Probing Macromolecular Complexes with a Reconfigurable Nanoscale DNA Force Spectrometer

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Single molecule force spectroscopy is a valuable approach to studying the structure of biological materials and their kinetic properties. Nevertheless, the probes limit integration into complex systems, and the cost and complexity of the equipment and assays limit broader use. DNA-based nanodevices are a promising alternative that allows for probing the force response of biomolecules such as nucleosome[1,2]. Here, we build on these prior works to develop a nanoscale DNA force spectrometer (nDFS)[3]. Specifically, the nDFS allow for enhanced control over forces and especially the application of compression forces. Moreover, the readout from electron microscopy can provide the unique chance to observe the detailed sample structure conformation under force instead of end-to-end distance.

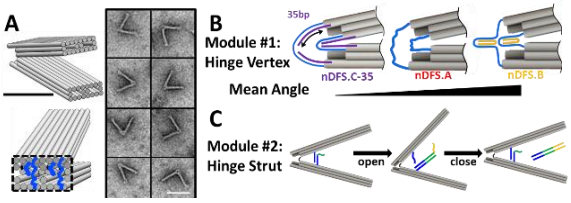

Figure 1 nDFS overview and two modules

The hinge structure nDFS is fabricated by scaffold DNA origami, and it consists of two arms connected by several single-stranded DNA scaffold linkers (Fig.1A). The device behaves like a torsional spring where the arms are stiff, and the mechanical properties are determined by the design of the hinge vertex. We demonstrated the ability to control both the equilibrium angle and stiffness by modifying the detailed vertex architecture (Fig. 1B). Specifically, the mean angle can be tuned over a range of 35 deg to 87 deg, which provides a passive approach to modulating forces applied by the nDFS. We also developed an active approach to the nDFS open or closed by forming or disrupting a DNA duplex strut between the arms, controlled via strand displacement (Fig.1C). The toggling strategy is used to 1) apply compressive forces to a 249 bp double-stranded DNA (dsDNA) 2) and apply tensile forces to tetra-nucleosome array. For the dsDNA compression, the nDFS was directly folded into the open state and the dsDNA sample was integrated via biotin-neutravidin binding. The nDFS was then toggled closed to induce bending of the dsDNA (Fig. 2A) The polymer model captured the behavior of the dsDNA[3]. For the tetra-nucleosome experiment, the nucleosome array was first incorporated into a closed nDFS and then tensile forces were applied by toggling the nDFS open (Fig. 2B), leading to decompaction of the array, as indicated by an increase in radius of gyration, Rg. For future work, we seek to expand the applicability of nDFS to a broader experimental environment by enhancing the structural stability in low ionic conditions and increasing the force measurement range.

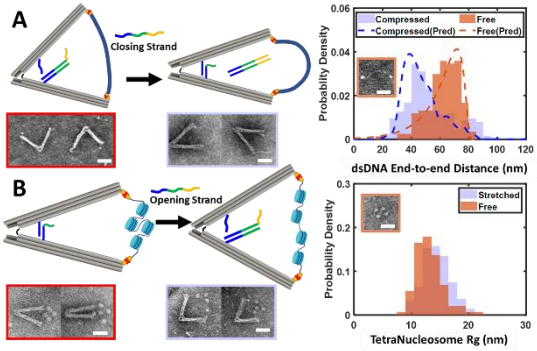

Figure 2 (A) nDFS provides compression force to dsDNA. (B) nDFS provides tensile force to tetra-nucleosome

[1] Le et al. (2016). ACS Nano, 10, 7073–7084.
[2] Funke et al. (2016). Sci. Adv., Vol 2. Issue 11
[3] Wang et al (2021) Nucleic Acids Research, Vol 49, 8987–8999