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Stretching DNA: Role of electrostatic interactions

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Abstract. The effect of electrostatic interactions on the stretching of DNA is investigated using a simple worm like chain model. In the limit of small force there are large conformational fluctuations which are treated using a self-consistent variational approach. For small values of the external force f, we find the extension scales as $fr_{\rm D}$ where $r_{\rm D}$ is the Debye screening length. In the limit of large force the electrostatic effects can be accounted for within the semiflexible chain model of DNA by assuming that only small excursions from rod-like conformations are possible. In this regime the extension approaches the contour length as $1/\sqrt{f}$ where f is the magnitude of the external force. The theory is used to analyze experiments that have measured the extension of double-stranded DNA subject to tension at various salt concentrations. The theory reproduces nearly quantitatively the elastic response of DNA at small and large values of f and for all concentration of the monovalent counterions. The limitations of the theory are also pointed out.

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1 Introduction

The technical advances in the manipulation of single molecules has enabled the probe of mechanical and relaxational measurement in both equilibrium [1,2] and non-equilibrium conditions [3]. Among the first of such experiments was the investigation of the elastic response of λ - bacteriophage DNA (λ DNA) molecules subject to tension [1]. These experiments and others have offered a window into the behavior of biological molecules on scales ranging from several nanometer to few microns. They also provide an opportunity to understand the limits of validity of theories based on generic polymer models such as Edwards model or simple model of semiflexible chain such as the worm like chain (WLC) model [4].

The earliest theories describing the elastic response of WLC subject to tension, which are relevant to the experiments of Smith *et al.* [1], were due to Fixman and Kovac [5] and Crabb and Kovac [6]. The precise experiments made possible by nanomanipulation of single molecules [1,2] has demanded more accurate theories. In these experiments the response to a constant force on a magnetic bead attached to λ DNA in a solution of varying salt concentration is used to probe DNA elasticity. After these pioneering experiments were reported several theoretical studies followed [7–9]. These papers showed that, when the salt concentration is sufficiently large, the simple WLC model subject to tension quantitatively reproduces the force-extension curves. In particular, the asymptotic approach to full extension at large force, was shown to follow the $f^{-1/2}$ law, where f is the external force.

Since DNA is highly charged one expects counterion effects to be extremely important in determining the elasticity. It has recently been established that the elastic response and persistence length of DNA greatly depend not only on the salt concentration but also on the valence and shape of counterions [10]. As in the case of simple polyelectrolytes, systematic theories of DNA subject to tension is difficult due to the presence of several competing length scales. In particular, at intermediate values of the external force, the interplay of several forces, namely intrinsic persistence length, electrostatic repulsion, counterion condensation, all conspire to determine the conformation of DNA.

Marko and Siggia [8] included the effects of electrostatic interaction, within a WLC picture of DNA by replacing the intrinsic persistence length by a scale dependent effective persistence length [11]. Such a description implicitly assumes that the intrinsic persistence length is large, and hence only small excursions in DNA from rod-like conformations are tolerated. The interaction between the charges is assumed to obey the Debye-Hückel potential, $V_{\rm DH}(r) = l_{\rm B}{\rm e}^{-\kappa r}/r$. The ionic strength of the solution I is related to the screening length $r_{\rm D} = \kappa^{-1}$ through the relation $\kappa^2 = 4\pi l_{\rm B} I$, and the Bjerrum length $l_{\rm B} = {\rm e}^2/(\epsilon k_{\rm B} T)$ represents the strength of interaction. The scale dependent effective persistence length $l_{\rm p}^{\rm eff}$ varies from $l_0 + l_{\rm OSF}$ (in length scale $r \gg r_{\rm D}$) to l_0 ($r \ll r_{\rm D}$), where $l_{\rm OSF} = l_{\rm B}/4\kappa^2 A^2$ [12,13] with A being the mean distance between charges. Marko and Siggia considered the limit

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when the applied force is so large that the angle fluctuation of tangent vector with respect to the direction of applied force is small which in consistent with the assumption of the OSF theory [12,13]. The chain extension z in the presence of tension approaches L like $f^{-1/2}$, in good agreement with experiment at large force. However, when the conformation of DNA is coil-like (at small force) the theory shows deviation from experimental results.

In order to probe the effects of electrostatic interactions at small values of f and I we suggest a mean field variational approach. The mean field model of semiflexible chains [14,15] successfully describes the conformations of the stretched DNA by a constant force at high salt concentration, where the electrostatic interactions are negligible [9]. In this model, the hard constraint on tangential vector $\mathbf{u}^2(s) = 1$ is replaced by a global constraint $\langle \mathbf{u}^2(s) \rangle = 1$ so that the average of the magnitude of a tangential vector $\mathbf{u}(s)$ is fixed even if the magnitude of tangential vectors fluctuate. This theory provides continuous crossover formula for the extension from the small force limit to the large force limit. In small force limit the extension z increases linearly whereas in large force limit the $f^{-1/2}$ rule is found. The theory also reproduces quantitatively the experimental force-extension curves at high salt concentration.

In this paper, we include electrostatic interactions in the mean field model of semiflexible chain. This approach provides an adequate theory of DNA elasticity in the small force limit. We compare our results with experiments at various salt concentrations.

