

Osmotic properties of DNA: Critical evaluation of counterion condensation theory

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The osmotic coefficient of *B*-DNA in water may, in dilute solutions, deviate by as much as 100% from predictions based on a simple line-charge “counterion condensation” theory. In contrast, a cell model description of the ionic atmosphere near a cylindrical polyelectrolyte predicts osmotic properties that are in surprisingly good harmony with all available experimental findings over a wide range of DNA concentrations. We argue that the neglect of molecular features, such as finite radius, makes line-charge condensation theory inapplicable at all but impractically low polyelectrolyte concentrations.

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Ionic screening of charge interactions remains one of the most vigorously discussed properties of polyelectrolyte solutions [1]. The osmotic pressure of *B*-DNA in dilute aqueous solutions [2,3] shows as much as factor-of-2 deviations from predictions based on the influential counterion condensation theory of Oosawa and Manning (OM) [4–6]: We shall argue that an earlier theory pioneered by Lifson and Katchalsky (LK) [7,8], based on the cell model formulation of the nonlinear Poisson-Boltzmann equation, provides a more successful starting point than counterion condensation theory for the osmotic properties of rigid cylindrical polyelectrolytes under low-salt conditions. Indeed, when the axial separation between cylinders is smaller than the persistence length, say, their osmotic properties are dominated by electrostatic effects that are accurately described by the LK cell model. By explicitly including the finite size of the cylinder, this cell model fundamentally disagrees with the line-charge OM picture. Neglect of finite radius is the main reason for the osmotic failure of OM theory [9].

We consider a rigid (hollow or solid), charged cylindrical polymer of radius a , coaxially enclosed in a cylindrical (Wigner-Seitz-like) “cell” whose radius R corresponds to the total system volume per polymer length. Because the cell acts as a neutralization volume for the counterions, the electric field vanishes at the cell wall. Counterions organize within the cell according to the nonlinear Poisson-Boltzmann (PB) equation for the double layer electric potential, u . In potential units of $k_B T/e$, and in the absence of added salt, this equation has the form

$$\frac{1}{r} \frac{d}{dr} \left(r \frac{du_*(r)}{dr} \right) = -\frac{\kappa_*^2}{2} e^{-u_*(r)}. \quad (1)$$

Here κ_*^{-1} denotes “screening length,” * for “inside” or “outside” the polyelectrolyte cylinder. Potentials $u_{in}(r)$ and $u_{out}(r)$ determine the charge densities,

$$n_*(r) = n_{*,0} e^{-u_*(r)}, \quad (2)$$

where $n_{*,0} = \kappa_*^2 (8\pi l_B)$, and $l_B = e^2/(\epsilon k_B T)$ is the Bjerrum length.

The major and minor grooves in DNA [10] require that we treat its “cylinder” as at least partly hollow to allow for solvent and ion access to the space within the grooves. Here, for simplicity to recognize the maximum possible range of “nonspecific” accumulation of countercharge near the cylinder, we consider cylinders that are either solid or completely hollow. Counterion accumulation is formulated via the PB equation with continuous variation in density at $r=a$, $u_o(r \geq a)$ and $u_i(r \leq a)$ are evaluated relative to a zero at the cell wall ($r=R$) with $\kappa_*^2 = \kappa^2 = 8\pi l_B n(R)$, where $n(R)$ is the number density of counterions at the cell wall.

For $a < r < R$ [7,8]

$$u_o(r) = \ln \left[\frac{(kr)^2}{2z} \cos^2 \left(2 \ln \left[\frac{r}{R_M} \right] \right) \right]. \quad (3)$$

For $r < a$ [11]

$$u_i(r) = u_o + 2 \ln(1 + cr^2). \quad (4)$$

The integration constants z , R_M , κ , c , and u_o are obtained from boundary conditions: $u_i(a) = u_o(a)$, $du_i(r)/dr|_0 = 0$, $du_o/dr|_R = 0$, and $[du_o(r)/dr - du_i(r)/dr]|_a = 2Q/a$. For DNA the dimensionless linear charge density $Q = l_B/l_{\text{DNA}} \approx 4.353$ is determined by the charge separation $l_{\text{DNA}} \approx 1.7 \text{ \AA}$ and the Bjerrum length $l_B \approx 7.14 \text{ \AA}$ for water at room temperature. The parameters are fixed by an iteration that starts with a trial partitioning of counterions, Q_i and Q_o ($Q_i + Q_o = Q$), inside and outside $r=a$ refined until the boundary conditions are exactly satisfied.

