Complexity Science DTC miniproject proposal

What can DNA-protein binding experiments tell us about the nature of overstretched DNA? A statistical mechanical approach

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Aim: You will use an existing computational statistical mechanical model (and simple analytic estimates) to suggest a set of experiments, involving DNA-binding proteins, that might reveal the nature of 'overstretched' DNA.

Background: Dynamics of conformational changes of DNA

DNA in the cell is subject to considerable forces and torques. DNA is spooled tightly around proteins in the cell nucleus, as a means of storage [1]; it is forced into viral capsids by molecular motors [2]; and it is stretched along its axis by the protein RecA [3]. Stretching facilitates a process called 'recombination', which promotes genetic diversity and DNA repair. DNA stretching is therefore crucial for the molecule's biological function, but is also of great importance technologically: elongating DNA changes its conductance [4], a useful property in the context of using molecules of DNA as wires in nanoscale devices. However, the fate of DNA under tension is not well understood. In this project we aim to understand it better.

The deformations that DNA suffers in the cell can be reproduced in experiment using recently-developed techniques for manipulating single molecules [5]. At forces below 10 picoNewtons (pN), DNA pulled along its axis behaves as a conventional polymer. Between 10 and about 65 pN it extends as if it were an harmonic spring. But as one exceeds a tension of about 65 pN, double-stranded DNA undergoes a surprising and abrupt elongation. The resulting 'overstretched' form of the molecule is 1.7 times longer than the familiar double-helix structure (called 'B-form DNA'). The nature of overstretched DNA is disputed: one view holds that overstretched DNA consists of an elongated double-stranded form called S-DNA [6] (the 'B-to-S' picture), while a competing picture considers overstretching to signal a conversion to single strands [7] (the 'force-melting' picture).

Thermodynamic data do not distinguish clearly between the two competing pictures of elongation. However, within a statistical mechanical model [8] we observe that the emergent *kinetics* associated with these pictures are distinctly different, with the B-to-S picture better describing experiment. The model offers predictions on length and time scales comparable with those of experiment (see Figure 1). In the proposed project you will use this model to understand what happens when proteins bind to DNA under tension.

Proposed Project: What can DNA-protein binding experiments tell us about the nature of overstretched DNA?

One means of determining the nature of overstretched DNA might be to use proteins that bind to DNA. For instance, RecA and T4 gene 32 protein bind to stretched DNA in general more rapidly than they do to unstretched DNA [3, 9]. It is difficult to infer immediately from these measurements what is the conformation that DNA adopts at a given tension, because the proteins in question bind to both double-stranded *and* single-stranded forms of the molecule. However, quantitative modelling may allow us to make such an inference. Specifically, if we know or can estimate the rates and thermodynamic affinities with which proteins bind to different conformations of DNA, then it will be possible to use the statistical mechanical model to make predictions for what would happen in experiment based on each of the two proposed pictures of DNA overstretching. On the basis of these predictions, we would like to discriminate between the B-to-S and force-melting pictures of overstretching.

Your task will be to use a statistical mechanical model (and simple analytic estimates) to suggest a set of exper-

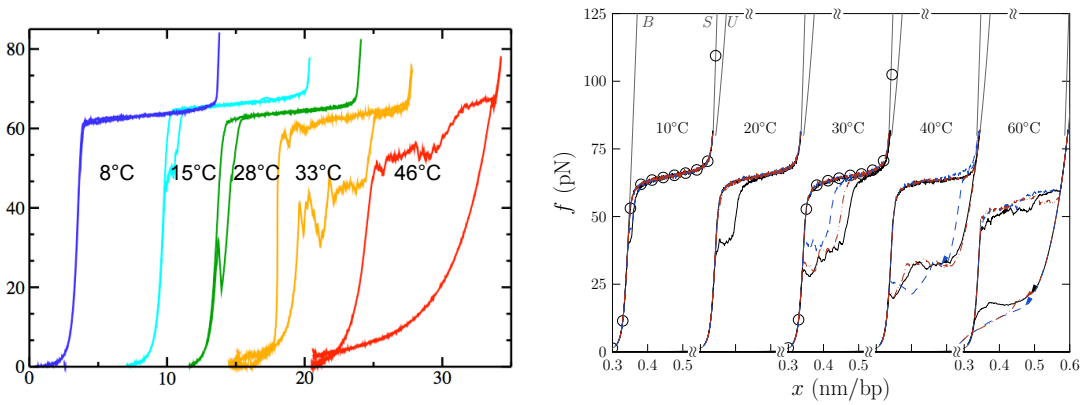

Figure 1: Using a statistical mechanical model to reproduce the kinetics of DNA overstretching experiments. Left panel: overstretching experiments [Mao, H.B. *et al. Biophys. J.* (2005)] (force in pN (vertical) versus extension (horizontal), curves offset horizontally) performed upon lambda DNA over a range of temperature T. The DNA is first stretched and then allowed to recover its original length. At low temperature stretching and shortening curves lie on top of each other, signaling a reversible transition. As temperature increases one observes a progressive increase in the degree of hysteresis, as well as a 'roughening' of the force-extension plateau at very high temperature. These features are reproduced by a statistical mechanical model [8] (right panel, three independent simulations shown for each T); within the model, melting at high temperature generates hysteresis by way of sluggish recombination of separated strands. Plateau-roughening is the signature of strand separation in concert with an inhomogeneous sequence. You will use this model to make predictions for experiments involving DNA-binding proteins.

iments, involving DNA-binding proteins, that might reveal the nature of 'overstretched' DNA. The most important question in this field remains, *Is overstretched DNA double-stranded?* You will adapt the statistical mechanical model mentioned above to accommodate proteins binding to DNA (this will be a relatively small overhead, since only minor modifications are required). Data for DNA-binding proteins are reported in the literature, and you will use this information to devise, and then test computationally, a means of discerning the mechanics of DNA under tension. I encourage you to follow your own ideas and shape the direction of the project as you see fit.

References

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