Supporting Information

Biancalana et al. 10.1073/pnas.0912654107

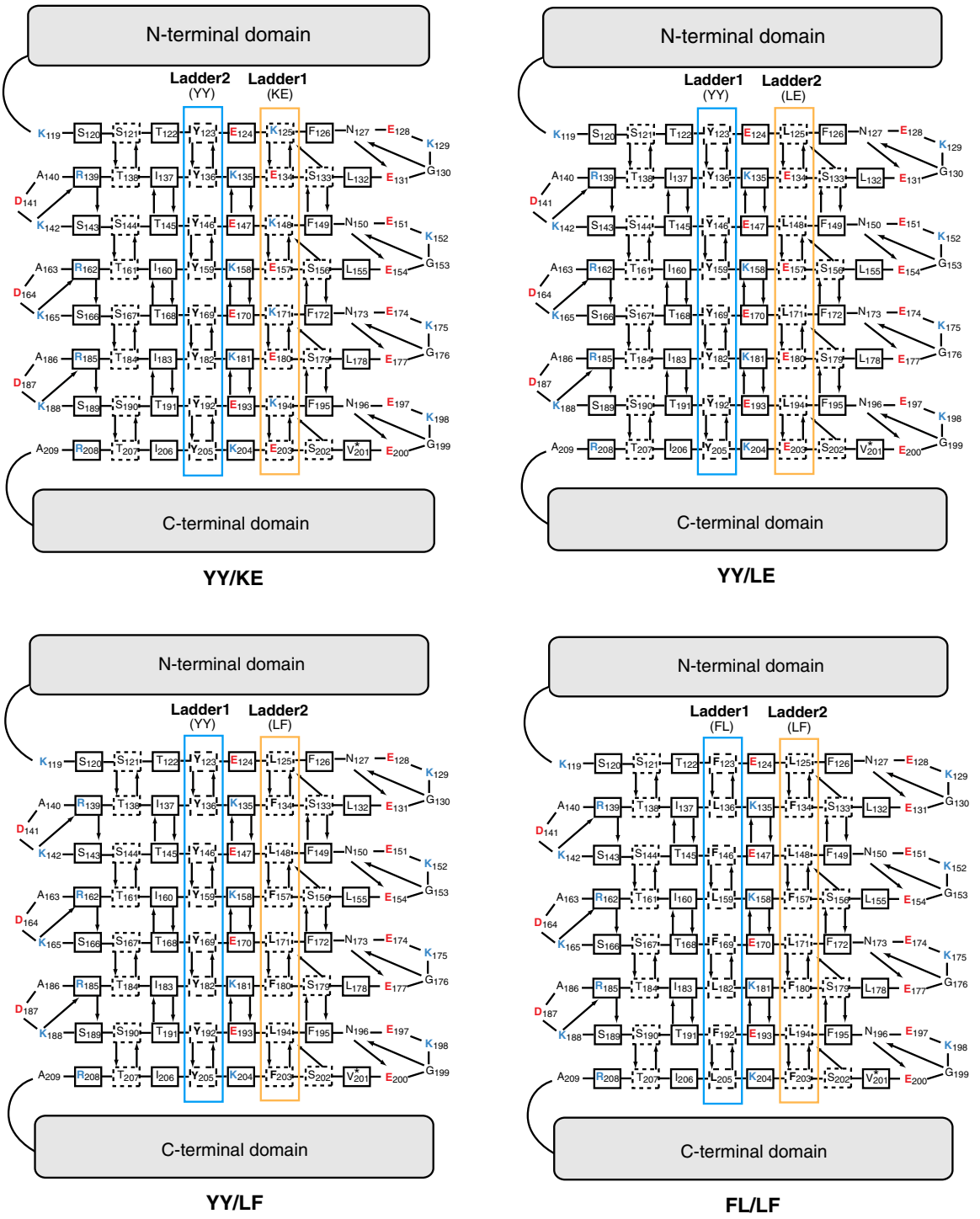


Fig. S1. Schematic drawings of the PSAMs described in this work. The amino acid sequences of the central β-sheet region are shown according to the β-sheet topology. Residues in boxes are those within β-strands. Residues in dashed line boxes have side chains protruding toward the reader. Hydrogen bonds in the wild-type OspA structure (PDB ID, 1OSP) are shown as arrows, with the arrowhead pointing to the carbonyl group. The N- and C-terminal domains correspond to residues 27-118 and 142-273, respectively of the surface engineered OspA (54)

