AN AUTOMATED BAYESIAN PIPELINE FOR RAPID ANALYSIS OF SINGLE-MOLECULE BINDING DATA

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Single-molecule binding assays enable the study of how molecular machines assemble and function. Current algorithms can identify and locate individual molecules, but require tedious manual validation of each spot. Moreover, no solution for high-throughput analysis of single-molecule binding data exists. Here, we describe an automated pipeline to analyze single-molecule data over a wide range of experimental conditions [1]. In addition, our method enables state estimation on multivariate Gaussian signals. We validate our approach using simulated data, and benchmark the pipeline by measuring the binding properties of the well-studied, DNA-guided DNA endonuclease, TtAgo, an Argonaute protein from the Eubacterium *Thermus thermophilus*. We also use the pipeline to extend our understanding of TtAgo by measuring the protein's binding kinetics at physiological temperatures and for target DNAs containing multiple, adjacent binding sites (Figure 1).

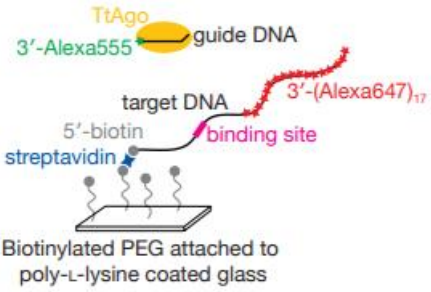

Figure 1 System for studying TtAgo

[1] An Automated Bayesian Pipeline for Rapid Analysis of Single-Molecule Binding Data; Carlas S Smith, Karina Jouravleva, Maximiliaan Huisman, Samson Jolly, Phillip D Zamore, David Grunwald; bioRxiv 261917; doi: https://doi.org/10.1101/261917