Anisotropic Elastic Model for Short DNA Loops

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Effect of bending anisotropy on a planar DNA loop, using energy minimization and neglecting entropic effects, is studied. We show that the anisotropy results in polygonal shape of the loop and increasing the anisotropy makes the edges sharper. Calculating the energy of such a loop lets us to find effective persistence length as the geometrical mean of hard and soft rigidities, which is quite different from harmonic mean for an unconstrained long DNA.

I. INTRODUCTION

A long time has been passed since a straight double helical structure had been proposed by Watson and Crick as relaxed configuration of a DNA, but today, we know it can almost never be found in this form in nature. The macromolecule is able to play its important and essential role in the life, when it is interacting with different proteins which force it to bend, twist, melt and/or pack [1, 2].

To understand the macromolecule’s functionality, DNA has been examined in different length scales. Single molecule force-extension studies on $10 - 100 \mu m$ DNAs [3], cyclization experiments on $100 - 1000 nm$ DNAs [4] and recent experiments on sharply bent DNAs in loops with the lengths less than $50 nm$, persistence length of DNA [5, 6, 7, 8]. All these experiments are of great biological interest. Depending on relevant length scale of the problem, experimental results are usually analyzed in two types of theoretical models: variants of the continuum elastic worm-like chain (WLC) and base-pair steps.

The worm-like chain models are based on the physics of elastic rod where the energy has a quadratic form in deformations. There are two approaches to this model. The first one is based on Kirchhoff-like equations of balance of forces and torques for every segment of the rod [9, 10] and the other one is based on energy minimization [11, 12]. In these models statistical mechanics of the rod at non-zero temperature is also encountered [13]. A force constant or equivalently a deformation module is defined for bending, twisting, stretching and their coupling terms. In the classical WLC model, all cross terms as well as stretching energy are neglected and the rod is isotropic in bending in different directions [14]. So the model is identified with just two persistence lengths assigned to each of bending or twisting degrees of freedom. Modifications to this model have been made by encountering twist-stretch coupling, twist-bend coupling and stretch-bend coupling [11, 12, 13, 15], and estimating the free parameters by fitting the theory to force-extension experimental data. WLC model has also been used to evaluate loop formation probability, J-factor [12, 14].

Looking at microscopic structure of DNA macromolecule suggests that bending toward the groove is easier than bending toward the backbone. Though, there is no direct experiment to measure the anisotropy, there are some theoretical postulates for double stranded DNA [19, 20, 21]. Monte Carlo simulations have also confirmed that B-DNA bends more easily in the groove direction (roll) than in the backbone direction (tilt) [22]. It has been encountered in some models for describing force-extension and DNA cyclization experiments and found to have no significant improvement to the results [11, 12, 23], though existence of bending anisotropy affects the existence of twist-stretch coupling [17]. Balaeff et al. have claimed that bending anisotropy can reduce the energy of loops in length scale of 76 bp by a factor of one third [24]. In a recent experiment an asymmetry in the periodic behavior of free energy of loop formation as a function of loop length for $60 - 100$ bp loops has been detected [7]. Fourier analysis of free energy shows two main frequencies. One with a period of $\sim 10.5$ bp due to helical shape of the double strand and one with a period of $\sim 5.6$ bp that might be a result of bending anisotropy.

Sequence dependence and anisotropy of bending persistence length has been widely noticed in base-pair steps approaches, in which relative rotation and displacement of every two segments are defined through six parameters slide, shift, rise, tilt, roll and twist [25]. These sequence dependent parameters for individual base-pair steps have been determined from their standard deviation in crystal complexes [26]. Theoretical work has also been done on extracting these parameters from atomic level parameters via the analysis of Molecular Dynamics (MD) trajectories [27]. Different simulation studies give estimations for roll and tilt values. Munteanu et al. have shown that for $3 - 11$ bp DNAs bending rigidity oscillates with bending direction and the values of roll is $\sim 8 - 10$ times greater than the values of tilt [28]. Olson et al. estimate the ratio of the hard bending rigidity, $A_1$, to the soft bending rigidity $A_2$ to be between $4$ and $16$ [24] and between $1$ and $5$ in a more recent study [29]. MD simulations of 17 bp dsDNAs gives $A_1$ almost twice as $A_2$ [30]. In a stack of plates simulations, Mergell et al. state that special constraints make roll twice as favorable as tilt [31].
Effect of sequence dependency on persistence length and also intrinsic curvature on loop formation problem have been studied analytically by Popov et al. which result in a wide distribution of cyclization probabilities.\textsuperscript{29}

