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Protein folding intermediates probed by ensemble of their transient stiffnesses in single-molecule force-quenched AFM¹ ROBERT SZOSZKIEWICZ, KATARZYNA MALEK, Kansas State University — By using force-quench AFM (FQ-AFM) spectroscopy molecular structures with transient stiffnesses are detected during folding of a recombinant protein with four I27 molecules linked in tandem. The intermediate stiffnesses are detected from shape and peaks of the autocorrelation of fluctuations in end-to-end lengths of the folding molecules, as well as by applying the equipartition theorem to the FQ-AFM experimental results. In the light of the relevant molecular dynamics simulations these intermediates are likely to probe the ensemble of random-coiled collapsed states present both in the force-quench and thermal-quench folding pathways.

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