

Free energy of twisted semiflexible polymers

Supurna Sinha*

Raman Research Institute, Bangalore 560 080, India

(Received 21 February 2008; published 4 June 2008)

We investigate the role of fluctuations in single-molecule measurements of torque-link (t -lk) curves. For semiflexible polymers of finite persistence length (i.e., polymers with contour length L comparable to the persistence length L_p), the torque versus link curve in the constant-torque (isotorque) ensemble is distinct from the one in the constant-link (isolink) ensemble. Thus, one encounters the conceptually interesting issue of a “free energy of transition” in switching ensembles while making torque-link measurements. We predict the dependence on the semiflexibility parameter $\beta=L/L_p$ of this extra contribution to the free energy, which shows up as an area in the torque-link plane. This can be tested against future torque-link experiments with single biopolymers. We bring out the inequivalence of torque-link curves for a stiff polymer and present explicit analytical expressions for the *distinct* torque-link relations in the two ensembles and the free-energy difference in switching ensembles in this context. The predictions of our work can be tested against single-molecule experiments on torsionally constrained biopolymers.

DOI: [10.1103/PhysRevE.77.061903](https://doi.org/10.1103/PhysRevE.77.061903)

PACS number(s): 87.15.-v, 05.40.-a, 36.20.-r

I. INTRODUCTION

The statistical mechanics of semiflexible polymers is of great current interest. Research in this area has been motivated by experiments [1,2] on biopolymers in which single molecules are stretched and twisted to measure elastic properties. These experiments are designed to understand the role of semiflexible polymer elasticity [3–5] in, for instance, the packaging of these polymers in a cell nucleus. The process of DNA transcription can generate supercoiling. It is also regulated by supercoiling [6]. In a typical experiment [6] probing the twist elasticity of a DNA molecule, the ends of a single molecule of double-stranded DNA are attached to a glass plate and a magnetic bead. Magnetic fields are used to rotate the bead and magnetic field gradients to apply forces on the bead. By such techniques the molecule is stretched and twisted and the extension of the molecule is monitored by the location of the bead. One can thus measure the extension of the molecule as a result of the applied link and force [6] and also make a measurement of the torque versus applied link [7].

In the context of force-extension measurements, an isometric setup is described by the Helmholtz free energy, whereas an isotensional setup is described by the Gibbs free energy [8]. In statistical mechanics, these two ensembles are distinct [9–11]. In the thermodynamic limit these two descriptions agree, but semiflexible polymers (those with contour length L comparable to the persistence length L_p —i.e., $\beta=L/L_p \approx 1$) are *not* at the thermodynamic limit.

In the present paper we explore this issue in the context of torque-link measurements. The two distinct statistical-mechanical ensembles here are the constant-torque ensemble (isotorque ensemble) and the constant-link ensemble (isolink ensemble). Here we focus on the role of fluctuations in single-molecule torque-link experiments. In order to correctly interpret such experiments one needs to understand the

effect of fluctuations on the measured quantities. For instance, it turns out that an experiment in which the link applied to a polymer molecule is fixed (isolink) and the torque fluctuates yields a different result from one in which the torque is held fixed (isotorque) and the link fluctuates [12]. This difference can be traced to large fluctuations about the mean value of the torque or the link, depending on the experimental setup. Experimentally, both isolink and isotorque ensembles are realizable. Here is a schematic description of an experimental setup for torque-link measurements of single biopolymer molecules. A polymer molecule attached to a glass plate on one end is suspended in a suitable medium with a magnetized bead attached to the other end. The magnetized bead is kept in a magnetic trap. One can realize an isolink setup by using a “stiff magnetic trap” and an isotorque setup by using a “soft magnetic trap,” which allows the applied link to fluctuate, but applies a fixed torque to the molecule [13,14]. The fluctuations in torque-link measurements vanish only in the thermodynamic limit of very long polymers. In the next section we describe the setup in more detail.

The paper is organized as follows. In Sec. II we discuss the isolink and isotorque ensembles. In Sec. III we illustrate the phenomenon of inequivalence of ensembles in torque-link measurements by explicitly presenting analytical expressions in the context of a stiff polymer. In Sec. IV we draw attention to the notion of the free energy of a transition in going from one ensemble to another and its dependence on semiflexibility, which can be tested against future single-molecule experiments on torsionally constrained polymers and simulations. Finally, we conclude the paper in Sec. V.

