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by

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Self-consistent-field theory investigation of behavior of hyaluronic acid chains in aqueous salt solution

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In this work we develop a field-theoretic methodology, which combines the concept of Gaussian equivalent representation for functional integrals with the continuous Gaussian thread model of flexible polymers for solving statistical-mechanical problems of polyelectrolyte solutions. We demonstrate the applicability of the method for systems of polyelectrolyte chains where the monomers interact via Yukawa-type pair potential. We develop the corresponding formulas and employ them to calculate structural and thermodynamic quantities of the polyelectrolyte system. As a specific example, the present work focuses on the aqueous solution of hyaluronic acid with added salts $NaCl$ and $CaCl_2$. Hyaluronic acid is a high molecular weight linear polysaccharide, which has a multitude of roles in biological tissues. We conclude that the effect of sodium chloride and calcium chloride on the osmotic properties of hyaluronic acid solutions can be accounted for by their contributions to the ionic strength. Nevertheless, the effects of coiling and self-association can be stimulated in solution by added salt.

Keywords: functional integral, partition functions, thermodynamic functions, polyelectrolyte solution.

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I. INTRODUCTION

Originally introduced by Edwards^{1,2} and by Helfand and Tagami³, mean-field theories for polymers, also commonly referred to as self-consistent-field theory, have been proven useful for estimating structure and thermodynamic properties of a wide variety of polymer systems^{4,5}. Their applications to polyelectrolyte and ionic systems have, however, only been rare^{4,6,7}. This is due to the fact that the mean-field approximation is only accurate for highly concentrated polyelectrolyte solutions, where local field fluctuations are averaged out, due to effective screening of the electrostatic interactions surrounding the monomers⁴. The mean-field approximation provides inaccurate or even qualitatively incorrect results for polyelectrolyte solutions in the regime of low to moderate concentrations⁴. However, these concentration regimes are highly relevant to biological applications. In such cases more sophisticated techniques beyond the mean-field level of approximation are required⁸.

In our previous works^{8,11,12,14,15} we developed the such technique beyond the mean-field level of approximation. Our approach is based on the method of the Gaussian equivalent representation (GER) for the partition functions in the form of functional integrals^{9,10,13}.

The goal of the present work to investigate a possibility of the appropriate description of a polyelectrolyte solution with added univalent and divalent ions in the semidilute concentration region within our model of polyelectrolyte solution^{14,15}, and to obtain the information about the influence of counterions on the thermodynamic functions and configuration features of the polyelectrolyte chains in solution. As a specific example the present work focuses on the aqueous solution of hyaluronic acid (HA) with added salts $NaCl$ and $CaCl_2$. The experimental data for osmotic pressure, which were obtained in Ref.¹⁶, are convenient and useful object for our investigation. In the work by F.Horkay et al.¹⁶ an empirical equation of state was obtained for the osmotic pressure in the semidilute concentration region, in terms of two variables, the polymer concentration and the ionic strength of the added salt. The conclusion of authors is that over the physiological ion concentration range, the effect of the sodium chloride and calcium chloride on the osmotic properties of HA solutions is fully accounted for by their contributions to the ionic strength.

The abundance of HA (also called Hyaluronan) and its multiple roles in biological tissues is unique among biopolymers^{16,18–20}. HA is a component of connective tissue whose function is to cushion and lubricate. HA, or commercial preparations containing HA, are in use,

or being studied to be used, to prevent, treat or aid in the surgical repair for many the types of problems people with connective tissue disorders tend to have such as: fractures, hernias, glaucoma, detached retinas, osteoarthritis. HA is a primary constituent of the extracellular matrix and also participates in a variety of cell-to-cell interactions. Among its many functions, HA plays a critical role in cartilage, where the collagen network enmeshes large aggrecan-HA complexes that provide resistance to compressive load¹⁷.

The repeating unit in this linear polymer is an AB disaccharide: [*D*-glucuronic acid (*A*)(β -(1 \rightarrow 3)) - 2 - (acetylamino) - 2 - deoxy - *D* - glucose (*B*)(β - (*l* \rightarrow 4))]_{*N*}, so that the polymer contains 2*N* monosaccharides and *N* ionizable groups located on the (*A*) glucopyranose rings of the *ABAB* ... structure. The distance of separation of these groups will depend on the angles at the glycoside bonds^{18-20,22}.

