Single-molecule sequence detection via microfluidic planar extensional flow at a stagnation point

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Supporting Information

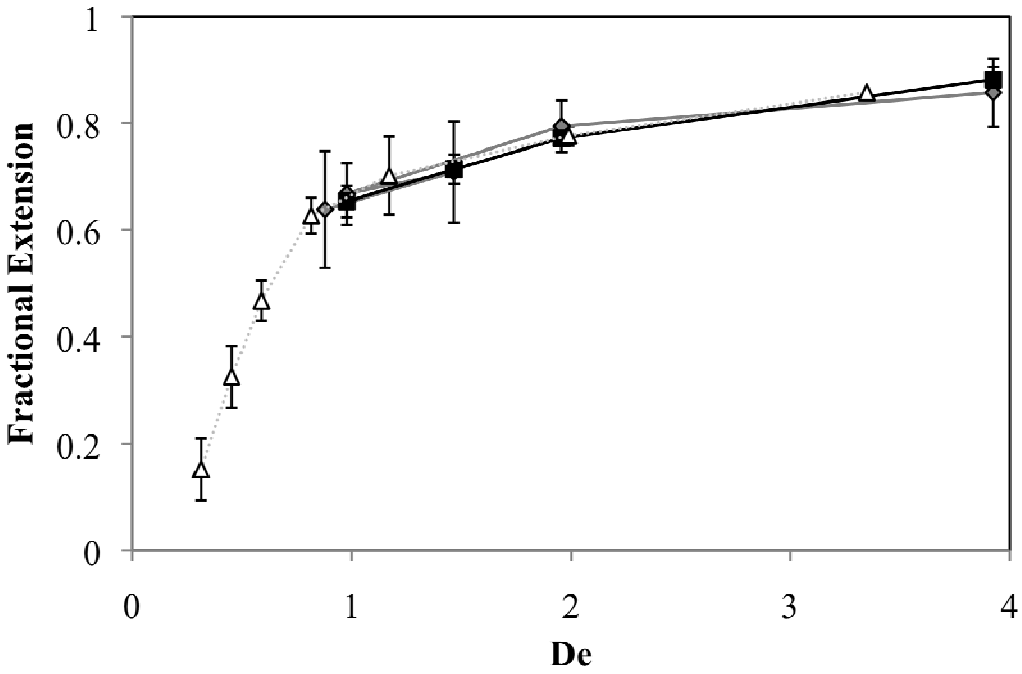

Figure S-1. DNA extension variation with De at the stagnation point of the cross slot. Results are shown for both DNA molecules (black squares) and marker-DNA complexes (gray diamonds) in EcoRI visualization buffer. Each data point is the average fractional extension of an ensemble of N > 22 stained λ-DNA molecules trapped for at least 3 strain units at the stagnation point. Strain rates were calculated using 2D fluid dynamics simulation at centerline velocities using Comsol. Results from Perkins *et al.* (white triangles) for DNA in a different buffer system are provided for comparison.[1]

1. T.T. Perkins, D.E. Smith, and S. Chu, *Science*, 1997, **276**, 2016-2021.

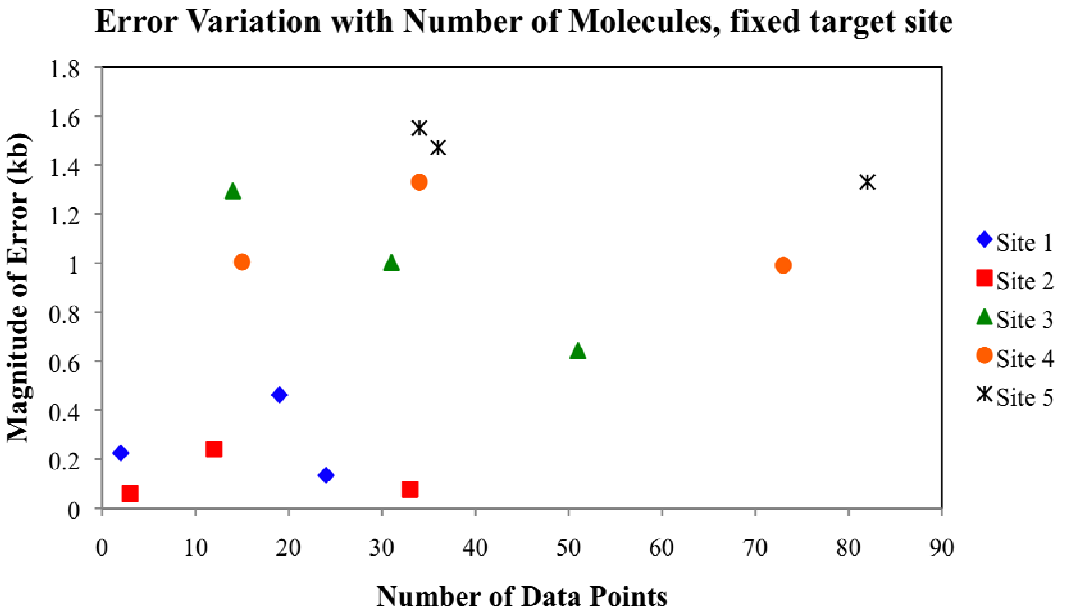

Figure S-2. Error variation with size of population at fixed target site for molecules extended at De = 1, 1.5, and 3.9. Note that the magnitude of the error does not uniformly decrease as more molecules are included in the analysis.

