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Formation of Loose Clusters in Polyelectrolyte Solutions

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ABSTRACT: We report further development of a pair potential for two identically charged rodlike polyions oriented in parallel. An attractive force mediated by condensed counterions of any valence, including univalent, is identified for separation distances between a Debye screening length and about a third of a Debye length. The characteristics of the potential are explored as a function of ionic strength, dielectric constant, counterion valence, and charge density of the polyions. The work required to assemble more than two polyions in parallel array is shown not to be pairwise additive. The free energy of assembling many parallel polyions in a hexagonally ordered cluster is calculated as a function of the number N of polyions in the cluster. The free energy has a minimum at a finite cluster size. Discrepancies between the theoretical structure of the stable cluster and experimental observations on polyelectrolyte clusters (with univalent counterions) are discussed, and it is tentatively concluded that the apparently loose and disordered clusters observed in solution may be understood as stabilized by relatively few pairwise attractive interactions.

1. Introduction

Aqueous polyelectrolyte solutions with univalent counterions exhibit a tendency toward polymer cluster formation. DNA clusters in aqueous NaCl have been visualized in the electron microscope.[1] The conditions of their appearance were found to be correlated with conditions required for detection of DNA aggregates from static and dynamic light scattering data. The phenomenon is salt-dependent. The DNA clusters were more stable at lower NaCl concentrations. Nucleosomal DNA fragments 160 base pairs in length were utilized in the experiment, an indication that polyelectrolyte clustering does not require chains of high molecular weight. A slow relaxation time in dynamic light scattering, the so-called slow mode, has been observed at low DNA concentration for a 20 base pair oligonucleotide[2] and for dilute solutions of poly(styrenesulfonate) with a molecular weight as small as 5000 (about 24 monomers).[3,4] Static light scattering intensities from sonicated DNA fragments are consistent with a cluster size of about 1400 Å.[5] For poly(vinylpyridinium chloride) in D_2O small-angle neutron scattering and static light scattering converge on a cluster size of about 900 Å.[6] The slow mode in light scattering is a polyelectrolyte effect. It is not present in a neutral poly(methacrylic acid) solution but emerges when NaOH is added to partially ionize the weak polyacid.[7] Analogous behavior (onset of clustering when a neutral polymer is ionized) has been observed in other systems.[8]

Energy transfer experiments were performed on aqueous solutions of poly(L-lysine) in NaCl.[9] When the Förster radius of the donor−acceptor pair was 28 Å or less, only baseline transfer was observed as the NaCl concentration was lowered. However, when the NaCl concentration was lowered in the case of a Förster radius of 58 Å, a clear enhancement of energy transfer was seen. This experiment shows that polymer segments in a poly(L-lysine) cluster approach to within about 60 Å but stay further away than about 30 Å. The attractive interaction responsible for clustering is not a short-range "contact force".

We have found in the framework of counterion condensation theory that the potential of mean force between two parallel identically charged rodlike polyions with univalent counterions features a deep attractive free energy well located inside a Debye length at intermediate distances.[10,11] The attraction is caused by a large increase of the internal partition function of the condensed counterions as the polyions approach (see Figure 8 of ref 10, or Figure 3 of ref 11). A qualitative physical interpretation could be that the condensed counterions spread entropically into the region between the polyions, unimpeded by forcible attraction back to their polyion of origin, since superposition of the electric fields of the two polyions creates a flat potential energy landscape between them (see Figure 11 of ref 10). Or, the augmented electric field between the two polyions further electrostricts the degrees of freedom of polar solvent molecules, extending local regions of low dielectric constant, which in turn provide an expanded low-energy environment for the condensed counterions. In either case, or as the combined effect of these and possibly other mechanisms, the attractive interaction can be conceptualized as a "covalent bond", in which the condensed counterions are the shared "electrons".[10] In this paper, we address the question of whether the attraction leads to discrete polymer clusters, as indicated by experiments, or to macroscopic phase separation and precipitation. The literature contains examples of other approaches to this question.[12,13]

2. Theory

2.1. The Interaction of Two Polyions. Here we summarize previous results[10,11] for a pair of identical rodlike polyions oriented in parallel (or parallel segments of flexible polyions with persistence length), while making some improvements. Recall our model for a polyion— a linear array of P point charges with fixed spacing b—and the associated dimensionless measure of charge density $\xi = l_B/b$, where l_B is the Bjerrum length q^2/Dk_BT, q the protonic charge, D the dielectric constant of bulk solvent, and k_BT the product of the Boltzmann constant and Kelvin temperature (there are

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ξ charges in a Bjerrum length polymer segment). We consider the potential of mean force $w_2(r)$ for two polyions, that is, the reversible work required to bring the two polyions from infinite separation to mutual distance r. The formulas for $w_2(r)$, as well as for all free energies in this paper, are expressed in units of Pk_BT. They are therefore dimensionless and represent free energies per charged site on one polymer (in our previous papers,[10,11] the units were $2Pk_BT$). The two polyions are immersed in a Z: Z' electrolyte solution, where Z is the absolute value of the valence of the counterion and Z' the absolute value of the valence of the coion.