HA is joined in class of polyacids which, even when fully neutralized in water (pH is 7), are completely ionized, or nearly so^{16,18}. According to the criterion of Manning^{23,24}, complete ionization occurs for the infinite line charge model of polyelectrolytes when the charge density parameter ξ is less than unity, where ξ is defined by $\xi = \lambda_B/b$, λ_B - Bjerrum length and *b* - the length per unit (electron) charge. The *b* must be greater than about 7Å to meet this criterion. As *b* decreases below this distance, the Manning theory predicts increasing counterion condensation in the near vicinity of the polyion, which partly shields the polyion charge. HA is one of the few available polyions which meet the criterion for complete ionization, because conformational calculations suggest an average ionizable site separation of about $\sim 10\text{\AA}$ ^{18,21,22}, depending on the energy parameters chosen to represent the molecule. Available experimental evidence supports the assumption, based on the Manning criterion, that hyaluronate is completely ionized in solution^{16,18}.

HA in neutral aqueous solution, at or near a physiological concentration of NaCl, generally behaves as a typical semi-flexible polymer molecule¹⁹. Short chains are somewhat extended, while longer chains show evident coiling. The molecular domain of a high molecular weight HA chain occupies a sphere, as the time average of all accessible conformations. Because the molecular domains are quite large (but not impenetrable), HA chains interfere with each other at even low concentrations¹⁹. However, the influence of counterions behavior on the configuration properties of hyaluronic acid chain is not yet fully clear.

In the work¹⁵ we studied the thermodynamic response of aqueous chondroitin sulfate solutions to changes in the monomer and added salt concentrations, using a field-theoretic

approach beyond the mean field level of approximation. We compared our calculation results to data from experiments as well as molecular dynamics calculations, and demonstrated that our method provides reliable information for the osmotic pressure and entropy. By adjusting the osmotic pressure to experimental data and analyzing the resulting effective excluded volume parameter in various concentration regimes, we have investigated the local electrostatic and solvent effects, influencing the condensation behavior of the counterions onto the chondroitin sulfate chains.

HA is similar in chemical structure to chondroitin, but is typically of very high molecular weight, ranging from 10^5 to more than $10^6 Da$, equivalent to $\sim 250-2500$ disaccharide repeat units, and is not sulfated. Though molecule of HA is semi-flexible chain, we'll consider HA chains within Gaussian thread model, inasmuch as HA chains are very long.

Our paper is organized in the following way. In the Section 2 we review the basic derivation of the field theory for flexible polymer chains, followed by the derivation of our GER theory. We would like to stress, that a form of the potential of mean force, which contains the Debye function^{2,4}, is a new result of our present work in compare to our previous works^{14,15}. The derivation for the potential of mean force is took out in Appendix A. Then, we demonstrate the applicability of the method on systems of polyelectrolyte chains, where the monomers interact via Yukawa-type pair potential, and develop the corresponding formulas, employed to calculate the structural and thermodynamic quantities considered in this work. In Section 3 we present and discuss the results of our calculations on the example of aqueous HA solutions at various monomer and salts concentrations by comparing them to the osmotic pressure measurements of Horkay et al.¹⁶.

II. METHOD

Let us consider a solution composed of n polymer chains. The solvent is taken into account implicitly. The polymer chain is described by the continuous Gaussian thread model^{2,4}. The grand partition function can be expressed as^{4,14}

$$\Xi(z, \beta, V) = \sum_{n=0}^{\infty} \frac{z^n}{n!} \prod_{i=1}^n \int \delta \vec{r}_i e^{\beta W[\vec{r}_i, \Phi]}, \quad (1)$$

where

$$W[\vec{r}_i; \Phi] = -\frac{3}{2Nb^2\beta} \int_0^1 ds \left(\frac{d\vec{r}_i(s)}{ds} \right)^2 - \frac{N^2}{2} \int_0^1 ds_1 \int_0^1 ds_2 \sum_{i \neq j}^n \Phi(\vec{r}_i(s_1) - \vec{r}_j(s_2)), \quad (2)$$

N -polymerization index, b-statistical average length of segment,

$$\beta = \frac{1}{k_B T}, \quad z = \left(\frac{2\pi m}{\beta h^2} \right)^{3/2} e^{\beta \mu}$$

-activity. The first term in (2) is the configuration potential energy. It represents the harmonic stretching energy of the chain. The second term describes the solvent-mediated interactions among the segments.