2 Semiflexible chain under tension

We model DNA as a semiflexible worm like chain (WLC) with contour length L. The chain can be parameterized by unit tangent vector $\mathbf{u}(s) = \partial \mathbf{r}(s)/\partial s$, where $\mathbf{r}(s)$ represents the position vector in three dimensions at the curvilinear position s. The energy cost for bending is characterized by the persistence length l_p. The tangent vector for WLC chain satisfies the local geometric constraint $\mathbf{u}^2(s) = 1$ for all s. The enforcement of this constraint makes the theory for interacting WLC extremely difficult. Recently, it has been shown [9] that one can get reliable results for a number of problems involving semiflexible chains by replacing the local constraint $\mathbf{u}^2(s) = 1$ by a global constraint $\langle \mathbf{u}^2(s) \rangle = 1$. The resulting theory, which in the absence of interactions reduces to the model for semiflexible chains proposed by Lagowski *et al.* [15], can be systematically derived from a functional integral approach. Here we adopt this mean field model to investigate the effects of tension.

The probability distribution for isolated WLC chain, which preserves the global constraint $\langle \mathbf{u}^2(s) \rangle = 1$ is given by [14,15]

$$P_0 = \mathrm{e}^{-\lambda \int_0^L \mathrm{d}s \mathbf{u}^2(s) - \eta \int_0^L \mathrm{d}s (\partial \mathbf{u}/\partial s)^2 - b(\mathbf{u}_L^2 + \mathbf{u}_0^2)}. \tag{1}$$

The Lagrange multiplier $\lambda = \frac{3}{2l}$ (used to enforce the global constraint $\langle \mathbf{u}^2(s) \rangle = 1$) is (roughly) inversely proportional to the Kuhn length l of the semiflexible chain, and the constant η is related to the persistence length l_p. When the free energy of the non-interacting chain is optimized with respect to λ we obtain $\lambda = 9/(8l_\mathrm{p})$ and $b = 3/4$. These values satisfy the constraint $\langle \mathbf{u}^2(s) \rangle = 1$.

The distribution function of the isolated semiflexible chain under tension is given by

$$P_0(f) = \\ \mathrm{e}^{-\int_0^L \mathrm{d}s \lambda(s) \mathbf{u}^2(s) - \eta \int_0^L \mathrm{d}s (\partial \mathbf{u}/\partial s)^2 + \int \mathbf{f}(s) \cdot \mathbf{u}(s) - b(\mathbf{u}_L^2 + \mathbf{u}_0^2)}. \tag{2}$$

If the applied force is constant, $\lambda(s)$ has a uniform value for all s [9]. It has been shown that this assumption gives a self-consistent solution to the stationary condition [9]. The optimization of the free energy, in the presence of a constant force leads to the modified relation between $\lambda = 2l/3$ and η

$$1 - \frac{3}{4}\sqrt{\frac{\eta}{\lambda}} = \frac{f^2}{4\lambda^2} \cdot \tag{3}$$

The mean square end-to-end distance of semiflexible chain under constant tension can be obtained as

$$\begin{aligned} \langle R^2 \rangle &= \int_0^L \int_0^L \langle \mathbf{u}(s') \cdot \mathbf{u}(s'') \rangle \mathrm{d}s' \mathrm{d}s'' \\ &= lL - l\sqrt{\frac{2l\eta}{3}} \left(1 - \exp\left(-\sqrt{\frac{3}{2l\eta}} L\right)\right) + \frac{l^2 f^2}{9} L^2. \end{aligned} \tag{4}$$

3 Self-consistent theory: small force regime

The WLC under the influence of constant tension yields the correct dependence of the extension (along the direction of the force) of f at relatively large value of the inverse Debye screening length. However, precise experiments on DNA have demonstrated that the simple elasticity model is inadequate to take electrostatic effects into account especially at small value of the f and at low salt concentrations. Since DNA is highly charged the response of the chain to tension clearly depends on the ionic strength and to the valence of the counterions. In the presence of the external force there are four important length scales. They are the Bjerrum length $l_\mathrm{B} (= \mathrm{e}^2/4\pi\epsilon k_\mathrm{B}T)$, the Debye screening length $\kappa^{-1} \equiv r_\mathrm{D}$, the persistence length l_p, and the Pincus length $\xi_\mathrm{P} = k_\mathrm{B}T/f$ [16]. For monovalent ions $\kappa^2 = 8\pi l_\mathrm{B} \rho$ where ρ is the salt (say NaCl) concentration. The interplay between these length scales makes the calculation of the elastic response of DNA under tension difficult. Here we provide a variational theory that is adequate to obtain force-extension curves that are in agreement with experiments.

In order to take electrostatic effects into account we assume, for simplicity, that DNA molecule is uniformly charged and each charged segment of the chain interacts *via* screened Coulomb interactions. The Hamiltonian of an isolated DNA molecule consists of non-Coulomb part H_0 and the electrostatic energy contribution ΔH.

$$H_\mathrm{t} = H_0 + \Delta H. \tag{5}$$

The new probability function $P[\mathbf{u}]$ including the electrostatic interaction is

$$P[\mathbf{u}] = P_0[\mathbf{u}] \exp\left(\frac{-\Delta H}{k_\mathrm{B}T}\right) = P_0[\mathbf{u}] \exp\left[-\omega \int_0^L \int_0^L \mathrm{d}s'' \mathrm{d}s' \frac{\mathrm{e}^{-\kappa|\mathbf{r}(s'')-\mathbf{r}(s')|}}{|\mathbf{r}(s'')-\mathbf{r}(s')|}\right] \tag{6}$$

where ω is equal to l_B/A^2, A is the distance between the charges.