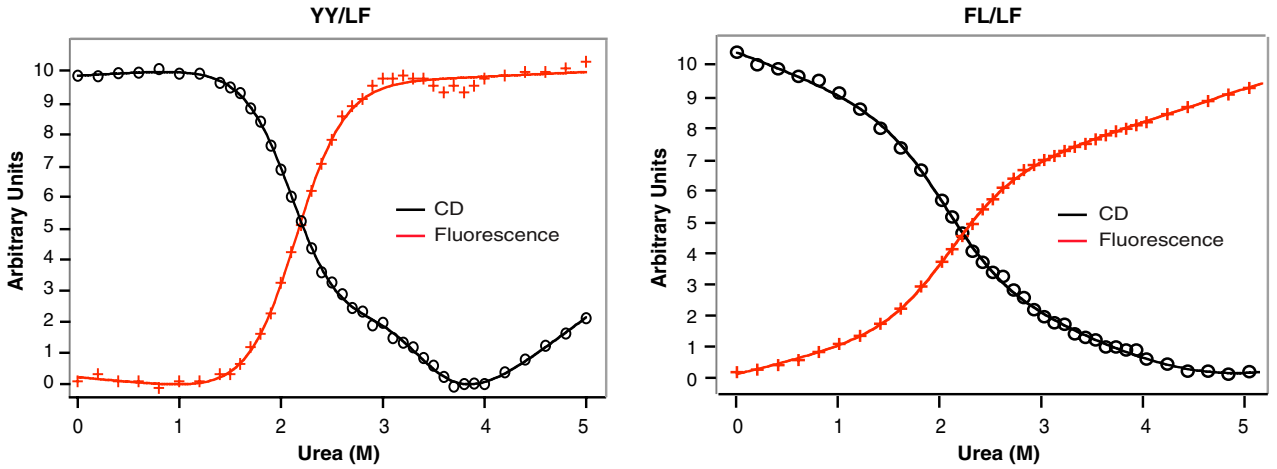


Fig. S2. Stability of YY/LF (*Left*) and FL/LF (*Right*) cross-β PSAMs measured by urea-induced unfolding. Circular dichroism ellipticity at 235 nm (*Black*) and Trp fluorescence emission at 290 nm (*Red*) are plotted as a function of urea concentration. The intensities were normalized from 0 to 10. The curves show the best global fits of the three-state model (Native–Intermediate–Unfolded)

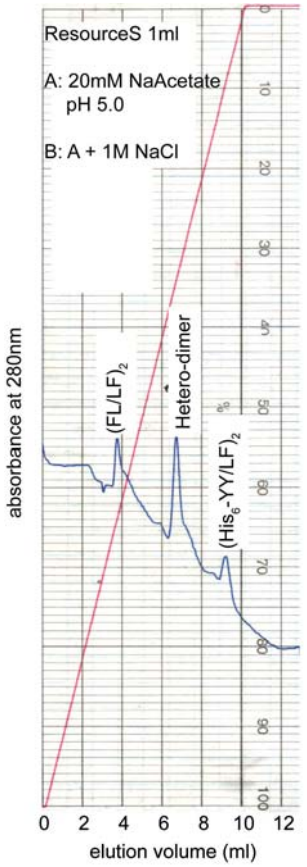


Fig. S3. The formation of the heterodimer of YY/LF and FL/LF. The sample was prepared by mixing equal amounts of FL/LF lacking a His-tag and YY/LF containing a His-tag in the presence of urea. Refolding was performed by dilution into 10 mM phosphate buffer, pH 6.0 with 150 mM NaCl. The refolded sample was concentrated and analyzed using cation exchange chromatography (SP-Sepharose, Amersham) in 20 mM sodium acetate buffer, pH 5.0 with a sodium chloride gradient. The identities of the three peaks are indicated.

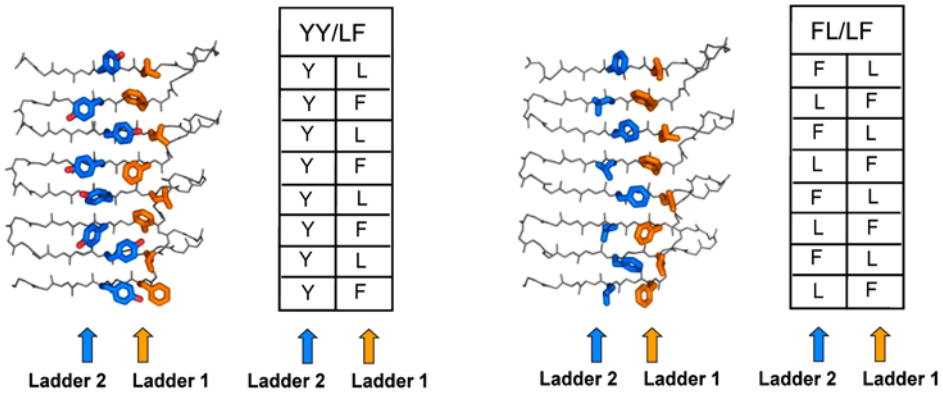


Fig. S4. Structure of the designed ladders of YY/LF (*Left*) and FL/LF (*Right*), depicted in the same manner as in Fig. 1C.