To derive the basic field-theoretical representation of the grand partition function, we introduce the segment density operator of the polymer system:

$$\rho(\vec{r}) = \sqrt{\beta N} \int_0^1 ds \sum_{i=1}^n \delta(\vec{r} - \vec{r}_i(s)). \quad (3)$$

We can recast the integrand exponent in the grand partition function in the form of functional integral^{4,14}, which results in

$$\begin{aligned} & \exp \left[-\frac{\beta N^2}{2} \int_0^1 ds_1 \int_0^1 ds_2 \sum_{i \neq j}^n \Phi(\vec{r}_i(s_1) - \vec{r}_j(s_2)) \right] = \\ & = \exp \left[-\frac{1}{2}(\rho \Phi \rho) + \frac{\beta N n \Phi(0)}{2} \right] = \int \frac{D\phi}{\sqrt{\det \Phi}} e^{-\frac{1}{2}(\phi \Phi^{-1} \phi) + i(\rho \phi) + \frac{\beta N n \Phi(0)}{2}} = \\ & = \int d\mu_\Phi[\phi] : \exp \left[i\sqrt{\beta N} \sum_{i=1}^n \int_0^1 ds \phi(\vec{r}_i(s)) \right] :_\Phi, \end{aligned} \quad (4)$$

where we introduced the concept of the *normal product* according to the given Gaussian measure $d\mu_\Phi[\phi]$:

$$\begin{aligned} & : e^{i\sqrt{\beta N} \int_0^1 ds \phi(\vec{r}(s))} :_\Phi = e^{i\sqrt{\beta N} \int_0^1 ds \phi(\vec{r}(s))} e^{\frac{\beta}{2} N \Phi(0)}, \\ & : \phi(\vec{r}_i(s)) \phi(\vec{r}_j(s')) :_\Phi = \phi(\vec{r}_i(s)) \phi(\vec{r}_j(s')) - \beta \Phi(\vec{r}_i(s) - \vec{r}_j(s')), \\ & \int d\mu_\Phi : e^{i\sqrt{\beta N} \int_0^1 ds \phi(\vec{r}(s))} :_\Phi = 1. \end{aligned}$$

This provides the correct account of the potential in zero, and leads to summation of so called tadpole diagrams^{9,10}.

Using the formula (4) one can represent the grand partition function in the form

$$\Xi(z, \beta, V) = \int d\mu_\Phi[\phi] \exp \left[z \int \delta \vec{r} e^{-\frac{3}{2N b^2} \int_0^1 ds \left(\frac{d\vec{r}(s)}{ds} \right)^2} : e^{i\sqrt{\beta N} \int_0^1 ds \phi(\vec{r}(s))} :_\Phi \right], \quad (5)$$

where

$$d\mu_{\Phi}[\phi] = \frac{\delta\phi}{\sqrt{\det \Phi}} \exp \left[-\frac{1}{2}(\phi\Phi^{-1}\phi) \right],$$

-Gaussian measure,

$$(\phi\Phi^{-1}\phi) = \int_V d^3r \int_V d^3r' \phi(\vec{r})\Phi^{-1}(\vec{r}, \vec{r}')\phi(\vec{r}'),$$

$$\int_V d^3r \Phi(\vec{r}', \vec{r})\Phi^{-1}(\vec{r}, \vec{r}'') = \delta^3(\vec{r}' - \vec{r}'').$$

A positivity of the Fourier-transformation for the potential Φ

$$\tilde{\Phi}(\vec{p}) = \int d^3r \Phi(\vec{r})e^{i(\vec{p} \cdot \vec{r})} > 0.$$

is sufficient condition for the existence of the functional integral (5).

The ordinary strategy for approximate calculation of the functional integral (5) is a mean field approximation. However, there is multitude of cases where the mean field approach has been found to provide either inaccurate or even qualitatively wrong results^{4,8}.

Our aim is to obtain the equivalent representation of the functional integral, which is suitable for any external parameters.

Let us perform the displacement of the functional variable $\phi \rightarrow \phi + \frac{ic}{\sqrt{\beta}}$ and go over to a new Gaussian measure $d\mu_D[\phi]$, where c and $D(x,y)$ are functions to be determined. The Grand partition function takes the next form:

$$\Xi(z, \beta, V) = \sqrt{\frac{\det D}{\det \Phi}} \int d\mu_D[\phi] e^{W_D}, \quad (6)$$

$$W_D = -\frac{1}{2} : (\phi[\Phi^{-1} - D^{-1}]\phi) :_D - \frac{1}{2}(D[\Phi^{-1} - D^{-1}]) - \frac{i}{\sqrt{\beta}}(c\Phi^{-1}\phi) +$$

$$+\frac{1}{2\beta}(c\Phi^{-1}c) + zAe^{-Nc} \int d\sigma[\vec{r}] : [1 + i\sqrt{\beta}N \int_0^1 ds \phi(\vec{r}(s)) -$$

$$-\frac{\beta N^2}{2} \int_0^1 ds_1 \int_0^1 ds_2 \phi(\vec{r}(s_1))\phi(\vec{r}(s_2))] :_D + W_2[\phi]. \quad (7)$$