An effective persistence length, $A$, can be assigned to a rod with hard and soft anisotropic bending rigidities $A_1$ and $A_2$ (corresponding to tilt and roll). In an analytical stack of plates study, done by Olson et al., a complicated dependence of $A$ on detailed couplings, anisotropic constants and sequence alphabet has been shown for a free DNA.\textsuperscript{29}

For a free, long and highly twisted DNA, the effective persistence length exactly equals the harmonic mean of soft and hard rigidities,

$$A = 2\left(\frac{1}{A_1} + \frac{1}{A_2}\right)^{-1}. \tag{1}$$

This can be easily deduced from the equipartition principal. The equipartition principle states that the total energy of a rod is equal to $\frac{1}{2}k_BT$ times number of its degrees of freedom. Bending of a segment of a rod around a principal axis with length equal to its persistence length can be considered as one degree of freedom. Therefore number of degrees of freedom in hard and soft directions is simply counted as $\frac{L}{A_1}$ and $\frac{L}{A_2}$ and the total energy is found to be $\frac{1}{2}k_BT\left(\frac{L}{A_1} + \frac{L}{A_2}\right)$. To find the efficient persistence length, the anisotropic rod is considered as an isotropic one. Since the isotropic rod is able to be bent around two similar directions, its energy is found to be $2\left(\frac{1}{2}k_BT\frac{L}{A}\right)$ in which $A$ is the efficient persistence length. Having the two estimations equal to each other results equation \textsuperscript{1} This is more accurately derived by Kehrbaum using an averaging theory for a non-isotropic elastic rod with high intrinsic twist.\textsuperscript{32} Adding geometrical constraints on a free DNA might affect the above relation.

Here we are going to give an expression for effective persistence length of in-plane DNA loops, by energy minimization, when the entropic effects are neglected. Although an isotropic WLC loop with length smaller than its persistence length is planar, it is not obvious for the case of anisotropic DNA. In fact experiments\textsuperscript{34} and MD simulations\textsuperscript{35} show that DNA mini-circles are not completely, but are almost planar.

The rest of paper is organized as follows: Section 2 describes the model that is used in studying the short DNA loops, followed by the presentation of the results in Section 3. Finally, Section 4 concludes the paper, while the overestimation of deviation of DNA local twist from its mean value appears in Appendix.

\section*{II. THE MODEL}

The anisotropic elastic model of DNA represents the macromolecule as an elastic rod of length $L$, parameterized by arclength $s$. As the double-strand is ribbon-like, it is anisotropic in bending around two different directions. On the other hand, DNA is twisted, so while bending around a fixed axis, it should bend around each of the two directions, periodically. It is energetically preferable for the DNA to bend more around the “soft” axis and less around the “hard” one. To reduce elastic energy, the DNA also may modify its twist to have more soft bending along its planar path.

For a planar DNA loop, the tangent unit vector $\hat{e}$ and the twist angle $\psi$ at each point contain enough information to parameterize DNA conformation. Actually an anisotropic bent DNA has a higher tendency to go out of the bending plane which depends on the value of anisotropy $(A_1 - A_2)/(A_1 + A_2)$. For short loops, this tendency decreases as the anisotropy becomes smaller.\textsuperscript{36}

Elastic energy of anisotropic rod is

$$E = \frac{1}{2}k_BT\int_0^L \left[ A(s)|\dot{\hat{e}}|^2 + C(\psi - \omega_0)^2 \right] ds, \tag{2}$$

where $A(s)$ is the local bending rigidity, $C$ is the twist rigidity, $\omega_0$ is the spontaneous twist of the helix, and the dots indicate derivatives with respect to $s$. The first term is the usual elastic term, with $s$ dependent persistence length $A(s)$ and the second term is the energy needed to over-(under-)twist the DNA, which may be implied by the boundary conditions on $\psi$ or the anisotropic effects. It should be noted that in this model $s$ dependence of $A$ is not because of sequence dependence, but it is due to rotation of soft and hard axes. As we will show later, this is a source of twist-bend coupling, although there is no explicit coupling term in the Hamiltonian.

