

Accessing the Low-Force Regime: Magnetic Tweezers Reveal Early Activation Steps in von Willebrand Factor

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Proteins are frequently regulated by mechanical forces, for example in hemostasis, cell adhesion, and mechanotransduction. In many cases, functionally relevant conformational transitions occur in the low-force regime (≤ 1 pN), which remains difficult to access with high precision. The blood coagulation protein von Willebrand Factor (VWF) is an important example, undergoing force-induced structural transitions that regulate its adhesive function in the bloodstream.

Here, we use single-molecule magnetic tweezers to resolve low-force transitions associated with early activation steps in VWF and quantify how disease-associated mutations reshape these mechanical equilibria. We directly show that the clinically relevant F2561Y mutation selectively destabilizes the VWF stem, shifting its opening transition to substantially lower forces (from ~ 1.1 pN in the wildtype to ~ 0.1 pN) while leaving higher-force domain unfolding unaffected.

These findings provide a direct molecular link between low-force mechanics and pathological function. More broadly, they highlight the potential of magnetic tweezers to quantitatively probe force-dependent conformational changes in multidomain and mechanosensitive proteins, opening new opportunities to investigate how mechanical perturbations regulate protein function across diverse biological systems.