Supporting Information

Free Energy Landscape of Lysozyme: Multiple Near-Native Conformational States and Rollover in the Urea Dependence of Folding Energy

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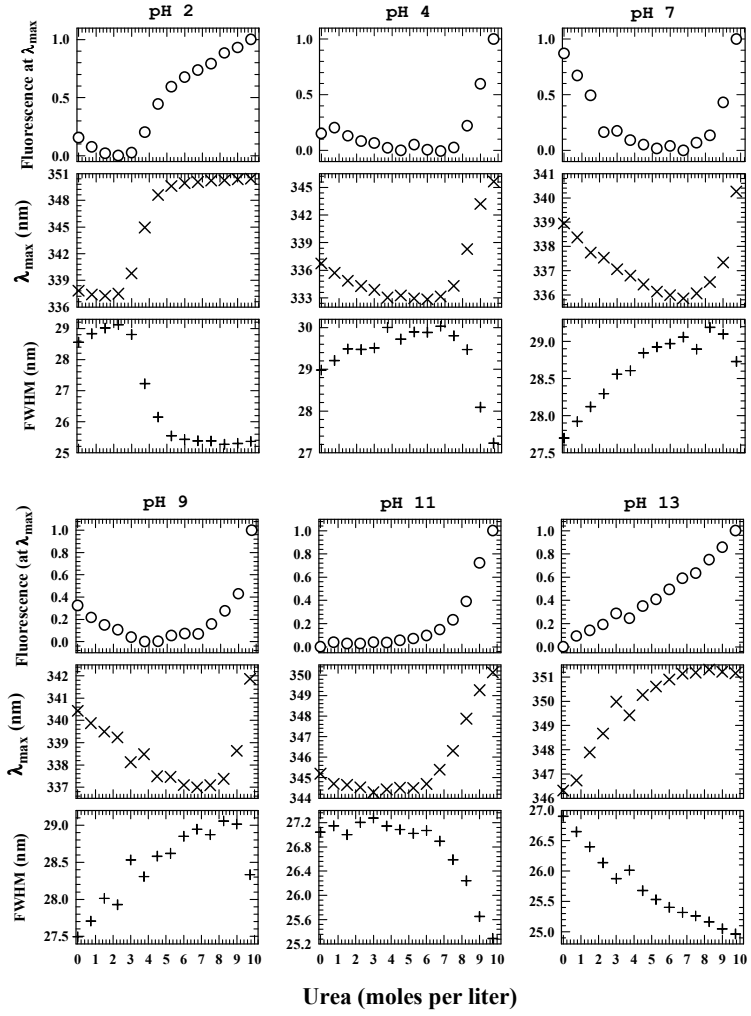

Figure S1. Urea dependence of tryptophan fluorescence intensity, emission wavelength maximum, and the full width at half maximum (FWHM) of the emission spectrum at indicated pH values. Incomplete unfolding at pH 4, 7, 9, and 11 within the urea solubility limit is evident.