II. CONSTANT-LINK AND CONSTANT-TORQUE ENSEMBLES

Consider an experiment, as described in the Introduction, in which one end of a biopolymer molecule is attached to a glass plate and the other end is attached to a magnetized bead (of magnetic moment $\vec{\mu}$) kept in a magnetic field \vec{B} , which is

*supurna@rri.res.in

used to rotate the bead. We suppose both \vec{B} and $\vec{\mu}$ are parallel to the glass plate. The energy of the bead is given by

$$E = -\vec{\mu} \cdot \vec{B} = -\mu B \cos(\theta - \theta_0), \quad (1)$$

where θ_0 describes the direction of the magnetic field and θ the direction of the magnetic moment $\vec{\mu}$. The variables lk and lk_0 , which keep track of the number n of turns of the bead, are related to θ and θ_0 as follows: $lk = \theta + 2n\pi$ and $lk_0 = \theta_0 + 2n\pi$. Consider $P(lk)d lk$, the number of configurations (counted with Boltzmann weight) for a polymer of contour length L and bend persistence length L_p , characterized by a semiflexibility parameter $\beta = \frac{L}{L_p}$, in a link interval $d lk$ of lk . The free energy defined by $\Phi(lk) = -\frac{1}{\beta} \ln P(lk)$ is the free energy pertaining to a fixed link. The partition function [11,15] for the combined system consisting of the polymer molecule and the magnetized bead in the magnetic field is given by

$$Z(lk_0, \beta) = \int_{-\infty}^{+\infty} d lk e^{-\beta\Phi(lk)} e^{\beta\mu B \cos(lk - lk_0)} \quad (2)$$

A. Constant-link ensemble: The limit of a stiff trap

In the limit of a stiff trap ($\mu B \rightarrow \infty$), $\vec{\mu}$ follows \vec{B} closely and $lk \approx lk_0$. Thus $\cos(lk - lk_0) \approx 1 - \frac{(lk - lk_0)^2}{2}$. In this limit the partition function for the combined system (molecule+trap) reduces to

$$Z(lk_0, \beta) = e^{\beta\mu B} \int_{-\infty}^{+\infty} d lk e^{-\beta\Phi(lk)} e^{-\beta\mu B(lk - lk_0)^2/2}. \quad (3)$$

Clearly, in this limit the Gaussian factor pertaining to the magnetic trap approaches a δ function and we have

$$Z(lk, \beta) \approx e^{-\beta\Phi(lk)}. \quad (4)$$

Here we have switched notation to write lk in place of lk_0 . Thus a stiff trap realizes the constant-link (isolink) ensemble by constraining fluctuations in lk . In order to change the applied link from lk to $lk + d lk$, one applies a torque $\langle t \rangle = \frac{\partial \Phi}{\partial lk}$. Thus one gets a torque-link ($\langle t \rangle, lk$) curve by plotting $\langle t \rangle$ versus lk .

B. Constant-torque ensemble: The limit of a soft trap

In the opposite limit of a soft trap, μB is small, but large enough that the polymer does not get untwisted. In such a situation the link fluctuates. One can adjust lk_0 such that $lk - lk_0 \approx \pi/2$. The magnitude of the torque, $|\vec{t}| = |\vec{\mu} \times \vec{B}| = t = \mu B$, is held fixed for a particular measurement. In this limit the torque t is the control parameter, which can be changed from one reading to the next by changing the magnitude B of the magnetic field. A feedback loop is used to ensure that $\langle lk - lk_0 \rangle$ is maintained at $\pi/2$. Clearly, the potential energy for the trap, on expanding around $lk - lk_0 = \pi/2$ and retaining terms to linear order in $lk - lk_0$, takes the form

$$E = -\vec{\mu} \cdot \vec{B} = \mu B(lk - lk_0) = t(lk - lk_0). \quad (5)$$

Thus, in this limit Eq. (2) gives the following expression for the partition function $\tilde{Z}(t, \beta) = Z(lk_0, \beta) e^{\beta t lk_0}$ (where lk_0 is determined by the condition $\langle lk - lk_0 \rangle = \frac{\pi}{2}$) for the combined system consisting of the polymer molecule and the trap [11,15]:

$$\tilde{Z}(t, \beta) = \int_{-\infty}^{+\infty} d lk e^{-\beta\Phi(lk)} e^{\beta t lk}. \quad (6)$$

Thus in a soft trap link lk fluctuates, but torque fluctuations are constrained [16]. One thus realizes the constant-torque (isotorque) ensemble. $\tilde{Z}(t)$ [17] is the generating function for the lk distribution. Given the constant-torque free energy $\Gamma(t) = -\frac{1}{\beta} \ln \tilde{Z}(t)$, one gets the mean link $\langle lk \rangle = -\frac{\partial \Gamma}{\partial t}$ and ($t, \langle lk \rangle$) torque-link relation.