When the applied force is smaller than $f < k_\mathrm{B}T/r_\mathrm{D}$, the orientation of the tangential vectors are not correlated at scales $r > r_\mathrm{D}$, which implies that the mean square average of angle fluctuation $\langle\theta(s)^2\rangle$ is not small. The chain becomes flexible on large scales and then adopts "coil like" conformation, although the chain is still stiff on small length scales. We use "coil-like" to imply that relatively large excursions from rod-like conformation are possible so that the Gaussian approximation, which would be valid when $\langle\theta^2(s)\rangle$ is small, breaks down.

For this problem the Gaussian approximation (employed to treat the long range interaction) to the WLC model is not valid [8]. This can be seen by deviations from experiments in the extension-force curves at small forces. (See Fig. 6 in Ref. [8].) Here we use a self-consistent variational theory to describe the effect of small force on the conformations of DNA.

In order to estimate the size of the charged DNA under tension we follow the uniform expansion method introduced by Edwards and Singh [17]. Accordingly, we write

$$H_\mathrm{t} = H_1 + B \tag{7}$$

where

$$B = H_0 - H_1 + \Delta H. \tag{8}$$

Hamiltonian and H_1 corresponds to the non-interacting theory in which l is replaced by a effective Kuhn length l_1 and ΔH is a perturbation in Hamiltonian. The appropriate value of l_1 should satisfy (see Eq. (4)).

$$\langle R^2\rangle = l_1 L - l_1\sqrt{\frac{l_1 l_\mathrm{p}}{3}}\left(1-\mathrm{e}^{-\sqrt{\frac{3}{l_1 l_\mathrm{p}}}L}\right) + \frac{l_1^2 f^2}{9}L^2. \tag{9}$$

For arbitrary choice of l_1, $\langle R^2\rangle$ can be rewritten up to first order in B as

$$\langle R^2\rangle = \langle R^2\rangle_1 - \langle BR^2\rangle_1 + \langle B\rangle_1\langle R^2\rangle_1 \tag{10}$$

where $\langle\,\rangle_1$ indicates average with weight factor $\exp(-\frac{H_1}{k_\mathrm{B}T})$. Self-consistency condition requires that $\langle BR^2\rangle_1 = \langle B\rangle_1\langle R^2\rangle_1$ so that $\langle R^2\rangle$ to first order in B coincides with that computed using a reference.

We assume that the Kuhn length $l = 2/3\lambda$ will be replaced by l_1 by the coarse graining processes in such a way that new parameter l_1 satisfies equation (4) at a given force and persistence length l_p. Therefore, we have the following self-consistent equation:

$$\left(\frac{1}{l}-\frac{1}{l_1}\right) l_1^2\Big[L - \sqrt{\frac{9l\,l_\mathrm{p}}{8}}(1-\mathrm{e}^{-\sqrt{\frac{3}{l_1\ l_\mathrm{p}}}L} + \frac{l_1}{2}\mathrm{e}^{-\sqrt{\frac{3}{l_1\ l_\mathrm{p}}}L} + \frac{2l_1}{9}(\beta f)^2 L^2\Big] = \langle\Delta z^2(l_1)\rangle_{\Delta H}. \tag{11}$$

The right hand side of equation (11) is evaluated using

$$\langle\Delta z^2(l_1)\rangle_{\Delta H} = \langle z^2\rangle - \langle z^2\rangle_0 = -\frac{\frac{\partial^2}{\partial k^2}G(\mathbf{k},L,\mathbf{f})}{G(\mathbf{k},L,\mathbf{f})}\Big|_{\mathbf{k}=0} - \langle z^2\rangle_0 \tag{12}$$

where $\langle\,\rangle_0$ indicate the average with weight $\mathrm{e}^{-H_0/k_\mathrm{B}T}$, and $G(\mathbf{r})$ is the Green function associated with the total Hamiltonian

$$G(\mathbf{r},L,\mathbf{f}) = \int_{\mathbf{r}(0)=\mathbf{0}}^{\mathbf{r}(L)=\mathbf{r}} D[\mathbf{r}(s)] \exp\left(\frac{-H_\mathrm{t}}{k_\mathrm{B}T}\right) \tag{13}$$

and its Fourier transform is

$$G(\mathbf{k},L,\mathbf{f}) = \int \mathrm{d}^3\mathbf{r}(s) \exp(-\mathrm{i}\mathbf{r}\cdot\mathbf{k}) G(\mathbf{r},L,\mathbf{f}). \tag{14}$$

If the applied force is constant along the contour then $G(\mathbf{k},L,\mathbf{f}) = G(\mathbf{k}-\mathrm{i}\beta\mathbf{f},L)$ [18]. We can obtain the mean-square average of the end-to-end distance from $G(\mathbf{k},L)$,

$$\langle z^2\rangle = -\frac{\frac{\partial^2}{\partial k^2}G(\mathbf{k},L,\mathbf{f})}{G(\mathbf{k},L,\mathbf{f})}\Big|_{\mathbf{k}=0} = -\frac{\frac{\partial^2}{\partial k^2}G(\mathbf{k},L)}{G(\mathbf{k},L)}\Big|_{\mathbf{k}=-\mathrm{i}\beta\mathbf{f}}. \tag{15}$$