In the theory the pair potential $w_2(r)$ comes in three separately calculated pieces: for near distances r, for intermediate distances, and for far distances. Near distances are those which remain asymptotically constant as the Debye screening length κ^{-1} of the electrolyte solution tends to infinity. Roughly, these distances occur when the two thin rodlike polyions have approached to distances well within their Debye atmospheres. Their electrical charges are not mutually screened in the Debye–Hückel sense. Far distances scale asymptotically with the Debye length. Two polyions separated at such a distance are screened by their Debye–Hückel clouds. Intermediate distances tend to infinity like a fraction of a Debye length, so two polyions of intermediate separation are only partially screened by their diffuse ionic clouds.

In our previous work, we did not specify the positions of the boundaries between the near and intermediate regions and between the intermediate and far regions, so there were gaps in the composite curve of $w_2(r)$. In a more recent theory of the counterion–polyion and coion–polyion pair potentials,[14] the same three regions of space were used, but we also produced reasonable, albeit approximate, positions for the boundaries. Our estimate is that the near region extends from some cutoff distance of closest approach $r = a$ to $r = e^{-1}\kappa^{-1}$, where e is the base of natural logarithms, and that the intermediate region falls between $r = e^{-1}\kappa^{-1}$ and $r = \kappa^{-1}$. The far region then consists of all separations larger than a Debye length. We also have a procedure, again approximate, for generating a continuous potential. However, the continuous potential is only piecewise smooth. It is smooth within each region but does not cross the interregional boundaries smoothly.

We recall the basic elements of the theory. For two parallel polyions separated by distance r, there are three components of free energy: the ionic interactions within each polymer, the ionic interactions between the two polymers, and the work of transfer of a subpopulation of the counterions from bulk solution to the counterion layer condensed on the two–polymer assembly. To realize the first component $w_p(r)$, we sum over all Debye–Hückel pairwise interactions among renormalized charges $[1 - Z\theta(r)]q_p$ within each polyion. Here, $q_p = q$ for a pair of polycations, or $q_p = -q$ for a pair of polyanions. The fraction of condensed Z-valent counterions per polymer charged site is denoted by $\theta(r)$ (so the total number of condensed counterions is $2P\theta$, and the polymer charge is reduced by the factor $(1 - Z\theta)$). The quantity θ depends on r because the number of counterions condensed on one polyion is influenced by the presence of the other polyion at distance r. We get (in units of Pk_BT)

$$w_p(r) = -2\xi[1 - Z\theta(r)]^2 \ln \kappa b \qquad (1)$$

The condition for validity of this equation is that the screening length κ^{-1} be much greater than the charge spacing b but much less than the length of the rodlike polyion segments $(P - 1)b$. The formula for the Debye screening parameter κ for our Z:Z' electrolyte is

$$\kappa^2 = \lambda c_s \qquad (2)$$

where c_s is the concentration (molarity) of the electrolyte, and

$$\lambda = 4\pi \times 10^{-3} N_A l_B Z Z'(\nu + \nu') \qquad (3)$$

with N_A being Avogadro's number, ν the number of counterions in the formula for the electrolyte, and ν' the number of coions.

The second free energy component $w_{pp}(r)$ is obtained by pairwise summation of Debye–Hückel interactions among renormalized polymer charges, one member of the pair on one polymer, the other on the other polymer. The result is

$$w_{pp}(r) = 2\xi[1 - Z\theta(r)]^2 K_0(\kappa r) \qquad (4)$$

where K_0 is the zeroth-order modified Bessel function of the second kind. The condition for validity of this equation is that the distance r between the polyions be much larger than the charge spacing b within a polyion, so that the summation may be replaced by integration, although we will not interpret this condition strictly in our numerical work. The third component of free energy is the work $w_{\mathrm{transfer}}(r)$ required to transfer $2P\theta$ counterions from the bulk electrolyte to the condensed layer

$$w_{\mathrm{transfer}}(r) = 2\theta(r) \ln [10^3\theta(r)/\nu c_s Q(r)] \qquad (5)$$

where $Q(r)$ is the internal partition function of the layer of counterions condensed on the two–polymer assembly, which is sensitive to the mutual positioning of the polyions.

To bring the reference state at infinity into play, we subtract from each of the components $w_p(r)$, $w_{pp}(r)$, and $w_{\mathrm{transfer}}(r)$ the corresponding components at $r = \infty$ to get the differences $\Delta w_p(r)$, $\Delta w_{pp}(r)$, and $\Delta w_{\mathrm{transfer}}(r)$. The overall potential of mean force for the polyion pair $w(r)$ is then set equal to $\Delta w_p(r) + \Delta w_{pp}(r) + \Delta w_{\mathrm{transfer}}(r)$. At this stage, $w(r)$ contains the unknown functions $\theta(r)$ and $Q(r)$. Both are determined from a single minimization condition, $\partial w(r)/\partial\theta(r) = 0$ for any fixed value of r. The left-hand side of this equation can be viewed as a dilute-solution expansion in electrolyte concentration c_s with retention of only a leading $\ln c_s$ term and a constant term. These two functionally independent terms can separately be set to zero, yielding closed formulas for $\theta(r)$ and $Q(r)$ if the reduced polyion charge density exceeds the threshold value $\xi = 1/Z$. Separate calculations must be made for each of the three ranges of distances r. In the near region, κr is small, and an asymptotic expansion of the Bessel function

$$K_0(\kappa r) \sim -\ln\left(\frac{1}{2}\kappa r\right) - \gamma \qquad (6)$$

where γ is the Euler constant equal to 0.5772..., can be seen to contribute to the $\ln c_s$ dependence and thus to

