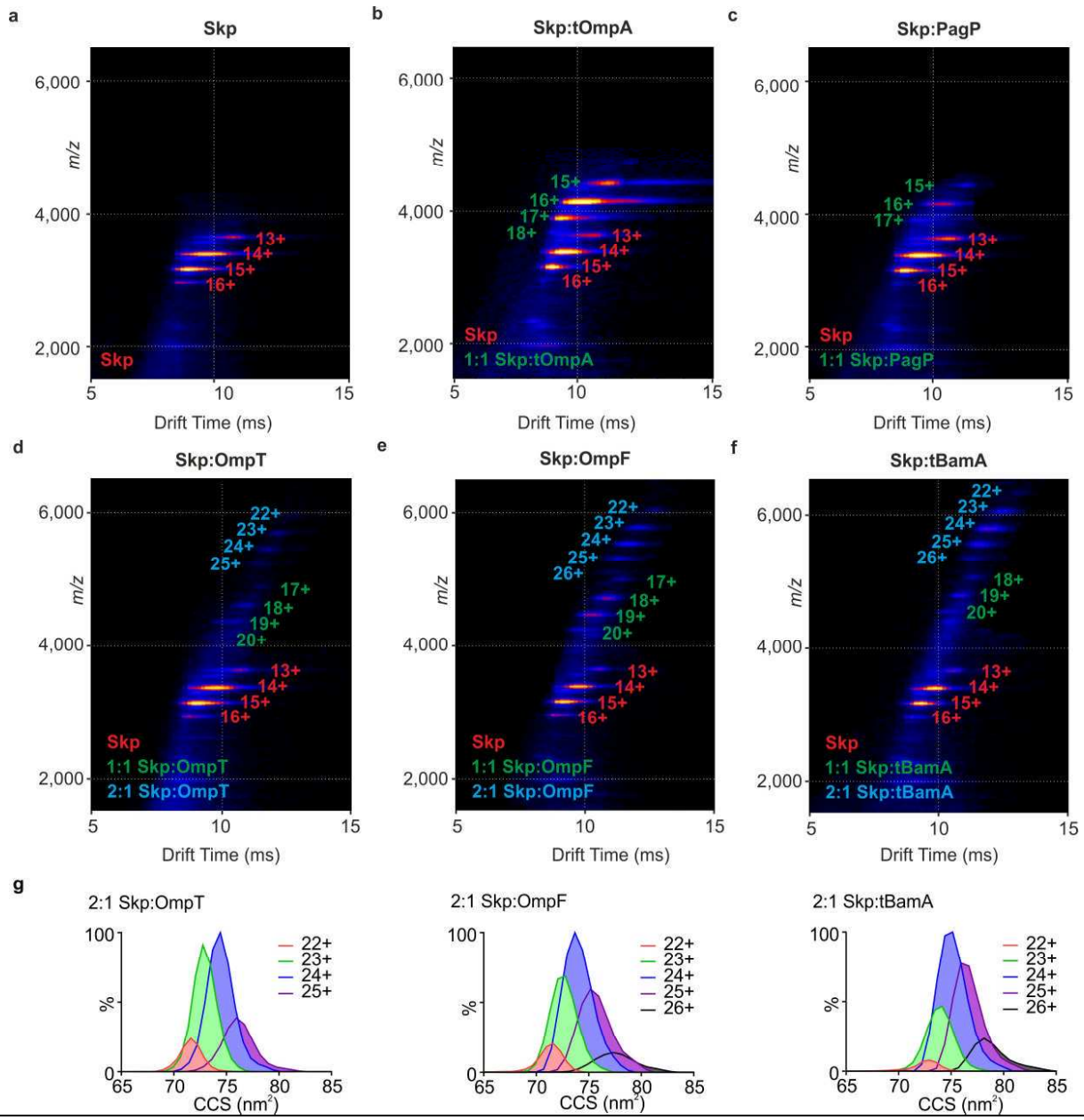
Example kinetic traces for **(a)** tOmpA, **(b)** PagP, **(c)** OmpF, **(d)** tBamA and **(e)** OmpT, in the absence of lipids, monitored by fluorescence emission spectroscopy. Assays were performed with OMP concentrations of 0.4 μM, in 0.24 M urea, 50 mM glycine-NaOH, pH 9.5, at 25 ºC. At least three replicates are shown for each protein. **(f)** Crystal structure of OmpT, PDB:1I78 (Vandeputte-Rutten, L. et al. *EMBO J* **20** 5033-9, 2001). Tryptophan residues are shown in stick representation and highlighted in red. The data for OmpT are well described by a single exponential indicated by black dashed lines.