In salt-free solutions the osmotic pressure is

$$\pi_{osm} = k_B T n(R) \quad (5)$$

on the cell wall. Because the electric field vanishes at the cell wall, the Maxwell stress is zero at $r=R$.

When the density of osmotically active buffer salt, $2n_o$ for monovalent salts, cannot be ignored, the cell model predicts that $\pi_{osm} = k_B T [n_+(R) + n_-(R) - 2n_o]$. $n_{\pm}(R) = n_o \exp[\pm u(R)]$; $u(R)$ is the difference in reduced potential between the cell wall and the bathing salt solution. In the artificial, but popular, “Donnan” limit [4,12,13] where the

variation in potential $u(r)$ within the cell is assumed to be small compared with $u(R)$ (as though $R \rightarrow a$), we decompose $n_+(R) \rightarrow n(R) + n_i, n_-(R) \rightarrow n_i$, where $n(R)$ is the counterion density in the absence of salt. In electrochemical equilibrium, the osmotically active fraction of counterions inside the polyelectrolyte assembly n_i is determined by

$$n_i(n(R) + n_i) = n_0^2, \quad (6)$$

(from cancellation of $\exp[\pm u(R)]$ factors). This approximation may be expected to work well when the concentration of osmotically active buffer salt $2n_o$ is small compared to the concentration of osmotically active counterions $n(R)$. Then the osmotic pressure is approximately

$$\pi \approx k_B T [n(R) + 2(n_i - n_o)]. \quad (7)$$

Though popular, the approximation employed here is difficult to justify from first principles [13,14].

By definition, the osmotic coefficient ϕ is the ratio of the actual osmotic pressure to the osmotic pressure of a hypothetical gas of uniformly distributed counterions, the number density $n(R) + 2(n_i - n_o)$ divided by the mean density $n_{\text{DNA}} = 1/(l_{\text{DNA}} \pi R^2)$:

$$\phi = \frac{n(R) + 2(n_i - n_o)}{n_{\text{DNA}}} \quad (8)$$

Figure 1 shows computed osmotic coefficients of rigid hollow or solid cylinders as a function of the molar concentration of DNA c_{DNA} for $c_o = 0, 2,$ and 10 mM.

(i) With no added salt ($c_o \approx 0$ mM), osmotic coefficients vary slowly vs c_{DNA} in good agreement with experiments [2] (diamonds). However, simple condensation theory (flat, dashed line) predicts no variation at all.

(ii) For concentrations of DNA phosphates from $c_{\text{DNA}} = 1 - 500$ mM, the calculated osmotic coefficients agree reasonably closely with experiment [2,3]: for $n_o \ll n_{\text{DNA}}$ [2], $\phi \approx 0.16$, compared to $\phi = 1/(2Q) \approx 0.11$ for line-charge condensation theory.

(iii) For $f = 2n_o/n(R)$ small but finite, the Donnan equilibrium approximation works well (2 mM and 10 mM data [3]).

Figure 2 shows the osmotic pressure π_{osm} vs molar phosphate concentration c_{DNA} for 2–300 mM [2,3] and 1–2 M [15]. As long as the ratio f is small, the simplest electrostatics model (dotted line for hollow cylinders and solid lines for solid cylinders) indeed explains quite well the magnitude and variation of the data. The hollow-cylinder model (dotted-line) gives a slightly better description of the data than the solid-cylinder model [16].

When the concentration of DNA is small, $c_{\text{DNA}} < 300$ mM, the simple Donnan-equilibrium model [Eqs. (7) and (8)] describes the experimentally observed increase in pressure vs c_{DNA} [3] (triangle, squares). In the high DNA-concentration range, the calculated variation of the pressure with concentration is clearly slower than that observed experimentally. The difference may be of nonelectrostatic origin [17] or may reflect charge discreteness [18]: salt concentration is likely to be unimportant here; renormalization