Notice that $\tilde{Z}(t)$ is the Laplace transform of $Z(lk)$. In the thermodynamic limit of long polymers ($\beta \rightarrow \infty$) the Laplace transform integral (6) is dominated by the saddle-point value and therefore $\Phi(lk)$ and $\Gamma(t)$ are related by a Legendre transform

$$\Phi(lk) = \Gamma(t) + t lk. \quad (7)$$

For finite β —i.e., for a polymer of finite extent—the saddle-point approximation no longer holds true and fluctuations about the saddle-point value of the free energy become important. Thus one finds that, for a finite β , $\Phi(lk)$ and $\Gamma(t)$ are not Legendre transforms of each other, but are related via a Laplace transform [Eq. (6)].

III. INEQUIVALENCE OF ENSEMBLES IN TORQUE-LINK MEASUREMENTS: A SPECIAL CASE

We illustrate the issue of the inequivalence of ensembles in the context of torque-link measurements explicitly by considering an analytically tractable and instructive special case: the torque-link relation for a stiff polymer.

Our starting point is the wormlike chain (WLC) Hamiltonian with bend and twist degrees of freedom in the presence of a stretching force f and a torque t [12]:

$$H = p_\theta^2/2 + (p_\phi - A_\phi)^2/2 \sin^2 \theta - f \cos \theta - \alpha t^2/2. \quad (8)$$

Here p_θ and p_ϕ are momenta conjugate to the Euler angles θ and ϕ . The momentum conjugate to the Euler angle ψ is $p_\psi = it$, a constant of motion, which contributes a term $-\alpha t^2/2$ to the Hamiltonian where α is the ratio of the bend persistence length L_p to the twist persistence length L_T . The “vector potential” $A_\phi = it(1 - \cos \theta)$ [4,12]. For a stiff polymer with one end clamped along the \hat{z} direction, we can approximate the sphere of directions by a tangent plane at the north pole of the sphere as the angular coordinate θ always remains small. In this limit [the paraxial wormlike chain (PWLC) model [4]] where the tangent vector never wanders too far away from the north pole of the sphere of directions, the polymer Hamiltonian [3,4,12,18] reduces to

$$H_{\text{PWLC}} = \frac{p_\theta^2}{2} + \frac{(p_\phi - A_\phi)^2}{2\theta^2} - \frac{\alpha t^2}{2} - f \left(1 - \frac{\theta^2}{2} \right) = H_P - f - \frac{\alpha t^2}{2},$$

where H_P is the Hamiltonian of interest in the paraxial limit after we take out a constant piece. In this limit $A_\phi = \frac{i\theta^2}{2}$. We introduce Cartesian coordinates $\xi_1 = \theta \cos \phi$ and $\xi_2 = \theta \sin \phi$ on the tangent plane R^2 at the north pole.

The PWLC model has been applied earlier in the context of flexible polymers at high tension [4]. Here we apply it in the context of stiff polymers at $f=0$. In the stiff limit, the tangent vector to the polymer points essentially along a fixed direction (north pole) even at $f=0$. As in Ref. [4], we restrict ourselves to the case of $\alpha=0$ [19].

In Ref. [20] we have derived the partition function $Z(f)$ of a stiff polymer with both end tangent vectors pointing along a fixed direction \hat{z} at a force f . In the presence of a force f and a torque t this expression goes over to

$$\tilde{Z}(t) = \sqrt{f - t^2/4} \exp(\beta f) / [2\pi \sinh(\beta \sqrt{f - t^2/4})]. \quad (9)$$

Notice that the ‘‘effective force’’ $\sqrt{f - t^2/4}$ replaces f when there is a competition between a force f and a torque t . This is simply due to the fact that the Hamiltonian H_P in the presence of a force f and a torque t pertains to that of a two-dimensional harmonic oscillator with frequency $\omega = \sqrt{f - t^2/4}$ rather than $\omega = \sqrt{f}$, which is the expression for the frequency of a two-dimensional harmonic oscillator in the presence of a force f and *no* torque.