The correlation function in k space is obtained by performing functional integral including phase factor $\mathrm{e}^{-\mathrm{i}\mathbf{k}\cdot\mathbf{r}}$. If we consider the electrostatic interaction as a perturbation, $H_\mathrm{t} = H_0 + \Delta H$, $\Delta H = V_\mathrm{DH}(r) = l_\mathrm{B}\mathrm{e}^{-\kappa r}/r$.

$$G(\mathbf{r},L) = \int \frac{\mathrm{d}^3\mathbf{k}}{(2\pi)^3}\langle\mathrm{e}^{\mathrm{i}kr}\rangle_0 - \sum_n \frac{\beta}{n!}\langle(\Delta H\mathrm{e}^{\mathrm{i}kr})^n\rangle_0. \tag{16}$$

We can write the correlation function $G(k) \approx G_0(k,L)\frac{1}{n!}\sum_{n=0} G_1(k,L) \approx G_0(k,L)\exp(G_1(k,L))$ under the Gaussian approximation.

$$G_0(k,L)G_1(k,L) = \langle V_\mathrm{DH}\mathrm{e}^{\mathrm{i}kr}\rangle_0 = \frac{1}{(2\pi)^3}\frac{\omega}{2}\int\mathrm{d}s''\int\mathrm{d}s'\int\mathrm{d}^3q\frac{\langle\mathrm{e}^{\mathrm{i}\mathbf{q}\cdot(\mathbf{r}(s'')-\mathbf{r}(s'))+\mathrm{i}\mathbf{k}\cdot(\mathbf{r}(L)-\mathbf{r}(0))}\rangle}{q^2+\kappa^2}$$
$$= \exp[-\frac{k^2 l}{6a}(aL-1+\mathrm{e}^{-aL})]\frac{\omega\sqrt{\pi^3}}{16\kappa^2}\int\mathrm{d}s\int\mathrm{d}s'\int_0^\infty\mathrm{d}\alpha \times \mathrm{e}^{-\alpha}\left(\frac{\pi}{g_1 l_1+\alpha/\kappa^2}\right)^{\frac{3}{2}}\exp\left(\frac{l_1^2 g_2^2 k^2}{g_1 l_1+\alpha/\kappa^2}\right) \tag{17}$$

with

$$a = (\frac{3}{2l\eta})^{\frac{1}{2}}$$

$$g_1(s''-s') = \frac{a(s''-s')-1+\mathrm{e}^{-a(s''-s')}}{6a}$$

$$g_2(s''-s') = \frac{a(s''-s') - 1 - 2\mathrm{e}^{\frac{aL}{2}} \sinh\frac{(s''-s')a}{2} \cosh\frac{(L-s''-s')a}{2}}{6a}$$

where the wave vector $\mathbf{q}$ is associated with the momentum transfer *via* electrostatic interaction and $\frac{1}{q^2+k^2}$ is the Fourier transform of $V_{\mathrm{DH}}(\mathbf{r}(s'') - \mathbf{r}(s'))$. We have introduced the dummy parameter α in order to replace three dimensional integral with respect to $\mathbf{q}$ to a one dimensional integral in w, The mean square average of the end-to-end distance is

$$\langle \Delta z^2 \rangle_{\Delta H} = \langle z^2 \rangle_0 + \left(2\frac{G_0'(\mathbf{k},L)G_1'(\mathbf{k},L)}{G_0(\mathbf{k},L)} + G_1''(\mathbf{k},L)\right)\Big|_{\mathbf{k}=-\mathrm{i}\beta\mathbf{f}}. \quad (18)$$

We solve equation (11) together with equations (17, 18) iteratively to find a new coarse grained Kuhn length l_1, which is related to the persistence length *via* equation (3). The correction to $\langle z^2 \rangle$ due to the electrostatic interaction is expected to be always positive since electrostatic interactions stiffen the chain. Therefore we would expect the effective Kuhn length l_1, which varies with salt concentration, to be larger than l.

In order to analyze the experimental measurements at small force using our theory we need the parameters L (the contour length), l_0 (the intrinsic persistence length of DNA), and the effective linear charge density $1/A$. The values of L and l_0 may be obtained by fitting the force-extension curve to the data of Smith *et al.* [1] at the monovalent salt concentration of 10 mM NaCl ($\kappa^{-1} \approx 3.2$ nm) using the WLC model [4]. For this condition the WLC gives an excellent description of the data because the electrostatic interactions are negligible [8,9]. The best fit is obtained with $L = 32.7$ μm and with the intrinsic persistence length $l_0 = 53$ nm. The effective charge density $1/A$ still remains free parameter. In this paper, we choose $1/A$ in order to find the best fit with experiments. We will discuss the effect of ion condensation in the following section. We use the Bjerrum length $l_{\mathrm{B}} = 0.7$ nm in water at room temperature with dielectric constant $\epsilon = 80$.