DNA bending could be decomposed into two principal axes $\hat{e}_1$ and $\hat{e}_2$, attached to the DNA. The hard one, $\hat{e}_1$ is perpendicular to double strand’s local plane, and the soft one, $\hat{e}_2$, is defined to lie in the local plane of the double strand, and to be perpendicular to both strands. Due to the helical structure of DNA, $\hat{e}_1$ and $\hat{e}_2$ rotate with the helix. Since $\hat{e}$ is perpendicular to $\hat{t}$ axis and lies in the $\hat{e}_1 - \hat{e}_2$ plane, by its decomposition in our coordinate system we obtain

$$E = \frac{1}{2}k_BT\int_0^L \left[ (A_1 \sin^2 \psi + A_2 \cos^2 \psi) \theta^2 \\ + C(\hat{\psi} - \omega_0)^2 \right] ds, \tag{3}$$

where $A_1$ and $A_2$ are constant bending rigidities about the rotating axes, $\hat{e}_1$ and $\hat{e}_2$, and $\theta$ is the angle between $\hat{t}$ and a fixed, arbitrary direction in loop’s plane. Using Euler-Lagrange equation and applying corresponding boundary conditions, we are able to find $\theta(s)$ and $\hat{\psi}(s)$ as well as DNA’s shape and bending energy of the loop. The integral form of closed loop or “ends meeting” condition is

$$\int_0^L \sin \theta(s) \, ds = \int_0^L \cos \theta(s) \, ds = 0. \tag{4}$$
Also because the DNA strands are antiparallel they can not switch in the ends and they should bind in phase, then we have \(\psi(L) - \psi(0) = 2k\pi\) with \(k\) being an integer that is the number of full turns of the helix. On the other hand to avoid any singularity in tangent vector changes we consider a constraint on \(\theta\) values in the ends by \(\theta(L) - \theta(0) = 2\pi\) (no self crossing).

III. RESULTS

Integrating Euler-Lagrange Equations of (3) results following equations of motions:

\[
\dot{\theta} = \frac{\gamma}{(A_1 + A_2) - (A_1 - A_2) \cos 2\psi},
\]

\[
\dot{\psi}^2 = -\frac{2\gamma^2}{C} \frac{1}{(A_1 + A_2) - (A_1 - A_2) \cos 2\psi} + \beta,
\]

where integral constants of \(\gamma\) and \(\beta\) should be determined from the boundary conditions. As it is seen in above equations, \(\psi\) depends on \(s\), as on the DNA’s local bending in a complicated form. This twist-bend coupling is a direct result of anisotropy of the model and vanishes in the case of isotropic rods. Even if \(A_1\) and \(A_2\) differ by one order of magnitude, which is the case of our double stranded DNA, the coupling is very weak and \(s\) dependence of \(\psi\) is negligible. Indeed, numerical studies show that the relative variations of \(\psi\) is less than one percent, even if two bending rigidities differ by two orders of magnitude [36]. This leads us to consider homogeneous twist along the DNA and set \(\psi(s) = \omega s\), in which \(\omega = 2k\pi/L\) is found by applying the “ends in phase” boundary condition. This is more discussed in Appendix. The above approximation decouples torsional part of energy from its bending part.

Integrating (5) with considering above approximation gives

\[
\theta(s) = \theta(0) + \frac{\gamma}{\omega \sqrt{A_1A_2}} \times \left( \tan^{-1} \left( \sqrt{\frac{A_1}{A_2}} \tan(\omega s) \right) + \pi \left[ \frac{1}{2} + \frac{\omega s}{\pi} \right] \right),
\]

where the bracket means “integer part”. This term is added to get rid of discontinuity in the \(\tan^{-1}\) function. Without lack of generality, we set \(\theta(0) = 0\) and therefore the condition on total bending results \(\theta(L) = 2\pi\). Applying this condition simply lets us to fix \(\gamma = (2\pi/L) \sqrt{A_1A_2}\). These would yield the functional form of \(\theta(s)\) and loop’s shape as

\[
\theta(s) = \frac{1}{k} \left( \tan^{-1} \left( \sqrt{\frac{A_1}{A_2}} \tan(\omega s) \right) + \pi \left[ \frac{1}{2} + \frac{\omega s}{\pi} \right] \right).
\]

The above solution automatically satisfies the “ends meeting” condition [4]. \(k\) in above equation counts the number of turns of helix along the loop. To have minimum torsional energy it should be fixed to the closest integer value to \(\omega_0L/2\pi\), so \(k = \text{round}(\omega_0L/2\pi)\). Thus the DNA is undertwisted in case \(\Delta \psi = \omega_0L - 2k\pi < 0\) and is overtwisted otherwise.

In the case \(A_1 >> A_2\) for a closed DNA loop, bending is not homogeneous and it is localized in “soft” parts. Here the loop looks more like a polygon rather than a circle. This is the direct effect of bending anisotropy. The shape of looped DNA for anisotropic model is given in figure 1. Increasing the ratio of \(A_1/A_2\), the polygonal shape is more visible (e.g. for \(A_1/A_2 > 50\), the DNA loop will have sharp edges at the soft points). In a full helix turn the DNA meets the soft axis of rotation two times when it bends in plane, thus the number of polygon edges equals \(2k\). As the number of edges (\(\sim\) helical turns) increases by length of the loop, the polygonal shape of the loop is less visible for larger DNAs.

