

Stress fluctuation calculated from DNA fluorescent images

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DNA に蛍光色素を結合させて DNA の分子運動を観察し、高分子のモデル系として議論する一連の研究がある。本研究では観察された DNA 画像から粗視化高分子モデルを構築し、応力緩和を推算する方法を開発した。

1 Introduction

It has been developed that direct observation of individual DNA dynamics can be achieved by fluorescent microscopy[1]. Although DNA is a rigid polyelectrolyte indicating characteristic features, in certain conditions the observed motion can be described by the established models in polymer physics for flexible neutral polymers[2]. Hence DNA observation has been used to investigate polymer dynamics in general for dilute and concentrated solutions on the conformational and transitional dynamics[3, 4, 5]. As an extension of the DNA fluorescent microscopy, in this study a method to estimate stress tensor from individual DNA images was proposed by reconstructing global conformation of DNA molecule.

2 Method

Aqueous solutions of T4-DNA (166kbp; 56 μ m, NIPPON GENE) with various concentrations (0.01, 600 and 1000 μ Mbp) including 0.5 TBE buffer, anti-bleach agent, 2-mercaptethanol, and oxygen scavengers were used. Motion of probe DNA labeled with YOYO-1 fluorescent dye (Molecular Probes) introduced in the solution was traced by fluorescent microscope (OLYMPUS IX-71) equipped with high sensitive camera (EB-CCD C7196-43, HAMAMATSU)

Assuming that the observed fluorescent image field ($I(r)$) directly reflects density distribution of segments on DNA, I construct an effective potential field $U(r)$ defined by

$$U(r) = U_m(r) + U_p(r) \quad (1)$$

$$U_m(r) = -a_m kT \frac{I(r)}{\int I dr} \quad (2)$$

$$U_p(r) = a_p kT \sum_i^N \delta(x - R_{xi}) \delta(y - R_{yi}) \quad (3)$$

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where a_m and a_p is tuning parameters being order unity. U_p is introduced to compensate effect of excluded volume being penalty to achieve adequately distributed conformation in whole area of the fluorescent domain. Here N is number of segments of the chain, $\mathbf{r} = (x, y)$ and position of the segment is $\mathbf{R}_i = (R_{xi}, R_{yi})$. According to the potential, freely-jointed chain is generated and equilibrated by MonteCarlo manner. From the generated chain conformation, stress tensor can be calculated by the standard expression written as

$$\sigma(t) = \nu \frac{3kT}{a^2} \langle u_\alpha(i, t) u_\beta(i, t) \rangle_i \quad (4)$$

where $\mathbf{u}$ is bond vector in the generated chain.

3 Results and Discussion

Figure 1 shows a particular working example. Since the observed image is restricted onto two dimensional plane, the generated conformation is arbitral to the vertical direction. However shear stress obtained for the plane of the observation, i.e., the projection of the conformation onto the plane should be reasonable. Assessment on the obtained stress relaxation by macroscopic rheological measurement is in progress and will be reported elsewhere.

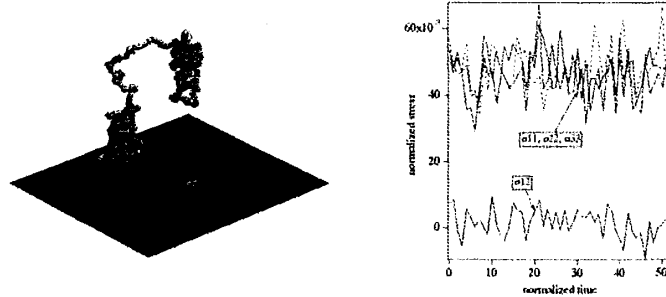


Figure 1: Left: observed DNA image and constructed chain conformation. Right: stress fluctuation calculated from the constructed chain conformation from each image in the observed movie.

References

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