Before we present the results of the force-extension it is useful to characterize the variation of the electrostatic persistence length of DNA with κ. Here $l_{\mathrm{e}} = l_{\mathrm{p}} - l_0$, $l_{\mathrm{p}} = 3l_1/4$ and $l_0 = 53$ nm. In Figure 1 we plot l_{e} as a function of κ. It is clear that there are two distinct scaling regimes of behavior.

$$\begin{aligned} l_{\mathrm{e}} &\sim \kappa^{-1} \quad (\kappa < \kappa_x) \\ l_{\mathrm{e}} &\sim \kappa^{-2} \quad (\kappa > \kappa_x) \end{aligned} \quad (19)$$

where $\kappa_x \approx 0.34$ nm^{-1} is roughly the crossover value.

The result in equation (19) requires discussion in light of recent development in the polyelectrolyte literature [19,20]. It has been shown that both for flexible ($l_0 l_{\mathrm{B}}/A^2 \ll 1$) and stiff ($l_0 l_{\mathrm{B}}/A^2 \gg 1$) chains the electrostatic persistent length scales as r_{D}^2 [19,20]. The crossover between the two limits is quite complicated and in this regime l_{e} can even depend sublinearly on κ [21]. In fact, it is possible that l_{e} can not be precisely determined in the crossover regime [19]. The precise scale of crossover has also been a point of consideration [21]. Simulations suggest that such a crossover occurs when $l_{\mathrm{e}} \approx l_0$. From Figure 1 we note that $l_{\mathrm{e}}(\kappa_x) \approx 70$ nm which is comparable to $l_0(\approx 53$ nm). Thus for $\kappa \le \kappa_x$ one expects deviation from the OSF prediction which, we believe, is not inconsistent with the prediction of the recent variational theories [19,20] and simulations [21]. The crossover, at least for this DNA, occurs when $l_{\mathrm{e}} \approx l_0$ [21], which is consistent with the condition $l_{\mathrm{OSF}} \approx l_0$ [20].

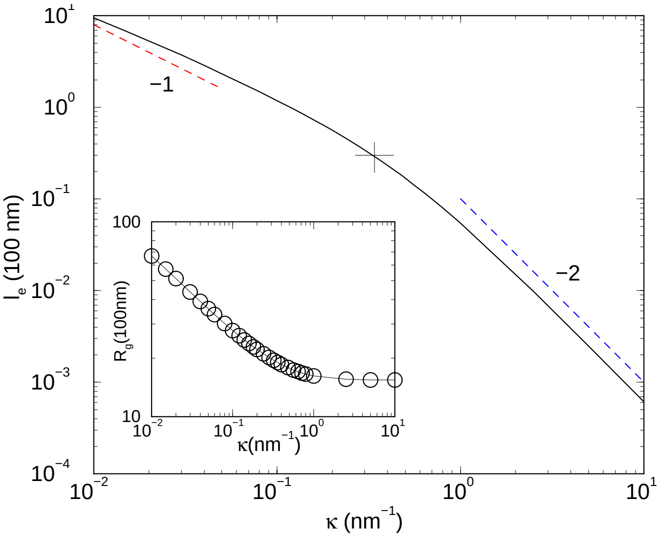

Fig. 1. The electrostatic contribution to the persistence length l_{e} as a function of κ. It is clear that l_{e} exhibits κ^{-1} and κ^{-2} dependence in low and high salt concentration respectively. The inset shows the dependence of the radius of gyration on the salt concentration, which is calculated by solving self-consistent equation (11). The value of parameters are $L = 32.7$ nm, $l_0 = 53$ nm, $1/A = 1.40$ nm^{-1}. The crossover point is indicated as a cross.

In the inset to Figure 1, we plot the salt concentration dependence of the radius of gyration of DNA. In the low salt concentration region, $\kappa < 0.1$ nm^{-1}, the radius of gyration R_{g} varies like $\kappa^{-1/2}$ and for high salt concentration, R_{g} shows little κ dependence. The $\kappa^{-1/2}$ variation, shown in the inset to Figure 1 implies $\nu = 1/2$ for R_{g}. The implication of this calculation, for our purpose, is that in the small κ regime $l_{\mathrm{e}} > l_0$ and as a result the effects of electrostatic interaction dominate at scales of l_{e}. Thus, a more detailed theory described here is required to describe the small force behavior of DNA.

In Figure 2, we plot force-extension curves for DNA molecule at 10 mM Na$^+$, 1 mM Na$^+$, 0.1 mM Na$^+$ ion concentration respectively. The fits at large forces are done by the calculation of the angle fluctuations of the tangential vector using $L = 32.7$ μm and $l_0 = 53$ nm. (See next section.) The small force fits are done with self-consistent mean field approximation using the same values for the parameters L and l_0.