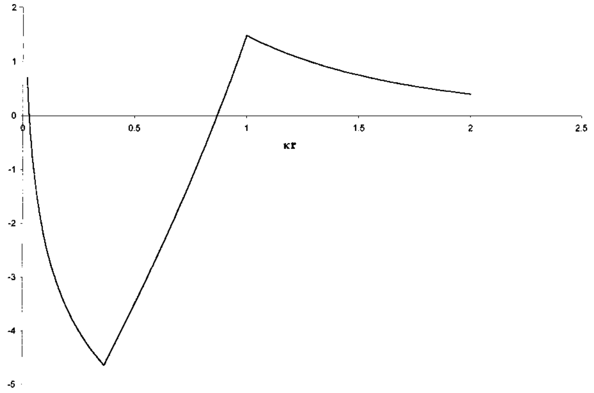

Figure 1. Reduced potential of mean force $w_2(\kappa r)$ (*i.e.*, in units of Pk_BT, where P is the number of charged sites on a single segment) for a pair of identical rodlike polyion segments in parallel orientation. The ratio κb of polymer charge spacing b to Debye screening length κ^{-1} equals 0.02, which corresponds roughly to DNA in aqueous 10^{-3} M NaCl.

the number of condensed counterions, while the constant terms influence the structure (partition function) of the condensed layer. The product κr is also small in the intermediate range of r, and the asymptotic expression for $K_0(\kappa r)$ can be used, but with the definition of r in the intermediate range

$$K_0(\kappa \mathrm{r}) \sim -(1-x)\ln(\kappa r_0) + 2 - \gamma \tag{7}$$

where x can take on values from zero to unity to cover the intermediate range, and r_0 is a fixed distance. A logarithmic dependence on c_s again influences the equilibrium number of condensed counterions. In the far range, κr is asymptotically constant, and the corresponding constant value of $K_0(\kappa r)$ contributes only to the partition function $Q(r)$.

The results for $\theta(r)$ and $Q(r)$ have been given previously.[10,11] We list the formulas for the potential $w_2(r)$, which are improved here by specification of the boundaries of the three regions of distance and by application of a simple procedure for ensuring that $w_2(r)$ be continuous across these boundaries.[14] For $\xi \geq 1/Z$, and $a \leq r \leq (1/\mathrm{e})\kappa^{-1}$

$$w_2(r) = -\frac{1}{2Z^2\xi}\ln\left[e^2\kappa^2b^2\left(\frac{1}{\kappa \mathrm{b}}\,\mathrm{e}^{-K_0(\kappa r)}\right)^{4Z\xi-1}\right] \tag{8}$$

For $(1/\mathrm{e})\kappa^{-1} \leq r \leq \kappa^{-1}$

$$w_2(r) = \frac{2\ln\kappa r}{Z^2\xi(1-\ln\kappa r)}(1+\ln\kappa b) - \frac{2[2Z\xi(1-\ln\kappa r)-1]}{Z^2\xi(1-\ln\kappa r)^2}\ln\left[\frac{1}{\kappa r}\,\mathrm{e}^{-K_0(\kappa r)}\,(\mathrm{e}\kappa b)^{\ln\kappa r}\right]. \tag{9}$$

and for $r \geq \kappa^{-1}$

$$w_2(r) = \frac{2}{Z^2\xi}(2Z\xi-1)K_0(\kappa r) \tag{10}$$

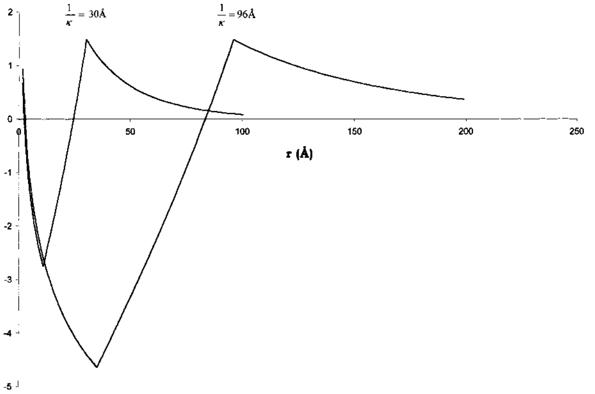

Figure 2. Ionic strength dependence of the reduced pair potential $w_2(r)$. The smaller Debye length (inner curve) corresponds to 10^{-2} M NaCl; the larger Debye length (outer curve), to 10^{-3} M NaCl. Charge spacing $b = 1.7$ Å.

