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Three-Pulse Photon Echo Peak Shift (3PEPS) as a probe of conformational distribution in heme protein folding ZHAOCHUAN SHEN, University of Colorado at Boulder, EMILY GIBSON, University of Colorado at Denver, RALPH JIMENEZ, University of Colorado at Boulder — Line broadening of electronic spectra of cofactor consists of two contributions: homogeneous broadening and inhomogeneous broadening. Inhomogeneous broadening reflects conformational diversity of the cofactor and its surrounding environment and is a key to quantifying the disorder of soft condensed matter system. To clarify the relation between contributions to the lineshapes and protein conformation, we investigated the equilibrium unfolding sample of Zn-cytochrome c (Zn-cyt c) in guanidine hydrochloride (GuHCl). Soret band UV-vis spectra were first measured as a function of GuHCl, and fitted with both two- and three- state models of protein folding. Then, we measured 3PEPS signal of the folded, midpoint and unfolded samples. By tuning the laser wavelength over Soret band on midpoint sample and comparing experimental asymptotic peak shift with simulated from two- and three-state models, we attributed folding of Zn-cyt c to a two-state model. This is a novel approach in understanding protein folding and should be applicable to investigate of other proteins.

Zhaochuan Shen University of Colorado at Boulder

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