Supplementary Figure 2

16-stranded OMPs require pre-incubation with a greater molar excess of Skp than 8-stranded OMPs to inhibit folding into synthetic liposomes.

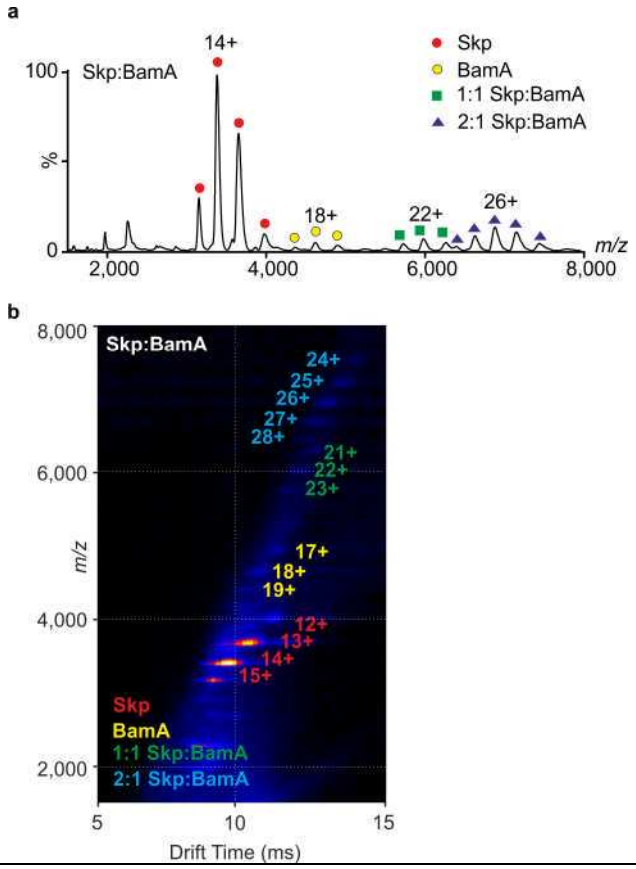
Example kinetic folding data for **(a)** tOmpA, left panel and tOmpA in the presence of a 1:1, centre panel, or 2:1, right panel, molar ratio of Skp:tOmpA; **(b)** PagP, left panel and PagP in the presence of a 1:1, centre panel, or 2:1, right panel, molar ratio of Skp:tOmpA; **(c)** OmpF, left panel and OmpF in the presence of a 2:1, centre panel, or 4:1, right panel, molar excess of Skp; **(d)** tBamA, left panel and tBamA in the presence of a 2:1, centre panel, or 4:1, right panel, molar excess of Skp. Pre-incubated Skp-OMP complexes were added to *di*$C_{11:0}$PC liposomes and OMP folding was monitored by fluorescence spectroscopy. Final OMP concentrations were 0.4 µM, with a molar lipid:protein ratio of 3200:1, in 0.24 M urea, 50 mM glycine-NaOH, pH 9.5. A minimum of three transients are shown in each panel. Single or double exponential fits to the data are indicated by black dashed lines (see Supplementary Table 2).



Supplementary Figure 3

Complexes of Skp with OMPs have variable stoichiometries revealed by ESI-IMS-MS.

