Supporting Information

Dao et al. 10.1073/pnas.1412165112

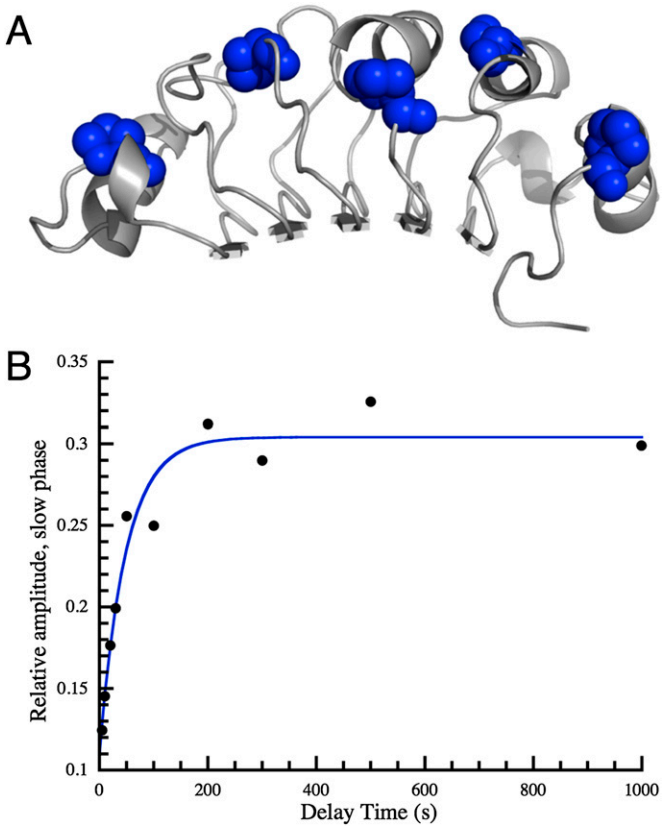

Fig. S1. *Cis–trans* prolyl isomerization during the refolding of PP32. (*A*) Ribbon representation of PP32. Residues P16, P60, P88, P108, and P140 are shown in blue spheres. (*B*) Double-jump refolding assay for PP32. PP32 was unfolded in 4 M urea, and was refolded after various delay times by dilution to a final urea concentration of 2 M. The relative amplitude is the fitted fluorescence change associated with the slow phase compared with the total refolding amplitude at each delay time. Line results from fitting a single-exponential model to the refolding amplitude.

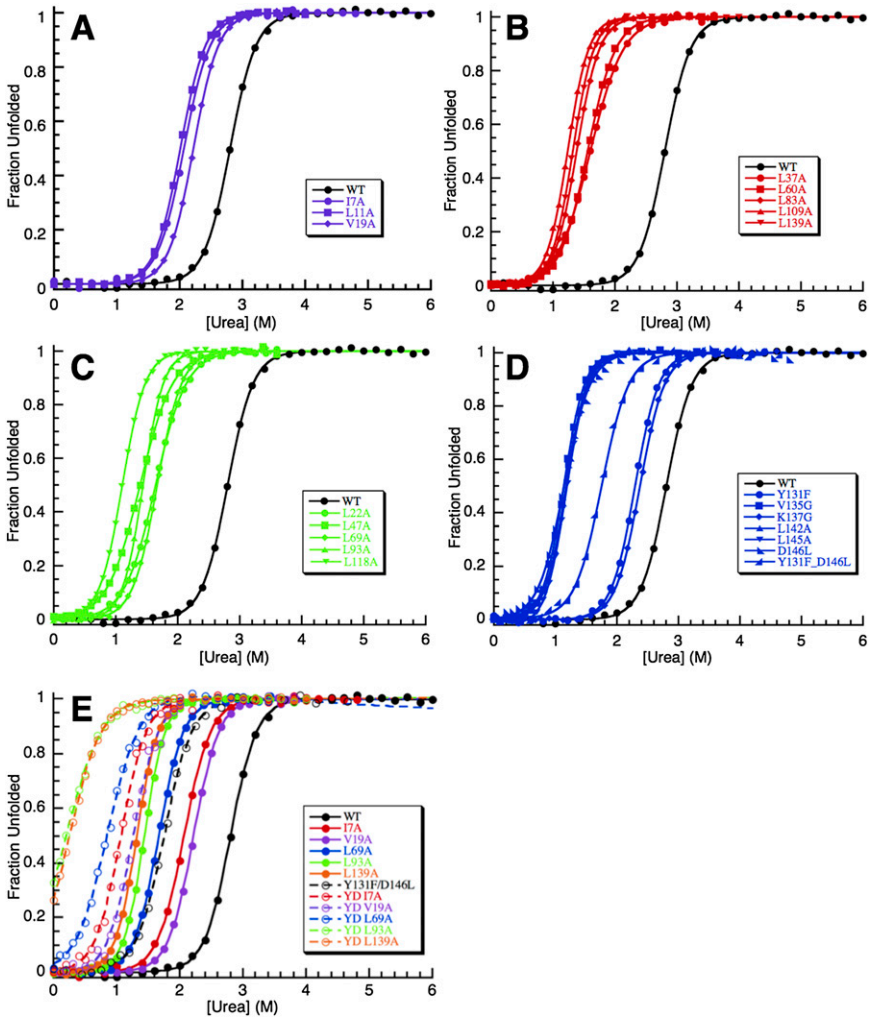

Fig. S2. Urea-induced equilibrium unfolding of PP32 variants. Transitions are monitored by far-UV CD at 220 nm. Lines result from fitting an equilibrium two-state unfolding model to the data. In each panel, WT PP32 is shown in black. (*A*) N-cap variants (purple), (*B*) conserved convex side L→A variants (red), (*C*) conserved β-sheet L→A variants (green), (*D*) C-cap variants (blue), and (*E*) a comparison between variants in WT (closed symbols) and YD (open symbol) backgrounds. The wild-type unfolding transitions, along with a subset of the single variant unfolding transitions (in panels *B*, *C*, and *D*), are adapted from ref. 1.

1. Dao TP, Majumdar A, Barrick D (2014) Capping motifs stabilize the leucine-rich repeat protein PP32 and rigidify adjacent repeats. *Protein Sci* 23(6):801–11.

Table S1. Fitted values for the Gaussian cosine equation

Parameter	Fitted value	Confidence intervals
a	1,510	900–2,120
μ	114 residues	91–139
σ	104 residues	60–149
b	−0.72	−1.1 to −0.38
n_{rep}	22.6 residues	21.6–23.6
c	−8.7 residues	−13.4 to −4.0

Parameters are as defined in Eq. **1** in main text. Parameters were fit using nonlinear least-squares optimization in Mathematica. Protection factors from the N-terminal α-helical cap were not included in the fit.