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Multiple attachments, heterogeneous binding and the high force tail in protein-protein binding force histograms ANWESHA SARKAR, EDWARD KRAMKOWSKI, Wayne State University, ESSA MAYYAS, Henry Ford Hospital, PETER M. HOFFMANN, Wayne State University — Atomic Force Microscopy (AFM) is a useful tool in measuring protein-protein interactions. However, a "clean" interpretation of the obtained data is not always easy. For instance, rupture force histograms generally do not fit simple theories. In particular, there is a high force tail that is not accounted for. This tail has variously been attributed to multiple binding (even though obvious multiple ruptures are excluded from analysis) or heterogeneity in the binding geometry. Here, we present a combined approach to answer the question of how much of the high force tail can be attributed to either cause. We used surfaces with well-controlled densities of active sites (biotin) to control multiple attachments with a functionalized tip (avidin). We found that the presence of multiple attachments, while significant, accounts for only a fraction of the events in the high force tail of the distribution. We also performed Monte Carlo simulations to match experimental results with theoretical expectations, confirming the importance of possible bond heterogeneity in these types of measurements.

Anwesha Sarkar
Wayne State University

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