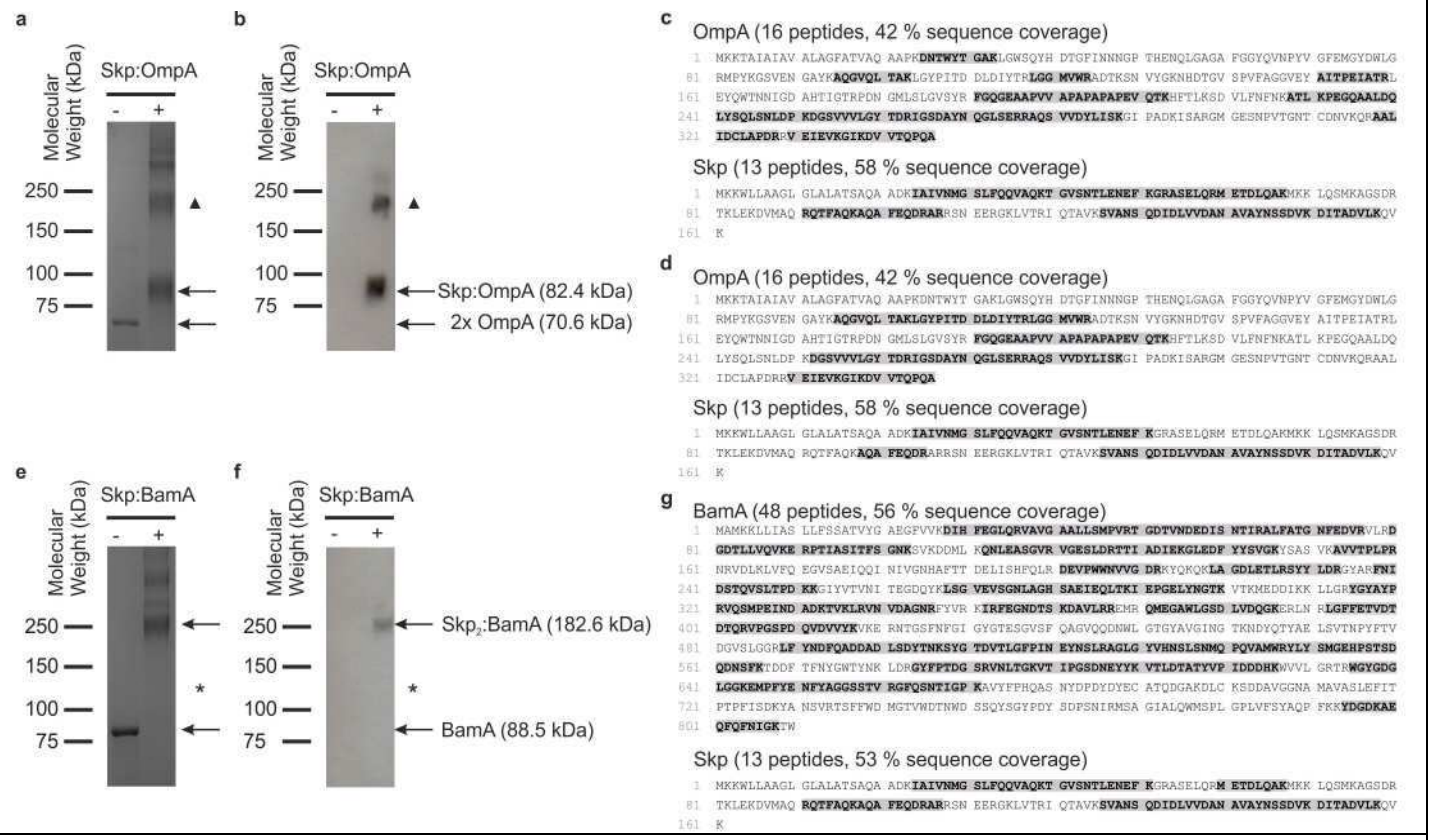
IMS driftscope plots of **(a)** Skp and Skp:OMP complexes with **(b)** tOmpA, **(c)** PagP, **(d)** OmpT, **(e)** OmpF or **(f)** tBamA. Peaks are labelled with their charge state. Charge states corresponding to Skp, 1:1 Skp:OMP and 2:1 Skp:OMP are labelled in red, green and blue, respectively. **(g)** CCS distributions (peak heights normalized to MS peak intensity) of 2:1 Skp:OMP complexes with OmpT (left), OmpF (middle), and tBamA (right) obtained from ESI-IMS-MS analyses. The charge state for each CCS distribution is indicated.



Supplementary Figure 4

ESI-MS shows that two copies of Skp bind to full-length BamA.