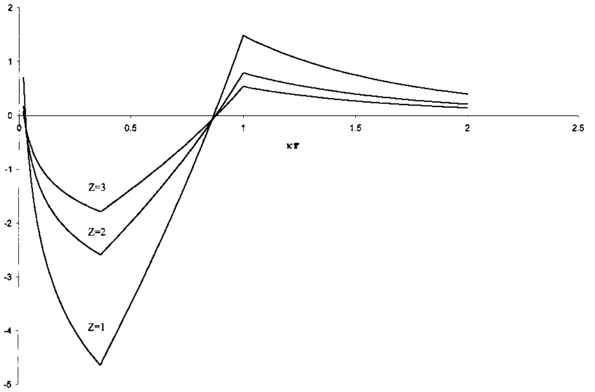

Figure 3. Dependence of the reduced pair potential $w_2(\kappa r)$ on counterion valence Z. $\kappa b = 0.02$.

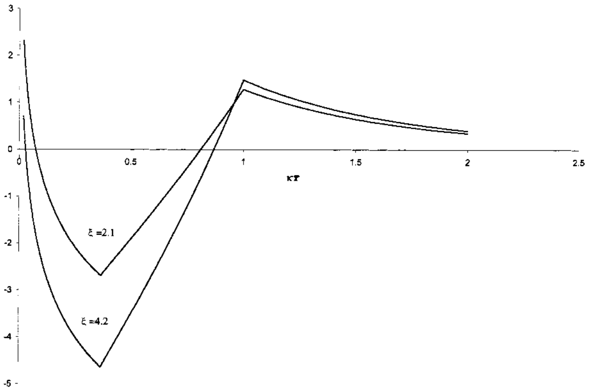

Figure 4. Dependence of the reduced pair potential $w_2(\kappa r)$ on reduced polymer charge density ξ. The Debye screening length is fixed by excess salt at 85 Å (0.0016 M NaCl) and is independent of ξ. Charge spacing $b = 1.7$ Å ($\xi = 4.2$), 3.4 Å ($\xi = 2.1$).

We give some illustrations. Figure 1 highlights the strongly attractive region of $w_2(r)$. The physical origin of the attractive interaction is discussed in detail in ref 10, and Figure 8 of that paper shows the behavior of the partition function of the condensed counterions $Q(r)$, which is primarily responsible for the attraction (in ref 10, the partition function was interpreted as the volume occupied by the condensed counterions). We note here only that the partition function, which reflects the distribution and energetics of the condensed counterions, is exquisitely sensitive to the distance r separating the two polyions. Figure 2 shows that both the range and strength of the attraction diminish at higher ionic strength. Figure 3 compares the effect of counterions of different valence at fixed ionic strength. The strength of the attraction is smaller for the counterion of higher valence, but the range does not change (at fixed ionic strength). In Figure 4, the charge density of the polyion is varied. The strength of the attraction is greater for

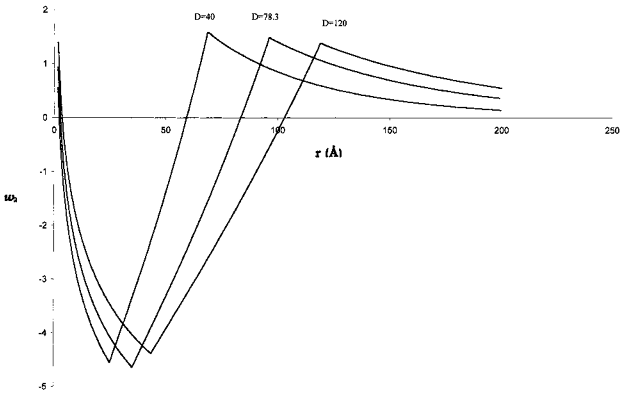

Figure 5. Dependence of the reduced pair potential $w_2(r)$ on solvent dielectric constant D. NaCl concentration is fixed at 10^{-3} M, so the Debye screening length scales as $\sqrt{D}$.

more highly charged polymers, but the range does not change. Finally, we see from Figure 5 that when the dielectric constant of the solvent increases at fixed salt concentration, the range of the attraction increases, but its strength changes only slightly. The possible significance of these properties will be elaborated in the Discussion section, but we can see here that the functional form of $w_2(r)$ is sufficiently complicated to produce qualitatively different behavior for each of the four ways of modulating the electrostatics.

2.2. Nonadditivity of the Pair Potential. The attractive force inherent in the polyion−polyion pair potential $w_2(r)$ is caused by an enhancement of the internal partition function of the condensed counterions as the polyions approach one another.[10,11] If the partition function is modeled in a simple way as consisting only of the volume of the condensed layer, then we can show graphically how the condensed layers disjoint on each individual polyion at large distances, swell on approach of the polyions, and finally merge into a layer held in common by both polyions (like a covalent bond formed by the counterions).[10] For a cluster of N polyions, a single commonly held layer of condensed counterions could not exist if the free energy of the cluster were given as a pairwise sum of terms $w_2(r_{ij})$, where r_{ij} is the distance between the ith and jth polyions of the cluster. We demonstrate nonadditivity for the concrete case of a cluster of three parallel polyions.