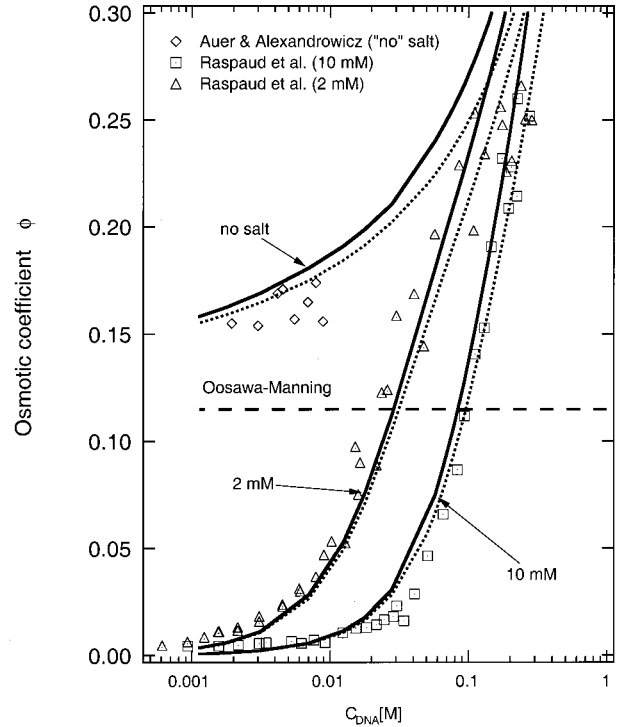


FIG. 1. Computed and measured osmotic coefficients, Eq. (8), vs c_{DNA} (molar DNA phosphates). The horizontal dashed line is the prediction of Oosawa-Manning line-charge condensation theory. Solid and dotted lines are computed for solid and hollow charged cylinders of radius $a = 10$ Å and linear charge density $1e/1.7$ Å contour length (or $Q = 4.353$). The “no-salt” curves are calculated from Eq. (5), data (diamonds) from Ref. [2]; lower curves computed from Donnan equilibrium model, Eqs. (6) and (7) for DNA in 2 mM and 10 mM salt solution, data (triangles and squares) from [3]. Concentrations $n(R), n_i, n_o$ are in number density units. Note complete irrelevance of OM predictions.

effects due to chain conformational fluctuations, discussed in Ref. [15], lead to predictions of decreasing rates of change of the pressure with concentration. Recent work on stretching of DNA under various ionic conditions [19] also suggests that there might be an additional strong coupling between DNA elasticity and electrostatics. Though the details of this coupling are only beginning to be elucidated [20] it is conceivable that local deformations of DNA would change the countercharge distributions at low-salt conditions and thus affect also the osmotic pressure.

Given the qualitative failure of Oosawa-Manning [4,5] line-charge condensation model (Figs. 1 and 2, dashed lines) what can it teach us about the counterion accumulation around any linear polyelectrolyte? In this OM picture, highly charged and rigid cylindrical macromolecules are portrayed as line charges with explicit neglect of finite macromolecular radius. The atmosphere of counterions is viewed as strongly perturbed: for $Q > 1$, condensation will bring down the effective (dimensionless) line-charge density from Q to 1; a fraction $f_c = (Q - 1)/Q = 1 - 1/Q$ of the counterions condense. The remaining fraction $f = 1/Q$ of the charge remains unbound and osmotically active. If the unbound fraction is

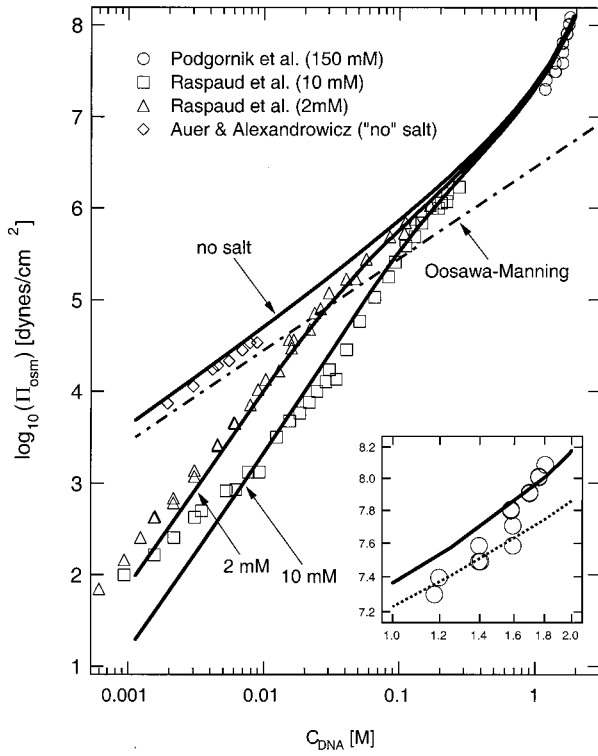


FIG. 2. Computed and measured osmotic pressures vs c_{DNA} . Dashed line, prediction of Oosawa-Manning line-charge model. Solid line, computations for solid cylinders, and data-point symbols as in Fig. 1. Except at high pressures (inset) on the log-log plot, the difference between hollow-and solid-cylinder predictions are difficult to detect. The arrows at $c_{\text{DNA}}=2$ mM, and 10 mM show that the Donnan finite salt correction goes awry when $2n_0/n(R)$ is no longer small, i.e., to the left of the arrows. Inset: solid and dotted lines for pressures calculated at high density for solid and hollow cylinders; circles data from [15]. Computed forces are of the same magnitude as those measured but a different function of concentration.