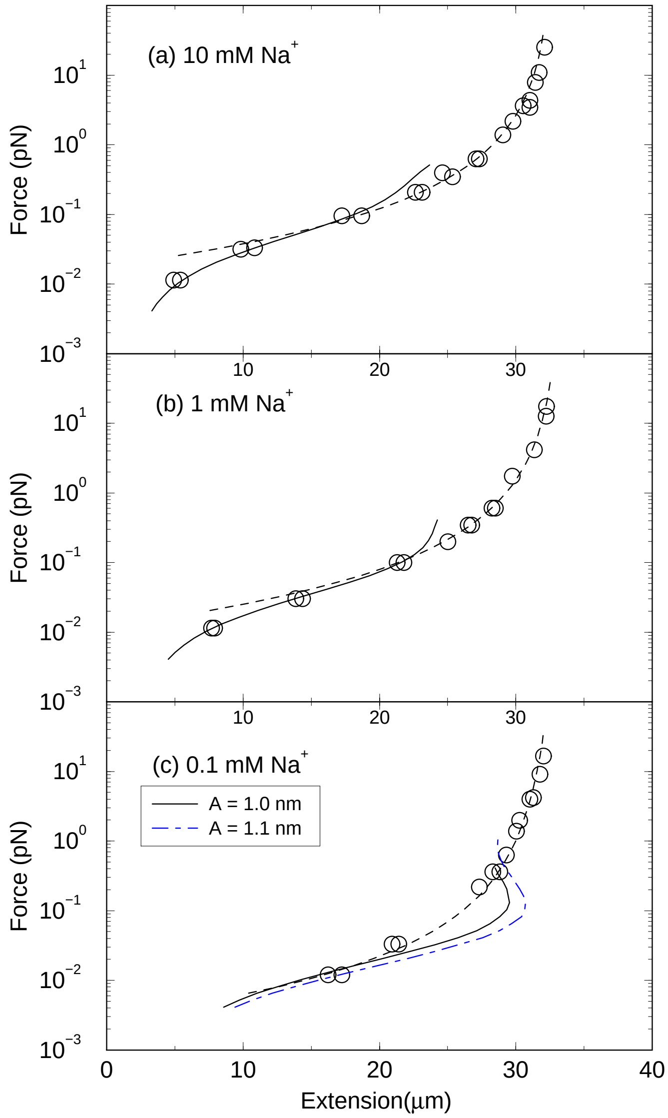

Fig. 2. Comparison between theoretical and experimental force-extension curves for DNA at various salt concentrations. (a) 10 mM Na$^+$, (b) 1 mM Na$^+$, (c) 0.1 mM Na$^+$ The symbols denote experimental results, and the dashed lines represent the functional integral estimation for high force limit. The force *vs.* extension curves for small force regime (solid lines) are calculated by numerically solving the self-consistent equation. In panel (c) the solid line is calculated with $A = 1.0$ nm while the dot-dashed line is computed with $A = 1.1$ nm. In all cases $L = 32.7$ μm and $l_0 = 53$ nm.

The choice of A, for which direct measurements are not available, requires explanation. We choose the effective charge density $1/A$ as 2.5 nm^{-1}, 1.4 nm^{-1} and 1.0 nm^{-1} for 10 mM Na$^+$, 1 mM Na$^+$, 0.1 mM Na$^+$ ion concentration respectively. These values give the best fit to the data. At large salt concentration (10 mM Na$^+$), the interaction range (Debye radius) $r_{\rm D} = 0.3$ nm is smaller than the size of each base of DNA. We conclude that counterion condensation is not relevant in this concentration range. If every base pair carries a charge $-1e$, the linear charge density will be 2.94 nm^{-1} since the size of base pair is approximately 0.34 nm. The choice of our effective charge density $1/A = 2.5$ nm^{-1} at 10 mM NaCl concentration indicates that most of the counterions dissociate from the monomers on the backbone. At smaller salt concentration, namely 1 mM Na$^+$ and 0.1 mM Na$^+$, the interaction range exceeds the Bjerrum length of aqueous solution, $l_{\rm B} = 0.7$ nm. We expect counterions are condensed in the vicinity of DNA, which leads to a reduction in the effective charge density. According to the Manning condensation theory [22,23], the charge density larger than one per Bjerrum length leads to counterion condensation. Therefore we choose $1/A = 1.4$ nm$^{-1} \approx 1/l_{\rm B}$. We used smaller value of $1/A$ for 0.1 mM concentration. However, all the data points from experiment correspond to the rod-like conformation even in the small force regime. We expect the self-consistent theory to be valid even at very smaller values of force, $f \leq 10^{-2}$ pN, and the approximate choice of $1/A$ is then of the order of $1/l_{\rm B}$.

It is clear from Figure 2 that there are two different regimes in the chain elasticity as the applied force increases. They correspond to the "coil-like" and rod-like conformations respectively. At a given ionic concentration, we can observe a plateau in the intermediate force regime. The self-consistent mean field calculation is valid before the onset of plateau, where "coil-like" conformations dominate. Our calculations provide the correct estimation of the chain extension when the extension z is much smaller than the total contour length L *i.e.* $z/L < 0.5$. The mean square average $\langle R^2 \rangle^{-1/2}$ with zero force is proportional to $\kappa^{-1/2}$ and increases as $f\kappa^{-1}$ as the magnitude of force increases. These results agree with experiments. At smaller ionic concentrations, the position of the plateau moves to the smaller values of force. In the following section, we discuss the force-extension curves for a semiflexible chain in a rod-like conformation.

4 Electrostatic effects on stretching stiff DNA: Large force regime

In this section, we apply the functional integral method to obtain the force-extension relation of an intrinsically rigid chain in the limit of large force. This regime was considered by Marko and Siggia [8] who noted that the effects of electrostatic interactions can be absorbed into an effective scale dependent persistence length. Here, we provide a derivation of this result using a functional integral approach.