At zero force, the expression simplifies and reduces to

$$\tilde{Z}(t) = t / [4\pi \sin(\beta t/2)]. \quad (10)$$

From the partition function $\tilde{Z}(t)$, we get the following analytical expression for the torque-link $(t, \langle \text{lk} \rangle)$ relation in the isotorque ensemble:

$$\langle \text{lk} \rangle = \frac{1}{\beta t} - \frac{1}{2} \cot\left(\frac{\beta t}{2}\right). \quad (11)$$

Given $\tilde{Z}(t)$, one can get the partition function $Z(\text{lk})$ in the conjugate domain of an isolink ensemble (see Sec. II) [20,21]:

$$Z(\text{lk}) = (\pi/2\beta) / [\cosh(\pi \text{lk})]^2. \quad (12)$$

This leads to the following torque-link $(\langle t \rangle, \text{lk})$ relation:

$$\langle t \rangle = (2\pi/\beta) \tanh(\pi \text{lk}). \quad (13)$$

Thus, the torque-link $(t, \langle \text{lk} \rangle)$ relation obtained in the isotorque ensemble [Eq. (11)] is distinct from the one $(\langle t \rangle, \text{lk})$ obtained in the isolink ensemble [Eq. (13)] (see Fig. 1).

In the next section we derive explicit expressions for the extra contribution to the free energy due to changes in the ensemble.

IV. FREE ENERGY OF THE TRANSITION IN TORQUE-LINK MEASUREMENTS

In an isotorque setup, torque is the control parameter and one measures the mean link to plot the torque-link $(t, \langle \text{lk} \rangle)$ curve. In an isolink setup the roles of link and torque are

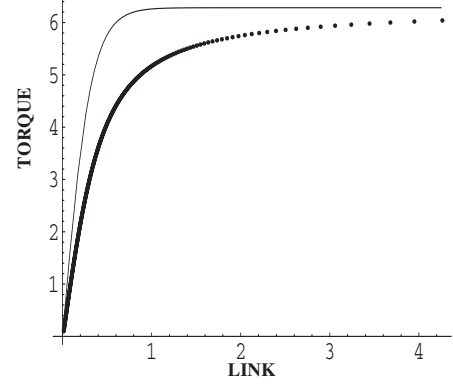


FIG. 1. Torque-link curve in the isolink (upper curve) and isotorque (lower curve) ensembles for $\beta=1$.

interchanged. Consider going from a small torque (lk_1, t_1) to a large torque (lk_2, t_2) configuration via an isotorque setup and returning from a large link (lk_2, t_2) to a small link (lk_1, t_1) configuration via an isolink setup. Since the torque-link relation depends on the chosen ensemble, in general there will be two distinct curves joining the points (lk_1, t_1) and (lk_2, t_2) in the torque-link plane describing the two processes. Such a transformation could lead to a net area being enclosed in the torque-link plane. This appears paradoxical since it seems to suggest that one can extract work from the system via a cyclic process. The resolution of this paradox is as follows. In completing the cycle and returning to the initial state, one is in fact changing ensembles twice at the two end points. Such *ensemble changes* in a cyclic transformation involve *finite changes* in free energy, which need to be taken into consideration. In particular, we notice for the special case of a stiff polymer the torque-link relations [Eqs. (11) and (13)] lead to a *free-energy difference* of

$$\Delta_{\text{stiff}} = \frac{2}{\beta} \ln \left| \frac{\cosh(\pi \text{lk}_2)}{\cosh(\pi \text{lk}_1)} \right| - (t_2 \text{lk}_2 - t_1 \text{lk}_1) + \frac{1}{\beta} \left[\ln \left| \frac{t_2}{t_1} \right| - \ln \left| \frac{\sin\left(\frac{\beta t_2}{2}\right)}{\sin\left(\frac{\beta t_1}{2}\right)} \right| \right]$$

for a transformation between the states (lk_1, t_1) and (lk_2, t_2) . In the stiff regime (i.e., at small β) the dependence of the free energy on β will be dominated by the first term. In other words, the free-energy difference $\Delta_{\text{stiff}} \sim \frac{1}{\beta}$. This is a prediction of our analysis, which can be tested against experiments with stiff biopolymers like actin filaments.

An analysis similar to the one in Ref. [9] applied to the context of torque-link measurements shows that one gets a contribution $\Delta = \frac{1}{2\beta} \ln \Phi''(\text{lk}_*)$ to the free energy coming from fluctuations around the long-polymer ($\beta \rightarrow \infty$) limit by expanding $\Delta(\text{lk}) = \Phi(\text{lk}) - t \text{lk}$ around the saddle-point value $\text{lk} = \text{lk}_*$ pertaining to the long-polymer limit. Here $\Phi(\text{lk})$ corresponds to the isolink free energy. This extra contribution $\Delta(\text{lk})$ vanishes in the limit of $\beta \rightarrow \infty$. For finite β , this non-zero contribution to the free energy accounts for the transition between the constant-link ensemble and the constant-

torque ensemble. Work is done on the bead by the trap in making the trap stiffer, while in going from a stiff trap to a soft trap work is extracted from the bead by the trap. The net work done is the difference between the work done at the two ends of the torque-link curves in switching ensembles. This net work exactly cancels out the nonzero area enclosed in the torque-link plane. The net area enclosed in the torque-link plane pertaining to the “free energy of transition” scales as $1/\beta$ and therefore grows with the rigidity of the polymer. These predictions of our study can be tested against future simulations and single-molecule experiments.