To find the energy of the loop, we read \((A_1 \sin^2 \psi + A_2 \cos^2 \psi) \dot{\theta} = \gamma\) from (5) and substitute it in (3),

\[
E = \frac{1}{2} k_B T \int_0^L \left( \gamma \dot{\theta} + C(\psi - \omega_0)^2 \right) ds.
\]

As \(\int_0^L \dot{\theta} ds = 2\pi\) and \(\dot{\psi}\) and \(\gamma\) are constant, the elastic energy is

\[
E = \frac{2\pi^2}{L} k_B T \sqrt{A_1A_2} + \frac{k_B T}{2L} C \Delta \psi^2.
\]

The second term is the twist energy which is due to the over-(under-)twist implied by the “ends in phase” condition on DNA loop. Because \(k\) is a step function of \(L\), this term leads to a well known oscillatory behavior which is damping by an \(L^{-1}\) factor. In analogy with the bending energy of a circular loop, \((2\pi^2 k_B T A/L)\), we can read the effective persistence length of in-plane small loops as \(\sqrt{A_1A_2}\). This is different from the effective persistence length of anisotropic DNA in larger scales (equation (11)).

IV. CONCLUSION

We studied the effect of bending anisotropy on planar DNA loops using energy minimization and neglecting entropic effects under constraints of parallel and in phase ends. Bending anisotropy causes anisotropy in curvature and twist. However, anisotropy induced a twist-bend coupling to the model even in the lack of explicit appearance of such coupling in the Hamiltonian, though, it is small enough to be neglected in the calculations. The bending anisotropy results in polygonal shape of the loop. Increasing the anisotropy makes the edges sharper where the number of helical turns and hence the number of edges of the polygon grows with loop length.

Energy of such a loop includes an oscillating term for twist energy and a bending term similar to that of anisotropic loop with an effective persistence length \(A = \sqrt{A_1A_2}\), which is different from harmonic mean of
that considering anisotropy in DNA bending rigidities is not able to explain large loop formation probability of DNA minicircles. Thus it seems other efforts, as like as bubble formation [37], effect of sequence dependence [32], or generalized semiflexible model [38] are more successful in this direction.

APPENDIX

We are going to give a simple numeric estimation for deviation of $\dot{\psi}$ from its mean value. Since $\cos 2\psi$ varies between $-1$ and $1$ the maximum value of the $\psi$ dependent term (first term) of equation (6) is

$$\max(x) = \frac{-\gamma^2}{C} \frac{1}{A_2} = \frac{-4\pi^2 A_1 A_2}{C A_2 L^2}. \quad (11)$$

Applying the typical values for a DNA minicircle (i.e. $\omega = 1.8$ nm$^{-1}$, $C = 75$ nm, $L = 94$ bp and bp = 0.34 nm) following by rough estimations, $A_1 = 100$ nm and $A_2 = 33$ nm (i.e $A_1/A_2 = 3$ with a harmonic mean of 50 nm), result a max($x$) $\approx 0.016 \omega^2$. So the deviation of $\dot{\psi}^2$ is small comparing to the mean value and therefore $\dot{\psi}^2 \approx \beta \approx \omega^2$. Now finding (max – min) of both sides of equation (6), we have:

$$2\dot{\psi} \Delta \dot{\psi} = \frac{\gamma^2}{C} \left( \frac{1}{A_2} - \frac{1}{A_1} \right) = \frac{4\pi^2}{L^2 C}(A_1 - A_2) \quad (12)$$

and as $\dot{\psi} \approx \omega$

$$\frac{\Delta \dot{\psi}}{\omega} = \frac{4\pi^2}{L^2 C \omega^2} (A_1 - A_2) \approx 1\% \quad (13)$$

It should be noted that above calculation is an overestimation of the range of $\dot{\psi}$ changes. As it was mentioned in the text, numerical study shows less than one percent fluctuation in the value even with considering two rigidities different by two orders of magnitude. That means, considering $\omega$ as a constant ($\dot{\psi} = \omega s$), is not a rude approximation.