It appears that when a large force is applied to the chain, the segments of the chain do not interfere with each other geometrically, because of tension induced stiffening occurs on scales from $\xi_{\rm P} = k_{\rm B}T/f$. If this assumption is valid the influence of the electrostatic interaction can be treated simply by replacing the persistence length $l_{\rm p}$ by an effective persistence length $l_{\rm p}^{\rm eff} = l_0 + l_{\rm OSF}$. With this replacement the force-extension curve can be easily calculated. Since the backbone is intrinsically stiff it follows that the fluctuation in angle $\theta(s)$, $\cos\theta(s) = \mathbf{u}(s)\cdot\mathbf{u}(0)$, is small, we can expand $\cos(\theta(s)) \approx 1+\frac{1}{2!}\theta^2(s)+\frac{1}{3!}\theta^3(s)+...$ If we consider only fluctuations of the angle, the expansion with respect to $\theta(s)$ guarantees the constraint of $|\mathbf{u}(s)^2| = 1$. The Hamiltonian of the system can be written in terms of $\theta(s)$ as,

$$\begin{aligned}\frac{H}{k_{\mathrm{B}}T} \propto{}& \frac{l_{\mathrm{p}}}{2}\int_0^L (\frac{\partial \mathbf{u}(s)}{\partial s})^2 \mathrm{d}s \\ &+ \frac{\omega}{2}\int_0^L\int_0^L \mathrm{d}s''\mathrm{d}s'\theta(s'')\theta(s')G^{-1}(s''-s') \\ &- \int_0^L \frac{|\mathbf{f}(s)|}{k_{\mathrm{B}}T}\cos\theta(s)\mathrm{d}s = \frac{1}{2}\int_{-\infty}^{\infty}\tilde{\theta}^2(q)Q^{-1}(q)\mathrm{d}q\end{aligned} \quad (20)$$

$$\tilde{Q}^{-1}(q) = \tilde{G}^{-1}(q) + l_{\mathrm{p}}q^2 + \frac{\mathbf{f}}{k_{\mathrm{B}}T} \quad (21)$$

where $\tilde{\theta}(q) = \sqrt{\frac{1}{2\pi}}\int_{-\infty}^{\infty} \mathrm{e}^{\mathrm{i}qs}\theta(s)\mathrm{d}s$, and in the limit of $q/\kappa \ll 1$,

$$\begin{aligned}\tilde{G}^{-1}(q) &= \frac{\omega}{2}[(1+\frac{1}{q^2\kappa^{-2}})\ln(1+q^2\kappa^{-2}) - 1] \\ &\approx \frac{\omega(q/\kappa)^2}{4} - \frac{\omega}{6}(q/\kappa)^4 + O((q/\kappa)^6).\end{aligned} \quad (22)$$

Therefore, the effective persistence length $l_{\mathrm{p}}^{\mathrm{eff}}$ can be identified with the coefficient of the quadratic term: $l_{\mathrm{p}}^{\mathrm{eff}} = l_{\mathrm{p}} + l_{\mathrm{OSF}}$, $l_{\mathrm{OSF}} = l_{\mathrm{B}}/4\kappa^2A^2$.

The generating function Z for the theory given in equation (20) is

$$\begin{aligned}Z \propto \int D[\theta]\exp\Bigg[&-\frac{\omega}{2}\int_0^L\int_0^L \theta(s'')G^{-1}(s''-s')\theta(s')\mathrm{d}s''\mathrm{d}s' \\ &+ \int_0^L \frac{|\mathbf{f}|}{k_{\mathrm{B}}T}\theta(s)\mathrm{d}s\Bigg].\end{aligned} \quad (23)$$

The ratio of the extension to the total length $\langle z/L\rangle$ can be obtained from the generating function Z

$$\langle \frac{z}{L}\rangle = 1 - \frac{\langle\theta^2(s)\rangle}{2} = 1 - \frac{1}{2}\frac{\partial}{\partial f}\frac{\partial}{\partial f}Z \quad (24)$$

where the mean square value of $\theta(s)$ can be found from $\tilde{Q}^{-1}(k)$,

$$\begin{aligned}\langle\theta^2(s)\rangle &= \frac{1}{2\pi}\int_{-\infty}^{\infty}\mathrm{d}k\int_{-\infty}^{\infty}\mathrm{d}k'\langle\theta(k)\theta(k')\rangle \mathrm{e}^{\mathrm{i}s(k-k')} \\ &= \frac{1}{2\pi}\int_{-\infty}^{\infty}\mathrm{d}k \mathrm{e}^{2\mathrm{i}sk}Q(k).\end{aligned} \quad (25)$$

In Figure 2 we show z/L (Eq. (24)) as a function of force. Our results (see Fig. 2) are in very good agreement with experimental results of Smith *et al.* [1] for all salt concentrations. In the large force regime, we find $\langle z/L\rangle \propto -1/\sqrt{f}$ which, of course implies, that is regime DNA does behave as WLC [8,9]. It is not surprising that the theoretical results for large forces start deviating when $z/L < 0.4$, and dramatically depart from the experimental results and when $z/L < 0.2$ "coil-like" conformations dominate at small forces. This suggests that for small force regime a more elaborate theory, such as the one presented in the previous section, is required.