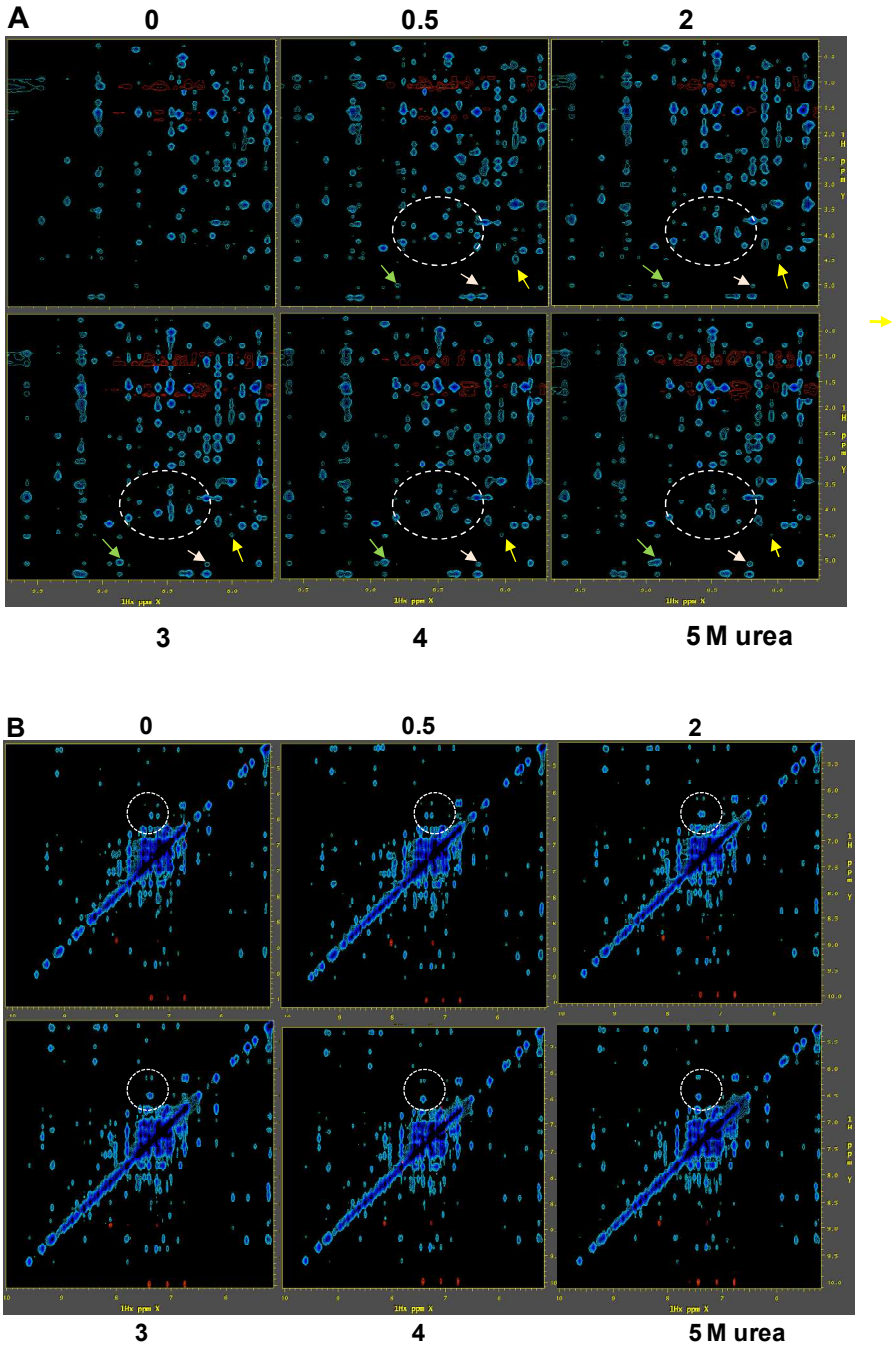

Figure S2. Screen shots of regions of NOESY spectra of lysozyme at indicated urea concentrations, pH 5, and 25°C. The crowded aliphatic region is not shown. Some of the minor changes at the indicated subdenaturing concentrations of urea are shown by circled arrowheads. .

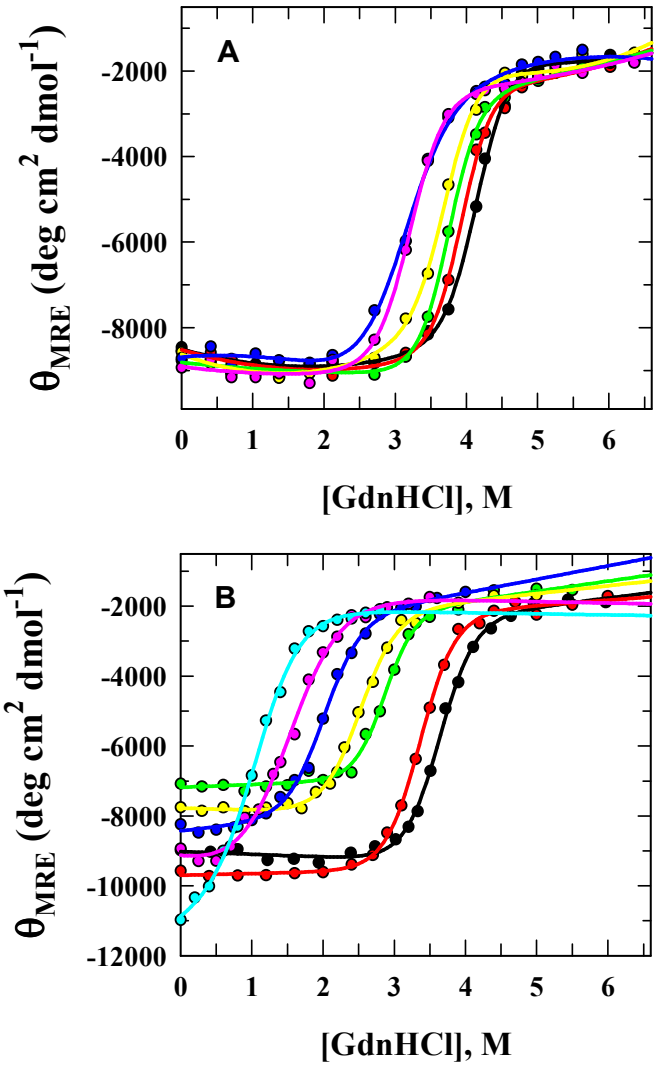

Figure S3. Far-UV monitored GdnHCl-induced equilibrium unfolding of lysozyme at different concentrations of urea, 20 mM sodim acetate, pH 5, 25°C. (*A*) 0 M (●), 0.55 M (●), 1.1 M (●), 2.3 M (●), 2.7 M (●), and 2.8 M (●) urea. (*B*) 1 M (●), 2.1 M (●), 3.2 M (●), 4.2 M (●), 5.0 M (●), 5.1 M (●), and 7.3 M (●) urea. The fits through data are according to the two state (N⇌U) transition assuming linear dependence of ΔG on GdnHCl molarity.