The components of the work required to assemble the three polyions from infinity are the same as for two polyions. There are the electrostatic repulsions among effective charges within each polyion, given by eq 1, where now w_p becomes a function of the three polyion−polyion distances through $\theta(r_{12}, r_{13}, r_{23})$ and the initial factor 2 is replaced by 3. There is a collective component accounting for the transfer of counterions from bulk solution to the condensed layer (considered as held in common by the three polyions). This contribution is given by eq 5 with r replaced by the three pairwise distance variables and the initial factor 2 replaced by 3. Finally, there are electrostatic interactions between pairs of polyions (because the free energy minimization involves all three components of the work, this last one does not imply pairwise additivity of the resulting three-body interaction). The formula is analogous to eq 4

$$w_{pp}(r) = 2\xi[1 - Z\theta(r_{12}, r_{13}, r_{23})]^2[K_0(\kappa r_{12}) + K_0(\kappa r_{13}) + K_0(\kappa r_{23})] \quad (11)$$

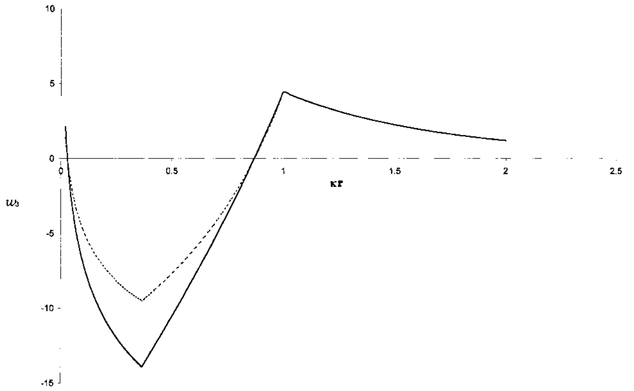

Figure 6. Three-body reduced polymer potential $w_3(\kappa r)$: solid curve, eqs 12−14 of text; dashed curve, pairwise additivity approximation. The rodlike polymers intercept an orthogonal plane at the corners of an equilateral triangle of side r. $\kappa b = 0.02$; $b = 1.7$ Å.

To show that the three-body potential is not pairwise additive, we reduce the problem by arranging the three parallel rods into a simple pattern: they cut an orthogonal cross section at the corners of an equilateral triangle of side r, so that $r_{12} = r_{13} = r_{23} = r$. Then the work required to assemble the three rods from infinity into this pattern is a function only of r, $w_3 = w_3(r)$; near, intermediate, and far regions for r can be defined just as in the two-body problem, and the calculation proceeds in analogous fashion. The result follows, and we see that $w_3(r)$ does not satisfy the pairwise additivity equation $w_3(r) = 3w_2(r)$, except in the repulsive Debye−Hückel-like far region (where the polyions do not share their condensed counterions).

In the near region, $a \le r \le 1/\mathrm{e}\kappa$,

$$w_3(r) = -\frac{1}{Z^2\xi}\ln\left[\mathrm{e}^2\kappa^2 b^2\left(\frac{1}{\kappa b}\,\mathrm{e}^{-K_0(\kappa r)}\right)^{(2/3)(6Z\xi-1)}\right] \quad (12)$$

In the intermediate region, $1/\mathrm{e}\kappa \le r \le 1/\kappa$

$$w_3(r) = \frac{3\ln\kappa r}{Z^2\xi(1-2\ln\kappa r)}\ln(e^2\kappa^2 b^2) - \frac{6[2Z\xi(1-2\ln\kappa r)-1]}{Z^2\xi(1-2\ln\kappa r)^2}\ln\left[\mathrm{e}^{-K_0(\kappa r)}\,(\kappa b)^{\ln\kappa r}\right] \quad (13)$$

In the far region, $r \ge 1/\kappa$

$$w_3(r) = \frac{6}{Z^2\xi}(2Z\xi - 1)K_0(\kappa r) \quad (14)$$

Figure 6 illustrates these formulas and compares them to pairwise additivity, $w_3 = 3w_2$. The three-body attractive well is deeper than predicted by additivity, as perhaps would be expected if the condensed counterions were shared among all three rods.

2.3. Clusters of *N* Polyions. When we try to analyze the work of assembling a cluster of N parallel rods, $N>3$, we encounter a different kind of problem. For three rods with tranverse section forming the vertexes of an equilateral triangle, the spacing between each pair is the same, and the single distance r can be in the near, intermediate, or far region. But suppose we have a parallel cluster of seven rods in hexagonal array, that is, a central rod surrounded by a single shell of nearest neighbors. Even in such a simple case, there are three distinct separation distances between various pairs, and

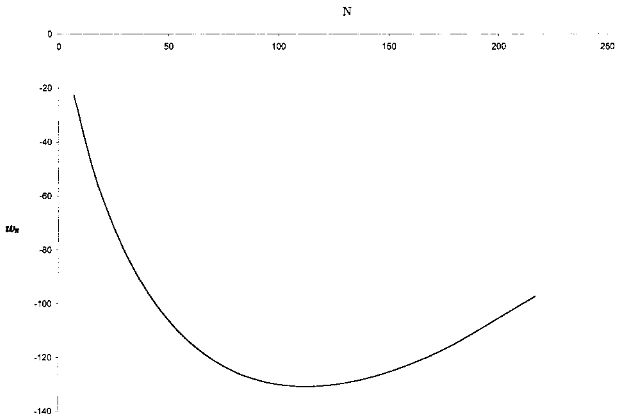

Figure 7. Dependence of the N-body reduced polymer potential on N (*i.e.*, the free energy required to assemble a cluster of N polyions as a function of N, expressed in units of Pk_BT). The N parallel rodlike polyions, each bearing P charges, are arranged on a hexagonal lattice with fixed lattice spacing $1/e\kappa = 20.4$ Å.