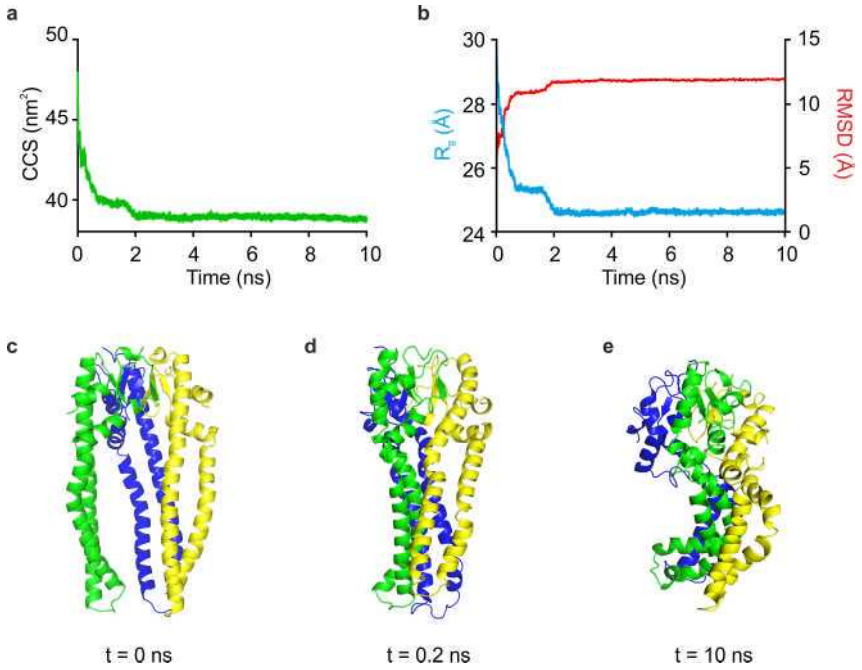
(a) Non-covalent ESI mass spectrum of full-length BamA binding to Skp. The spectrum is annotated with yellow circles (BamA), red circles (Skp), green squares (1:1 Skp:BamA) and blue triangles (2:1 Skp:BamA). The most abundant charge state in each distribution is labelled. **(b)** IMS driftscope plot corresponding to the mass spectrum in (a). Charge states corresponding to Skp, BamA, 1:1 Skp:BamA, and 2:1 Skp:BamA are labelled in red, yellow, green and blue, respectively. The ions at *m/z* ~2000 arise from Skp subunits.



Supplementary Figure 5

Chemical cross-linking and SDS-PAGE/western blotting/MS analysis of Skp:OMP complexes.