modeled as a polarized Debye-Hückel gas of counterions, this gas contributes to the osmotic coefficient ϕ , as $\phi_M = \pi/(k_B T n_Q) = f/2 = 1/(2Q)$. For DNA $\phi_M \approx 0.11$, with a significant fraction $f \approx 0.75$ of countercharge “bound.”

In the line-charge limit [4,5] there is no disagreement at all between cell model predictions and predictions based on condensation theory. In the cell model it is possible to define an effective “condensation range” [8] or a Manning radius R_M such that the fraction of the countercharge contained within a shell $a < r < R_M$ is $f = 1 - 1/Q_o$, the fraction of charges that are predicted to condense on the line-charge. In the small a/R limit, $R_M \approx R e^{-\pi/2z} \approx \sqrt{Ra}$. That is in this limit the Manning radius disappears; the effects of the fraction $f = 1 - 1/Q_o \rightarrow 1 - 1/Q$ of ionic atmosphere can be absorbed in a redefinition of the line charge, as in condensation theory. However when $a > 0$ the Manning radius R_M is finite and even diverges as one approaches the infinite-dilution limit $R \rightarrow \infty$.

Consider the asymptotic expression for the salt-free osmotic coefficient in the limit where a/R is small (the line-charge limit, or the infinite-dilution limit): from the deriva-

tives of the outer solution Eq. (3)

$$Q_o = 1 - z \tan\left(z \ln\left[\frac{a}{R_M}\right]\right) \quad (9)$$

$$0 = 1 - z \tan\left[z \ln\left(\frac{R}{R_M}\right)\right] \quad (10)$$

$$\kappa^2 R^2 = 4(1 + z^2). \quad (11)$$

we determine z :

$$\ln\left(\frac{a}{R}\right) = \frac{\arctan\left(\frac{1-Q_o}{z}\right) - \arctan\left(\frac{1}{z}\right)}{z}. \quad (12)$$

When a/R is small, $z \rightarrow 0$, the $\arctan(\cdot)$'s may be replaced by, respectively, $-\pi/2$ and $\pi/2$. Then z has a weak logarithmic dependence on a/R_0 :

$$z \approx \frac{\pi}{\ln\left(\frac{R}{a}\right)}. \quad (13)$$

The corresponding asymptotic expression for the osmotic coefficient is

$$\phi = \frac{n(R)}{n_{\text{DNA}}} = \frac{\kappa^2}{8\pi l_B} \pi R^2 l_{\text{DNA}} = \frac{1}{2Q_o} (1 + z^2) G\left(\frac{a}{R}\right). \quad (14)$$

$G(a/R)$ is a geometric factor that equals 1 for hollow cylinders, and is $1 - (a/R)^2$ for solid cylinders. Therefore, for small values of a/R , in the absence of salt, Eqs. (13) and (14) give

$$\phi \approx \frac{1}{2Q_o} \left(1 + \frac{\pi^2}{\ln^2\left(\frac{R}{a}\right)}\right). \quad (15)$$

Any concentration dependence of the osmotic coefficient is incompatible with simple condensation theory. Only impractically slowly does the osmotic coefficient approach the line-charge OM limit.

Is it fair to test the OM model against low-salt-concentration data when that model is usually applied to high-salt systems? The foundation of line-charge condensation theory is a condition that applies only in the limit of zero added salt. Recall that the electrostatic potential around an isolated charged line varies as $\ln(r)$. Paradoxically, line-charge condensation theory is usually derived for string-of-beads linear polyelectrolytes in salt solutions whose screening lengths are much too short even to allow the $\ln(r)$ potentials on which the concept of line charge condensation

is based. For example, the radius of the ‘line’ would have to be much less than the 10 Å screening length of the 0.1 M solution in which 10-Å-radius DNA usually lives.

Will it ever be instructive to reformulate [8,21–23] or “extend” line-charge condensation theory to include finite macromolecular radius and other molecular features? Reformulations would have to be merely linguistic. We see nothing

to be gained by retrofitting a structurally accurate model of counterion organization to the fiction of a line-charge model.

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