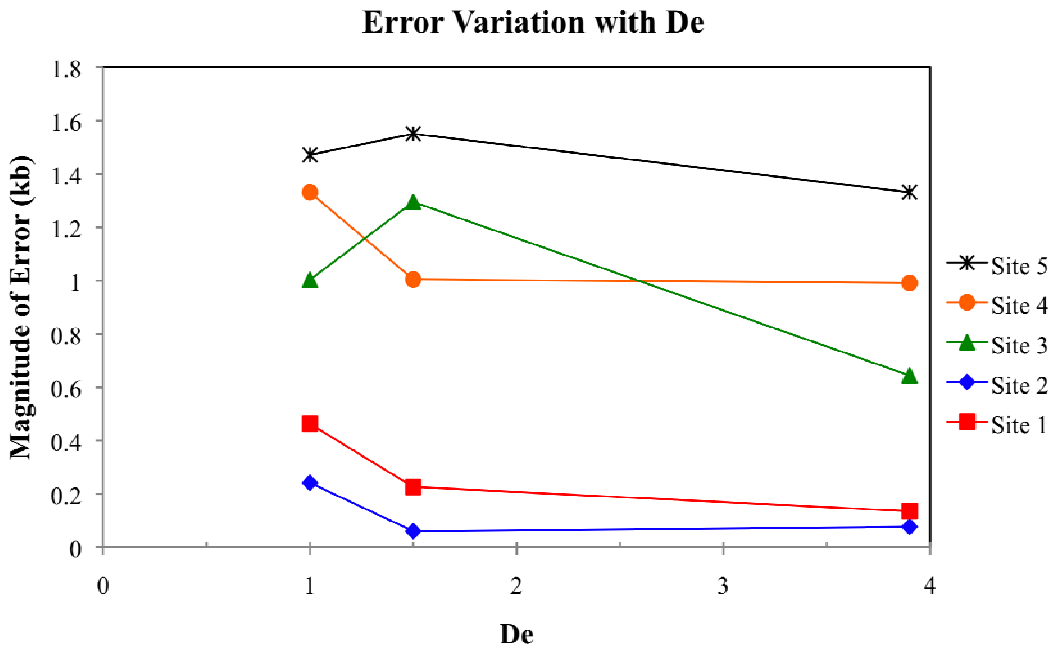

Figure S-3. Variation in error with De at the five target sites. The sites are ordered sequentially on λ-DNA as described in the text, with site 5 being nearest the end of the DNA. Sites 1 and 2 straddle the center of the molecule. Error magnitude increases with distance from the center of the molecule.

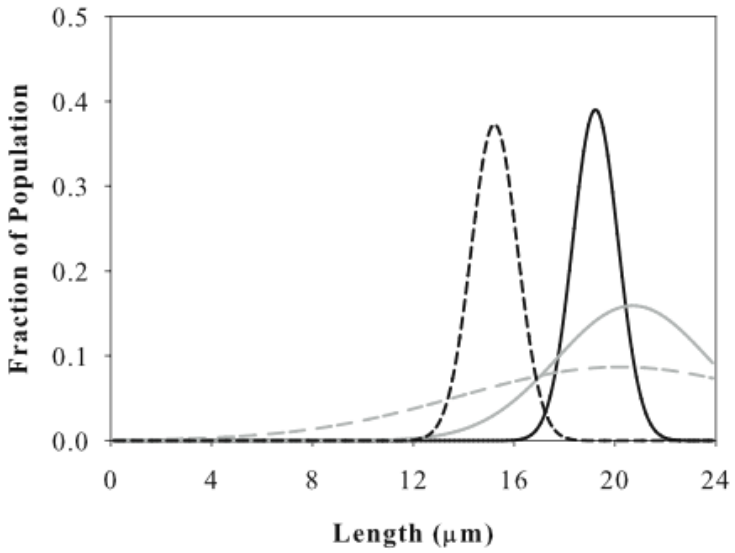

Figure S-4. Best-fit Gaussian length distributions for populations of measured marker-DNA complexes. Broken gray line includes data for all complexes stretched and measured on slides. Solid gray line represents only slide-stretched complexes with x/L > 0.75. Black lines represent molecules stretched and measured in extensional flow at De = 1 (broken) and De = 3.9 (solid) and give much narrower distributions than molecules stretched on slides.

Incubation Condition	N	<x>/L	Site 1: 21.226 kb		Site 2: 26.105 kb		Site 3: 31.747 kb		Site 4: 39.169 kb		Site 5: 44.973 kb	
			Δ (kb)	σ (kb)	Δ (kb)	σ (kb)	Δ (kb)	σ (kb)	Δ (kb)	σ (kb)	Δ (kb)	σ (kb)
Unmixed	130	0.89 (0.04)	-0.1	0.8	+0.2	1.0	+0.9	1.5	+0.9	0.9	+1.2	0.9
Mixed	133	0.86 (0.03)	-0.2	0.8	0	0.5	+0.7	1.2	+0.8	1.1	+1.2	1.0

Table S-1. Gaussian best-fit statistics for histograms produced using marker-DNA complexes trapped and stretched at the stagnation point of a planar extensional flow at De = 3.9. Populations were either continually mixed during marker-DNA incubation or incubated with no mixing. Error Δ and precision σ are reported as described in Table 1.