these distances may bear different relations to the Debye screening length $1/\kappa$. If the nearest neighbor separation is in the intermediate range, and the longer distances are in the far range, then attractive and repulsive interactions will coexist within the cluster (but recall the nonadditivity property, which makes this language imprecise). For a small cluster, the attractions among near neighbors might stabilize the cluster. But as the cluster grows, the number of repulsive interactions involving a given rod scales like the volume, while the number of attractions stays constant. There is then the possibility of a maximum cluster size, beyond which destabilizing repulsive forces dominate.

The mathematical analysis reflects this discussion in the way it handles the sum of the Bessel K_0 functions over polymer pairs ij in the interpolymer work component w_{pp} (the intrapolymer and transfer components are obtained from eqs 1 and 5, respectively, by replacing the initial factors 2 by N and the single variable r by the set of all pairwise rod–rod distances)

$$w_{pp}(\{r_{ij}\}) = 2\xi[1 - Z\theta(\{r_{ij}\})]^2 \sum_{i<j} K_0(r_{ij}) \tag{15}$$

where $\{r_{ij}\}$ is the entire set of rod–rod distance variables. We do the analysis for a regular hexagonal array. If the separation distance r_{ij} for a particular polyion pair is in the near ($r_{ij} < 1/e\kappa$) or intermediate ($1/e\kappa < r_{ij} < 1/\kappa$) region, we use an asymptotic expansion for the corresponding Bessel function $K_0(r_{ij})$, eq 6 or eq 7, respectively, generating logarithmic terms which affect the number of condensed counterions and give rise to attractive forces. If $r_{ij} > 1/\kappa$, the corresponding interaction $K_0(r_{ij})$ is treated as in the far region, $\kappa r_{ij} = O(1)$, and contributes to the repulsive free energy of the polymer cluster.

The calculation then proceeds in a manner similar to previous examples, and the work $w_N(\{r_{ij}\})$ of assembling a cluster of several shells of a hexagonal parallel array of N polyions can be obtained. In Figure 7, we see that the free energy for small clusters decreases as the cluster grows, but that a minimum is reached past which further growth is unstable. In this example, the lattice spacing is fixed at the value $1/e\kappa$, which gives the minimum of the 2-body potential $w_2(r)$. Figure 8 shows that w_N increases at fixed N if the spacing is chosen to be larger. One might have thought that the free energy would increase as well if the spacing is taken as less, since then near-region repulsions come into play. For $N = 7$, corresponding to a central rod surrounded by one complete shell, w_N indeed does increase. But Figure 8 indicates that a smaller spacing results in a lower free energy of assembly for larger clusters, because the increased number of intermediate-range attractive interactions dominates, and it appears that the most stable cluster in our model is a contact cluster. Possible implications are discussed in the next section.

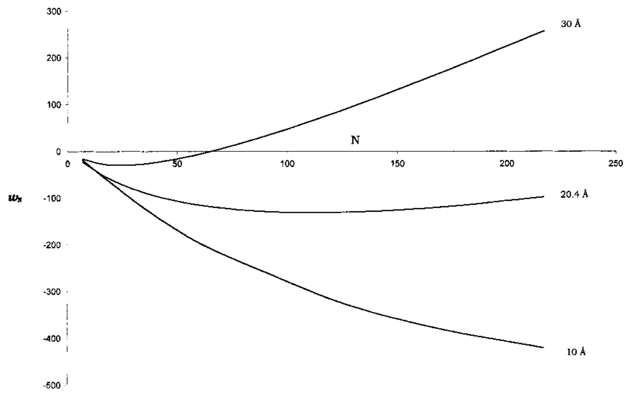

Figure 8. Dependence of cluster free energy w_N on lattice spacing. For example, the curve labeled 20.4 Å is identical to the curve in Figure 7 for lattice spacing 20.4 Å.

3. Discussion

When two identical rodlike polyions—or rodlike segments of polyions—are lined up in parallel, counterion condensation theory predicts a pair potential $w_2(r)$ that contains an attractive region at intermediate distances with a minimum at about a third of a Debye screening length (more precisely, at $r = 1/e\kappa$). The attractive well is deep and narrow; the energies in the graphics that illustrate this paper have all been divided by Pk_BT, where P is the number of charged groups on each polyion (the result is independent of P, since the energy scales like P). The attraction is mediated by the response of condensed counterions to mutual approach of the polyions. It is possible to formulate a classical analogy to a covalent bond.[10] The attraction does not require the counterions to be multivalent. In fact, the attraction is weaker for multivalent counterions than for univalent counterions (Figure 3).

