Materials List for:

Stretching Short Sequences of DNA with Constant Force Axial Optical Tweezers

Krishnan Raghunathan[1], Joshua N. Milstein[2], Jens -Christian Meiners[2]

[1]LSA Biophysics, University of Michigan

[2]LSA Biophysics, Department of Physics, University of Michigan

Correspondence to: Krishnan Raghunathan at krish@umich.edu

URL: https://www.jove.com/video/3405
DOI: doi:10.3791/3405

Materials

Name	Company	Catalog Number	Comments
Reagent/Equipment	Company	Catalog number	Comments
Nd:YVO4 laser	Spectra Physics	T40-Z-106C	
Acousto-optic deflector	IntraAction	DTD-274HA6	
Microscope Objective	Olympus	PlanApo	60X, NA 1.4
Piezo stage	Mad City Labs	Nano-LP100	XYZ stage
CCD camera	PixeLink	PL-A741	
Photodetector	Electro-Optics Tech	ET-3020	
Polystyrene Beads	Spherotech	SVP-08-10	800nm, streptavidin coated
Anti-digoxigenin	Roche	11333089001	From sheep
Primers	MWG operon	Custom oligos	One primer: biotin Other : digoxigenin
PCR reagents	New England Biolabs	TAQ polymerase, dNTPs	
Coverglass	Fisher Scientific		
Other chemicals for buffer	Fisher Scientific		

Supplementary Materials

A. Hydrodynamic Friction C–fficient

For determining the hydrodynamic friction c–fficient of the microsphere near a surface one can use the following expansion[5,10]:

where the following shorthand has been introduced:

The friction c–fficient is defined in terms of the fluid viscosity η and the radius of the microsphere, with the microsphere's center located a distance η above the surface. The summation converges reasonably well when expanded to about ten terms.

B. Influence of Axial Position on Stiffness Calibration

The calibration of the trap stiffness involves a tradeoff between the accuracy of the calibration, which increases with increasing distance from the surface, and the actual axial position where the trap is used experimentally. In general, the trap is calibrated at around 800-1000 nm from the surface, which is higher than the actual experimental condition.

C. Modified Worm-Like Chain (WLC) Model

The force extension curves can be fit to a modified WLC model that accounts for volume exclusion effects at zero optical force as follows:

where F_{opt} is the optical force, x_o is a fit parameter for the zero force extension,x_{opt} is the extension under force, l is the contour length of the DNA, and l^*_p is a second fit parameter for an "effective" persistence length. F_{wlc} is given by the usual WLC model[11] where ε is the relative DNA extension.