V. CONCLUSION

To summarize, in this paper we have studied the inequivalence of ensembles for torque-link measurements. We have calculated the free-energy difference between torque-link measurements in the isotorque and isolink ensembles for a stiff polymer. In addition, we have determined the contribution to the “free energy of transition” in going between the isolink and isotorque ensembles by expanding the free-energy difference $\Delta(\text{lk})$ around the long-polymer limit. We

predict the dependence on the semiflexibility parameter β of this extra contribution to the free energy, which shows up as an area in the torque-link plane. For the special case of a stiff polymer we find explicit analytical expressions for the torque-link $(t, \langle \text{lk} \rangle)$ relation obtained in the isotorque ensemble [Eq. (11)] and show that it is distinct from the one $(\langle t \rangle, \text{lk})$ obtained in the isolink ensemble [Eq. (13)]. We also show that in this stiff regime the free-energy difference $\Delta_{\text{stiff}} \sim \frac{1}{\beta}$. All the predictions mentioned here can be qualitatively and quantitatively tested against future single-molecule experiments on torsionally constrained biopolymers.

The theoretical predictions presented in this study are expected to generate interest in torsionally constrained single-molecule experiments, which will eventually lead to a deeper understanding of the role of twist elasticity in biological processes involving gene regulation [22].

ACKNOWLEDGMENT

It is a pleasure to thank A. Ghosh and D. Marenduzzo for discussions.

-
- [1] C. Bustamante *et al.*, *Curr. Opin. Struct. Biol.* **10**, 279 (2000).
 - [2] Yuri Tsuda *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **93**, 12937 (1996).
 - [3] J. Samuel and S. Sinha, *Phys. Rev. Lett.* **90**, 098305 (2003).
 - [4] Supurna Sinha, *Phys. Rev. E* **70**, 011801 (2004).
 - [5] J. Samuel, Supurna Sinha, and A. Ghosh, *J. Phys.: Condens. Matter* **18**, S253 (2006).
 - [6] T. R. Strick *et al.*, *Science* **271**, 1835 (1996).
 - [7] Zev Bryant *et al.*, *Nature (London)* **424**, 338 (2003).
 - [8] L. D. Landau and E. M. Lifshitz, *Statistical Physics*, 3rd ed. (Pergamon Press, New York, 1980), Pt. 1.
 - [9] Supurna Sinha and Joseph Samuel, *Phys. Rev. E* **71**, 021104 (2005).
 - [10] A. Rosa, D. Marenduzzo, and S. Kumar, *Europhys. Lett.* **75**, 818 (2006).
 - [11] D. Keller, D. Swigon, and C. Bustamante, *Biophys. J.* **84**, 733 (2003).
 - [12] C. Bouchiat and M. Mezard, *Eur. Phys. J. E* **2**, 377 (2000).
 - [13] A. La Porta and M. D. Wang, *Phys. Rev. Lett.* **92**, 190801 (2004).
 - [14] One way to apply torque could be to explore some intrinsic birefringence property of the material by using polarized light; see, for instance, A. Ghosh *et al.*, *Phys. Biol.* **3**, 67 (2006).
 - [15] We drop additive constants in the free energies or, equivalently, multiplicative constants in the partition functions.
 - [16] H. J. Kreuzer and S. H. Payne, *Phys. Rev. E* **63**, 021906 (2001).
 - [17] We suppress the β dependence and write $\tilde{Z}(t)$ instead of $\tilde{Z}(t, \beta)$ and $Z(\text{lk})$ instead of $Z(\text{lk}, \beta)$ in our subsequent discussion.
 - [18] J. D. Moroz and P. Nelson, *Proc. Natl. Acad. Sci. U.S.A.* **94**, 14418 (1997).
 - [19] There is no loss of generality in choosing $\alpha=0$. One can extend the analysis to finite α by multiplication by a Gaussian factor in the generating function space as outlined in [4].
 - [20] A. Ghosh, J. Samuel, and S. Sinha, *Phys. Rev. E* **76**, 061801 (2007); M. Emanuel *et al.*, *ibid.* **76**, 061907 (2007).
 - [21] Supurna Sinha and J. Samuel, *Phys. Rev. B* **50**, 13871 (1994).
 - [22] See, for instance, B. Alberts *et al.*, *Molecular Biology Of The Cell*, 3rd ed. (Garland, New York, 1994).