The model and the ingredients of the theory of two interacting rods are the same as those that have always been present in counterion condensation theory,[15] which is frequently found to be in quantitative agreement with laboratory data, including the sharp onset of condensation at a threshold polymer charge density,[16] and which has never previously yielded qualitatively misleading results. The effectively infinite length of the polyion rod does not mean that the rod is literally (and unrealistically) infinitely long. The rod need only be long compared to the Debye screening length of the solution.[17] We can envisage the construction of a theory for the interaction of two rods of length the same order as the Debye length and possibly shorter,[17] which may be a better way to describe the interaction of roughly rodlike segments of flexible charged polymers.

Counterion condensation theory is not a Debye–Hückel theory. If it were, it could not have predicted an attractive interaction between like-charged polyions, and moreover, it would inevitably have produced a pairwise additive potential. Counterion condensation theory is applicable to systems well beyond the range of applicability of a Debye–Hückel theory. For example, it succeeds in describing important aspects of the binding of multivalent cations to DNA[18] and to synthetic polyelectrolytes.[19] It is true that a screened Debye–Hückel potential is an important ingredient of the theory (but there are other important ingredients). The approximation involved is mitigated both by the strong reduction of polymer charge by the layer of condensed counterions and by the relatively large distance scale (tens of angstroms) in the loose polymer clusters of interest in this paper. The use of a screened potential in the context of counterion condensation theory is directly supported by simulated data.[16,20]

The pair potential developed here is not entirely realistic. The actual potential must be a smooth function of distance. Ours features discontinuities of slope at the boundaries between near and intermediate regions and between intermediate and far regions. We therefore have errors near the discontinuities. These errors, however, cannot change our results qualitatively. For example, they cannot be spuriously responsible for the attractive interaction. In our original paper,[10] we showed the existence of an intermediate region where the potential was attractive. That initial analysis was not accompanied by joining conditions for the two abutting regions.

It might be of interest to repeat here from ref 14 that our *numerical* estimate $r = e^{-1}\kappa^{-1}$ for the position of the boundary between near and intermediate regions for any nonzero numerical value of κ is not an *asymptotic* formula implying that this boundary recedes to infinity like the Debye length κ^{-1}. In fact this estimate of the position of the boundary was derived from an asymptotic region of space that recedes to infinity like $\ln(1/\kappa)$ and hence does lie between the near and intermediate regions.[14] There can be no definite distance that asymptotically divides near and intermediate distances. A numerical estimate is necessary, however, for numerical applications of our formulas. We could have joined the asymptotic branches of the potential in ref 10 by a smooth interpolation polynomial, but because we were able to derive, albeit approximately, reasonable numerical formulas for the boundaries, we chose not to pursue this empirical path. The penalty is a potential with unrealistic cusps at the boundaries.

In this paper, we have extended the pair potential for two polyions to the work of assembling a cluster of N polyions. There are two new results. We have shown that pairwise additivity does not hold (Figure 6). More importantly, we have found a minimum in the free energy of a cluster as a function of the number N of polyions in the cluster (Figure 7). The only forces we have considered are electrostatic. The electrostatic potential is attractive at intermediate range and repulsive at far range. Electrostatic attraction to the surface causes the cluster to grow, but electrostatic repulsion from the volume eventually dominates and growth stops at a finite cluster size. Although the minima in Figures 7 and 8 give the impression of being broad, they actually are deep and narrow, when one considers that the free energy scale in these figures must be multiplied by the number of charges P per polymer to get the free energy of the actual cluster. This result is consistent with experimental observation on cluster formation in the presence of univalent counterions; the clusters have a well-defined radius of gyration independent of polymer[6] or salt[5] concentration.

We now discuss some big discrepancies between experimental data and the behavior of the model and try to learn something from them. We highlight two areas of disagreement: the size of the cluster and the polymer spacing within the cluster. From Figures 7 and 8 and the geometry of the hexagonal lattice, we find a theoretical cluster size (cross section radius) on the order of 100 Å. The apparent radius of gyration extracted from scattering data is an order of magnitude greater, about 1000 Å,[5,6] a value confirmed by direct visualization.[1]

As regards the polymer spacing, small-angle X-ray and neutron scattering data suggest that the average spacing between clustered polymer segments in solution may not be much less than the average spacing of single chains calculated from the nominal polymer solution concentration.[2] At the concentrations typical of the experiments, a spacing on the order of 100 Å is indicated. Our model for the cluster is a bundle of N parallel rodlike polyions. If the free energy of assembling an N cluster is considered as a function of the lattice spacing for a fixed number N of polyions, we find that the free energy decreases as the lattice spacing decreases (Figure 8). Although our model polyion becomes unrealistic in detail at close range, we interpret this result as indicating that the cluster becomes stable only when closely packed, which clearly disagrees with the experimental evidence.

Fluorescence transfer experiments on poly(L-lysine) solutions have indicated that enhancement of cluster formation is correlated with the movement of polymer chains to within 60 Å of each other.[9] However, it appears that each chain is the neighbor of only one or two others on this distance scale.[9] Polyelectrolyte clusters are apparently much more expanded, looser, and more open than predicted on the basis of our calculations for regular hexagonal arrays.