5 Conclusions

In this paper we have considered the effects of electrostatic interactions on the stretching of DNA. It is already well established that the simple elastic model at high salt concentration gives an excellent description of the response of DNA to tension. Furthermore, it is clear that at small values of the concentration of salt and at small force "coil-like" conformation become important and an elaborate theory is required. Here we have shown that when the applied force is small, the long range interaction in DNA molecule can be properly treated by self-consistent variational mean field approximation. Our theory gives excellent agreement with experimental results in this regime. The larger the ionic strength, the larger the overlap range in the cross over regime from the mean field calculation (coil conformation) to the WLC limit (stretched conformation). If the screening length r_{D} is large compared to the total contour length L ($r_{\mathrm{D}}/L \sim 1$), then electrostatic interactions dominate at all scales so that the chain is effectively stiffened even when the applied force is small. In this case, the self-consistent theory is valid only in the limit of very small force.

One of the limitations of treating the electrostatic interactions, even at the primitive Debye-Hückel level, is that there is no easy way to choose the linear charge density $1/A$. The value of A is essentially controlled by counterion condensation effects. Since the conformation of DNA changes upon addition of salt dramatically a proper theory of describing DNA elasticity should include fluctuations due to counterion condensation. In the absence of such a theory we have used physical arguments to choose a value for A. A more elaborate theory that would treat fluctuations on small scales of the value of A as well as larger length scale is required to obtain the line density independently.

It appears that in the limit of small f the self-consistent variational theory may be adequate for all values for κ. For large value of κ and f the effects of electrostatic interaction may be treated using such a scale dependent persistence length [8]. It would be desirable to have a unified theory that can treat both regimes, and hence the case of intermediate values of f and κ. Such a theory would require using a more elaborate variational Hamiltonian, perhaps, similar to the ones used recently to treat the persistence length of polyelectrolyte chains [19].

Despite the success of the theory outlined here it is worth pointing out that the WLC can only explain the force-extension curves when the counterion is monovalent. It is only in the presence of monovalent ions that the electrostatic persistence length of DNA displays the well accepted dependence on κ. Baumann *et al.* [10] have found that multivalent salt ions (counterions) have dramatically different effects on persistence length of DNA.

By performing force measurement using laser tweezers they discovered that not only valence but also the shape of counterions profoundly affect elasticity of single DNA molecules. In order to fully understand these experiments effects due to the counterion condensation should also be taken into account. For the monovalent ions we have accounted for counterion condensation effects simply by using the mean distance between charges A as adjustable. These observations and other findings by Baumann *et al.* [10] clearly suggest that theories and simulations that go beyond the simple Debye-Hückel theory will be required to provide a complete description of the response of DNA to external tension.

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References

1. S.B. Smith, L. Finzi, C. Bustamante, Science **258**, 1122 (1992).
2. T.R. Stick, J.E. Allemand, D. Bensimm, A. Bensimm, V. Crognelle, Science **271**, 1835 (1996).
3. T.T. Perkins, D.E. Smith, R.G. Larson, S. Chu, Sience **268**, 83 (1995).
4. M. Doi, S.F. Edwards, *The Theory of Polymer Dynamics* (Oxford University Press: Oxford, U.K., 1986).
5. M. Fixman, J. Kovac, J. Chem. Phys. **58**, 1564 (1973).
6. J. Kovac, C.C. Crabb, Macromolecules **15**, 537 (1982).
7. F. Brochard-Wyart, Europhys. Lett. **23**, 105 (1993): F. Brochard-Wyart, H. Hervet, P. Pincus, Europhys. Lett. **26**, 511 (1994).
8. J.F. Marko, E.D. Siggia, Macromolecules **28**, 8759 (1995); Science **265**, 1599 (1994).
9. B.-Y. Ha, D. Thirumalai, J. Chem. Phys. **106**, 4243 (1997).
10. G. Baumann, S.B. Smith, V.A. Bloomfield, C. Bustamante, Proc. Natl. Acad. Sci. **94**, 6185 (1997).
11. J.L. Barrat, J.F. Joanny, Europhys. Lett. **24**, 333 (1993).
12. T. Odijk, J. Polym. Sci. Polym. Phys. Ed. **15**, 477 (1997).
13. J. Skolnick, M. Fixman, Macromolecules **10**, 944 (1977).
14. B.-Y. Ha, D. Thirumalai, J. Chem. Phys. **103**, 9408 (1993).
15. J.B. Lagowski, J. Noolandi, B. Nickel, J. Chem. Phys. **95**, 1266 (1991).
16. P. Pincus, Macromolecules **9**, 386 (1976).
17. S.F. Edwards, P. Singh, J. Chem. Soc. Faraday Trans. II **75**, 1001 (1979).
18. P. Haronska, J. Wilder, T.A. Vilgis, J. Phys. II France **7**, 1273 (1997).
19. R. Netz, H. Orland, Eur. Phys. J. **8**, 81 (1999).
20. B.-Y. Ha, D. Thirumalai, J. Chem. Phys. **110**, 7533 (1999).
21. U. Micka, K. Kremer, Europhys. Lett. **38**, 279 (1997).
22. G.S. Manning, J. Chem. Phys. **51**, 954 (1969).
23. J.L. Barrat, J.F. Joanny, Adv. Chem. Phys. **94**, 1 (1996).