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Hand-Over-Hand Binding in a Tethered-Particle Method Study of DNA Hybridization MARIE T. UNG, JOHN C. CROCKER, University of Pennsylvania — The tethered-particle method is used to probe the complex process of DNA hybridization. This experiment analyzes tethers formed between PEGylated polystyrene microspheres and PEGylated glass surfaces, to which DNA strands have been grafted. Previous studies have looked at single duplex dissociation in isolation. We seek to understand cooperativity effects of short duplexes in multiple tethered states. This experiment is performed near the duplex melting temperature, resulting in dynamic binding and unbinding. Single molecular tethers are analyzed using high performance particle tracking with a high-speed camera. By analyzing 2D trajectories of the particle's centroid versus time, we can distinguish single binding and unbinding events from multiple bridged states. The trajectories' analysis suggests that multiple bridged states are less stable than expected for a zero cooperativity model. We hypothesize that this instability may be due to extreme force sensitivity of individual DNA duplexes.

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