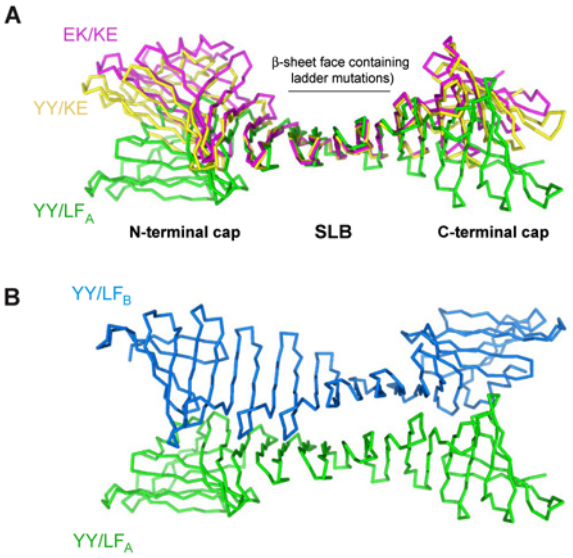
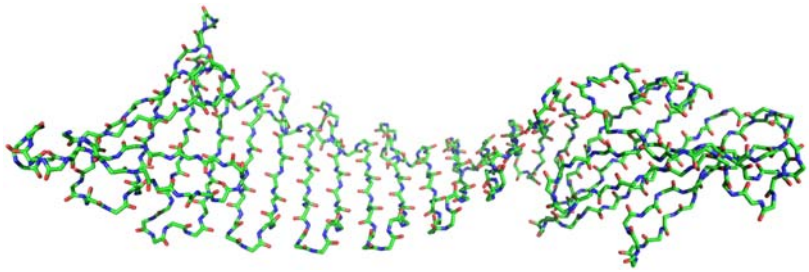


Fig. S5. Hinge-like bending of the capping domains in the cross-β PSAMs. (*A*) EK/KE (monomer), YY/KE (monomer), and subunit A of YY/LF (dimer) aligned using the SLB of the PSAM scaffold. Structures are shown as ribbons. (*B*) Both subunits A and B of the YY/LF dimer shown in same relative orientation as in *A*. Note the designed ladders are located at their dimer interface.

Table S1. Data collection and refinement statistics for YY/LF and FL/LF

	YY/FL	FL/LF
PDBID	3CKA	3EEX
	Data collection statistics	
Space group	$P2_12_12_1$	$P2_1$
Cell parameters	a=75.29	a=36.50
	b=83.65	b=82.40
	c=106.1	c=109.4
	$\alpha=\beta=\gamma=90.0$	$\beta=91.6$
Beamline	APS-23ID-B	APS-23ID-B
Wavelength	0.9793 Å	0.9793 Å
Resolution (Å) (highest resolution shell) [a)]	50-1.65(1.71-1.65)	50-2.50(2.59-2.50)
Completeness(%)	98.8(100)	99.0(100)
I/s(I)	22.2(2.50)	13.2(2.33)
R_{merge} [b)]	0.074(0.780)	0.094 (0.616)
Average redundancy	22.2(5.90)	3.10(3.10)
	Refinement statistics	
Resolution range (Å)	20.0-1.65	20.0-2.49
Reflections used (free)	77805(4100)	20097(2264)
R_{factor} [c)]	0.202	0.261
R_{free} [d)]	0.240	0.299
RMS deviations		
Bonds (Å)	0.020	0.013
Angles (°)	1.833	1.559
No. protein residues	315	315
No. waters	524	0
Average B factor ($Å^2$)	21.52	38.19
	Ramachandran plot statistics	
Most favored (%)	89.5	85.9
Additionally allowed (%)	10.0	12.2
Generally allowed (%)	0.5	1.0

Crystals were obtained at 19 °C using the hanging drop vapor-diffusion method. Crystallization conditions were as follows: 30% PEG400 and 0.1 M Tris-HCl pH 9.0 for YY/LF (10 mg/mL protein with 4 mM Thioflavin-T); 34% PEG400 and 0.1 M Tris-HCl pH 9.0 for FL/LF (20 mg/mL protein). Paratone oil was used for cryo-protection. Data collection and processing as well as structure determination were performed as described previously (29). Molecular replacement was performed using the YY/KE scaffold (the Protein Data Bank entry 2oY7) as a search model. The model was split into N- and C-terminal halves (residues 28–167 and 168–342, respectively) and used simultaneously.



Movie S1. Conformational rearrangement of cross-β PSAMs. An animated sequence of interpolated structures between the starting YY/EK (monomeric) and YY/LF (dimeric) PSAMs demonstrate the dramatic conformational changes occurring upon lamination of the central β-sheet region.
Movie S1 (MOV)