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[1] K.S. Matthews, Microbiol. Rev. 56, 123 (1992)
[2] R. Schleif, Annu. Rev. Biochem. 61, 199 (1992)
[3] S.B. Smith, L. Finzi, C. Bustamante, Science 258, 1122 (1992)
[4] W. Han, M. Dlakic, Y.J. Zhu, S.M. Lindsay, R.E. Harrington, Proc. Natl. Acad. Sci. USA 94, 10565 (1997)
[5] T.E. Cloutier, J. Widom, Molecular Cell 14, 355 (2004)
[6] T.E. Cloutier, J. Widom, Proc. Natl. Acad. Sci. USA 102, 3645 (2005)
[7] L. Saiz, J.M. Rubi, , J.M.G. Vilar, Proc. Natl. Acad. Sci. USA 102, 17642 (2005)
[8] Q. Du, C. Smith, N. Shiffeldrim, M. Vologodsii, Proc. Natl. Acad. Sci. USA 102, 5397 (2005)
[9] L. Mahadevan, J.B. Keller, Proc. R. Soc. Lond. A 452, 1679 (1996)
[10] T.P. Westcott, I. Tobias, W.K. Olson, J.Chem. Phys 107, 3967 (1997)
[11] J.D. Moroz, P. Nelson, Macromolecules 31, 6333 (1998)
[12] Y. Zhang, D.M. Crothers, Biophysical Journal 84, 136 (2003)
[13] H. Yamakawa, J. Shimada, M. Fujii, The Journal of Chemical Physics 68, 2140 (1978)
[14] J. Shimada, H. Yamakawa, Macromolecules 17, 689 (1984)
[15] J.F. Marko, E.D. Siggia, Macromolecules 27, 981 (1994)
[16] R.D. Kamien, T.C. Lubensky, P. Nelson, C.S. O'Hern, Europhys. Lett 38, 237 (1997)
[17] C.S. O'Hern, R.D. Kamien, T.C. Lubensky, P. Nelson, Eur. Phys. J. B 1, 95 (1998)
[18] F. Mohammad-Rafiee, R. Golestanian, Phys. Rev. Lett. 94, 238102 (2005)
[19] J.A. Schellman, Biopolymers 13, 217 (1974)
[20] V.B. Zhurkin, Y.P. Lysov, V.I. Ivanov, Nucleic Acids Res 6, 10811096. (1979)
[21] A. Matsumoto, W.K. Olson, Biophysical Journal 83, 22 (2002)
[22] V.B. Zhurkin, N.B. Ulyanov, A.A. Gorin, R.L. Jernigan, Proc. Natl. Acad. Sci. USA 88, 7046 (1991)
[23] S.D. Levene, D.M. Crothers, J. Mol. Biol. 189, 61 (1986)
[24] A. Balaeff, C.R. Koudella, L. Mahadevan, K. Schulten, Phil. Trans. R. Soc. Lond. A 362, 1355 (2004)
[25] W.K. Olson, N.L. Marky, R.L. Jernigan, V. Zhurkin, J. Mol. Biol 232, 530 (1993)
[26] W.K. Olson, A.A. Gorin, X.J. Lu, L.M. Hock, V.B. Zhurkin, Proc. Natl. Acad. Sci. USA 95, 111631168 (1998)
[27] O. Gonzalez, J.H. Maddocks, Theor. Chem. Acc. 106, 76 (2001)
[28] M.G. Munteanu, K. Vlahovicke, S. Parthasarathy, I. Simon, S. Pongor, Trends Biochem. Sci. 23, 341 (1998)
[29] W.K. Olson, D. Swigon, B.D. Coleman, Phil. Trans. R. Soc. Lond. A 362, 1403 (2004)
[30] F. Lankas, J. Sponer, P. Hobza, J. Langowski, J. Mol. Biol 299, 695 (2000)
[31] B. Mergell, M.R. Ejtehadi, R. Everaers, Phys. Rev. E 68, 021911 (2003)
[32] Y.O. Popov, A.V. Tkachenko, Effects of sequence disorder on dna looping and cyclization (2006), cond-mat/0510302
[33] S. Kehrbaum, Ph.D. thesis, University of Maryland (1997)
[34] A. Amzallag, C. Vaillant, M. Jacob, M. Unser, J. Bednar, J. Kahn, A. Stasiak, J. Dubochet, J.H. Maddocks, to appear to Nucl Acids Res (2006)
[35] F. Lankas, R. Lavery, J.H. Maddocks, Structure 14, 1527 (2006)
[36] F. Mohammad-Rafiee, R. Golestanian, Eur. Phys. J. E 12, 599 (2003)
[37] J. Yan, J.F. Marko, Phys. Rev. Lett. 93(10), 108108 (2004)
[38] P.A. Wiggins, P.C. Nelson, Phys. Rev. E 73(3), 031906 (2006)