We are led to suggest that our theoretical results for $w_2(r)$, the pair potential for only two polyions, as illustrated in Figures 1–5, may be more pertinent for an understanding of polyelectrolyte clustering than the calculations for N polyions. A polyion cluster may consist of a diffuse domain of chains of polymer concentration not much greater than the average bulk solution concentration, stabilized by relatively few pairwise attractions at an appreciable distance. The theoretical pairwise attraction is weaker at higher ionic strengths (Figure 2), a trend that is consistent with observations on clustering.[1,2,21] If univalent counterions are replaced by divalent counterions, the theoretical attraction again becomes weaker (Figure 3), which again is consistent with the behavior of clusters in experiments.[11,22,23] The pairwise attraction does *not* weaken if the dielectric constant increases (Figure 5), in qualitative accord with dynamic light scattering results in nonaqueous solvents.[24]

Figure 4 shows that the attractive well in $w_2(r)$ is strongly enhanced by increases in the polymer charge density, even above the threshold value (where the net charge density of isolated polyions does not change). The reason is that the attractive force is caused by the condensed counterions, and there are more condensed counterions at higher charge densities. Experimentally observed characteristics of clustering, however, appear not to depend on charge density above the condensation threshold.[25] A cluster may depend for its existence on stabilizing attractive forces, but the size and number of clusters may depend more on limiting repulsive forces within it. A feature of our theory is that the origin of both the attractive and repulsive forces is electrostatic. Repulsive electrostatics predominate in overwhelmingly the largest part of the configuration space of far distances and nonparallel orientations.

An important issue arises from the exclusively repulsive forces found in currently available simulations with explicit counterions, if the counterions are univalent.[26−28] A remark not meant to be facetious is that the simulation— and not our theory—may be in trouble, since attractive interactions in the presence of univalent counterions are strongly suggested by laboratory data.[1−9,21−25,29] But we are also able to discuss the matter constructively. An attraction in a range of intermediate distances is, according to our theory, inextricably correlated with the Debye length of the solution, and a condition for simulating it is that the number of small ions and the size of the simulated region must be large enough to generate a well-defined Debye screening length. A first version of the analytical fluctuation theory of Ha and Liu[30] was formulated with unscreened Coulomb potentials (infinite Debye length), so, in our sense, it is a "near region" theory. The quantitative agreement of this theory with the simulated results of Gronbach−Jensen et al.[26] suggests that the simulation did not extend into the range of intermediate distances, which is the location of our attractive interaction. Our theory concurs with the simulations in finding a repulsive force in the near region. Incidentally, the simulation provided only the force between two rods, not the potential.

Another question that should be asked of simulations that employ the "primitive model" is whether the use of a uniform dielectric constant provides the condensed layer of counterions with a realistic structure. Although a sharply defined, visually discernible condensed layer containing the same number of counterions as predicted by condensation theory is indeed observed in simulated models equipped with a uniform dielectric,[31] the layer is relatively diffuse in comparison with the predicted radial distribution function.[14] A much tighter simulated layer is obtained if a more realistic dielectric function is employed in the near neighborhood of the polyion.[32] This point may be important, because if the starting entropy of the condensed layer is relatively high on an isolated polyion, then there is less entropy gain when two layers expand and merge to envelope a pair of polyions,[10] and whatever attraction may be caused by the entropy gain could be missed. Counterion condensation theory itself uses a uniform dielectric constant in the near region,[14] but it is almost automatic in providing a low-entropy condensed layer due to its realistic,[33] albeit not atomistic, modeling of the condensed counterions as renormalizing the bare polyion charge. Simulations with explicit counterions may not adequately approach the renormalization of polymer charge (which physically requires intimate association of the condensed counterions with the polymer chain) without a realistically small dielectric constant in a near neighborhood that, after all, is only a few angstroms wide. The use of explicit water molecules as well as explicit ions would of course solve this problem.[34]

A related comment is that simulations using a primitive model are in good agreement with univalent counterion distributions predicted by Poisson−Boltzmann theory. Poisson−Boltzmann theory, however, is incapable of reproducing the sharp jumps, breaks, bifurcations, and conformational transitions observed in the laboratory at the critical onset of counterion condensation.[16] In other words, Poisson−Boltzmann theory, primitive model simulations, and counterion condensation theory all concur in the prediction of a sharp onset of counterion condensation at a critical polymer charge density,[31] but the two former methods of analysis do not produce the intimate association of the condensed counterions with the polymer chain that must underlie the observed abrupt changes of binding behavior and polymer properties when the condensed layer forms.

The model presented in this paper does not allow for fluctuations of the condensed counterions along the axis of the polyion rod, so the attractive force discussed here is not caused by rod−rod correlations among these longitudinal fluctuations (a classical thermal analogue of van der Waals' forces, which reduces to something akin to ionic crystallization at low temperatures, and which also produces a potential that is not pairwise additive).[13,26,30,35−41] One way to distinguish the "covalent bond" of our theory from the "van der Waals' force" of fluctuation theories is that the latter has a much shorter range (as a quasi-contact force, it is sensitive to the size of the counterion[26]). We think it likely that fluctuation attraction is the mechanism underlying the collapse of DNA at low polymer concentration[42] as well as polyelectrolyte phase separation,[38,43] both induced by addition of multivalent cations. We offer our variant as an explanation for the loose open clusters that appear to form in the presence of univalent counterions.

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