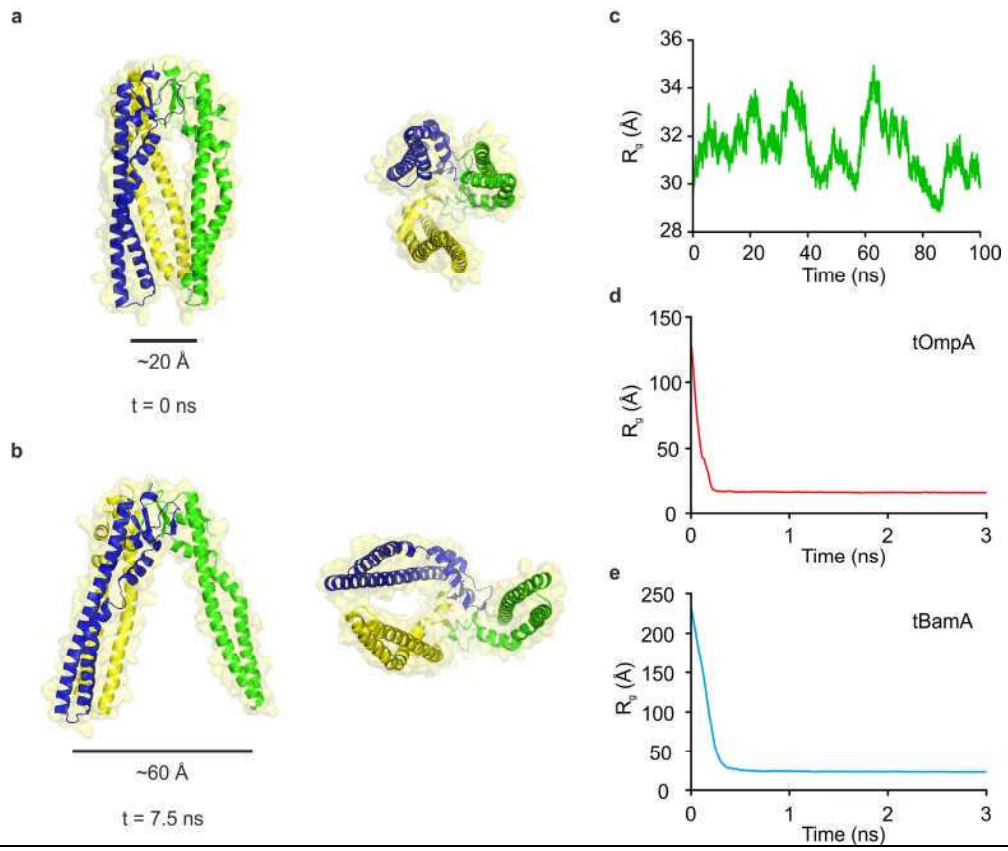
(a) SDS-PAGE analysis of a mixture of Skp and OmpA incubated without (-) or with (+) cross-linker. Upon addition of cross-linker a new band appears at approx. 85 kDa, consistent with a cross-linked Skp:OmpA complex (Skp:OmpA: 89.3 kDa, Skp: 54.0 kDa, OmpA: 35.3 kDa). An additional band (triangle) may correspond to $(Skp:OmpA)_2$ (178.6 kDa), likely due to dimerization via the periplasmic domain as previously observed (Marcoux, J. et al. *Structure* **22** 781-790, 2014). **(b)** Western blotting analysis with an anti-His antibody to confirm the location of His-tagged Skp-containing bands in the cross-linked Skp:OmpA sample. Higher molecular weight bands in the cross-linked sample (> 250 kDa) are therefore likely due to intermolecular OmpA cross-links. **(c-d)** Peptides identified from excised and digested bands from SDS-PAGE gels of cross-linked Skp:OmpA complexes consistent with **(c)** a 1:1 Skp:OmpA complex (the molecular weight band at ~90 kDa in **a,b**), **(d)** a larger Skp:OmpA complex (the molecular weight band labelled with a triangle in **a,b**). **(e)** SDS-PAGE analysis of a mixture of Skp and BamA incubated without (-) or with (+) cross-linker. Note that upon addition of cross-linker, no band appears at the expected mobility for a 1:1 Skp:BamA complex (*, 142.5 kDa). Instead, a single high molecular weight band is observed which corresponds to a higher order Skp:OMP assembly, consistent with the 2:1 Skp:BamA complex observed by ESI-MS (196.5 kDa). **(f)** Western blotting analysis with an anti-His antibody showing the location of His-tagged Skp-containing bands in the cross-linked Skp:BamA sample. Higher molecular weight bands in the cross-linked sample in (**e**) (> 250 kDa) are therefore likely due to intermolecular BamA cross-links. **(g)** Peptides identified from the excised and digested band of a cross-linked Skp:BamA complex. Regions of Skp or the corresponding OMP identified by database searching in (**c, d, g**) are denoted in bold with grey shading.



Supplementary Figure 6

Molecular dynamics simulations of Skp *in vacuo*.

(a) Theoretical CCS, and **(b)** backbone RMSD (red line) and radius of gyration (R_g) (light blue line) calculated for the initial 10 ns of a 100 ns molecular dynamics simulation in the gas-phase. **(c)** The starting model Skp structure used for the MD simulation (PDB: 1U2M (Walton, T.A. & Sousa, M.C. *Mol Cell* **15** 367-74, 2004), with missing residues in chains B and C modelled from chain A) (**Supplementary Data Set 1**). Skp subunits are colored green, blue and yellow. (d,e) Structures of Skp after a simulation time of **(d)** 0.2 ns and **(e)** 10 ns (**Supplementary Data Set 2**). The CCS values of the collapsed Skp structures after a simulation time of 10 ns and 100 ns (38.0 ± 1.8 nm^2 and 37.3 ± 1.9 nm^2, respectively) (see also **Supplementary Table 5**) agree favorably with the modal CCS of Skp at the lowest observed charge state (37.9 ± 0.6 nm^2) in native-MS experiments (**Fig. 4a**).



Supplementary Figure 7

Molecular dynamics simulations of Skp in solvent.

(a) Starting structure used for explicit solvent MD simulation of Skp (PDB: 1U2M3, with missing residues in chains B and C modelled from chain A), shown from the side (left) and bottom (right) (**Supplementary Data Set 1**). **(b)** Example structure of Skp in an 'open' conformation (t = 7.5 ns), shown from the side (left) and bottom (right) (**Supplementary Data Set 3**). **(c)** Radius of gyration of Skp over the course a MD simulation in explicit solvent. Structural collapse of initially extended chains of **(d)** tOmpA and **(e)** tBamA simulated with an implicit solvent model (see Online Methods).