Functional $W_2[\phi]$ is written in the form, which does not have linear and quadratic terms over the integration variable $\phi(x)$. Our basic idea is that the main contribution to the functional

integral is concentrated in the new quadratic Gaussian measure. It means, that the linear and quadratic terms over the integration variable $\phi(x)$ should be absent in the integrant exponent. Thus, we obtain two equations.

$$-\frac{i}{\sqrt{\beta}}(c\Phi^{-1}\phi) + z_1 i \sqrt{\beta} N e^{-Nc} \int_0^1 ds \int d\sigma[\vec{r}] \phi(r(\vec{s})) = 0 \quad (8)$$

$$-\frac{1}{2}(\phi[\Phi^{-1} - D^{-1}]\phi) - \frac{z_1 N^2 \beta e^{-Nc}}{2} \int_0^1 ds_1 \int_0^1 ds_2 \int d\sigma[\vec{r}] \phi(\vec{r}(s_1)) \phi(\vec{r}(s_2)) = 0, \quad (9)$$

where

$$d\sigma[\vec{r}] = \frac{\delta\vec{r} \exp\left[-\frac{3}{2Nb^2} \int_0^1 ds \left(\frac{d\vec{r}(s)}{ds}\right)^2\right]}{\int \delta\vec{r} \exp\left[-\frac{3}{2Nb^2} \int_0^1 ds \left(\frac{d\vec{r}(s)}{ds}\right)^2\right]},$$

$$z_1 = \frac{\exp\left[\beta\mu + \frac{\beta}{2}N[\Phi(0) - D(0)]\right]}{h^3 \left(\frac{\beta}{2\pi m}\right)^{3/2}} \int \delta\vec{r} \exp\left[-\frac{3}{2Nb^2} \int_0^1 ds \left(\frac{d\vec{r}(s)}{ds}\right)^2\right],$$

$$W_2 = z A e^{-Nc} \int d\sigma[\vec{r}] : e_2^{i\sqrt{\beta}N \int_0^1 ds \phi(\vec{r}(s))} :_D,$$

$$e_2^x = e^x - 1 - x - \frac{x^2}{2}.$$

The equations (8,9) connect the effective potential $D(r)$ with temperature, activity, polymerization index and statistical average length of segment. These are the equations of a self-consistency in our method.

The Eq.(8) can be written in the form

$$c = z\beta N A \tilde{\Phi}(0) e^{-Nc}. \quad (10)$$

The derivation for the solution of equation (9) is took out in Appendix A. The Fourier transformation for the function $D(r)$ can be expressed as

$$\tilde{D}(q^2) = \frac{\tilde{\Phi}(q^2)}{1 + cG(q^2) \frac{\tilde{\Phi}(q^2)}{\tilde{\Phi}(0)}}, \quad (11)$$

where

$$G(q^2) = \frac{12}{q^2 b^2} + \frac{72}{N b^4 q^4} (e^{-\frac{N b^2 q^2}{6}} - 1) = N f\left(\frac{N q^2 b^2}{6}\right) \quad (12)$$

- Debye's structure-factor, and

$$f(x) = \frac{2}{x^2} (e^{-x} - 1 + x)$$

- Debye function².

This is a new result in compare to our previous works^{14,15}. In the works^{14,15} we used approximation $G(q^2) \approx N$, because $G(0) = N$ and for not large N the Debye's factor can be written in form

$$G(q^2) \approx \frac{12}{q^2 b^2} + \frac{72}{N q^4 b^4} \left(1 - \frac{N b^2 q^2}{6} + \frac{N^2 b^4 q^4}{72} - 1\right) = N \quad (13)$$

However, for large N this approximation is not sufficiently accurate. The new form of the effective potential (11) leads to the new performance capabilities for description of polymer chains.

The grand partition function can be written in the form

$$\Xi(z, \beta, V) = e^{\beta P V} = e^{-\beta \Omega_{GER}^{(0)}} \int d\mu_D[\phi] e^{W_2[\phi]}. \quad (14)$$

The linear and quadratic terms over the integration variable $\phi(x)$ are absent in $W_2[\phi]$. Therefore, we can choose $W_2 \approx 0$. We estimated this approximation⁹. The contribution of the following terms is smaller than 10 percents in the wide region of external parameters.

The average polymer chains density can be expressed as

$$\langle \rho \rangle = \frac{z_1}{V} \frac{1}{\Omega_{GER}^0} \left(\frac{\partial \Omega_{GER}^0}{\partial z_1} \right)_{\beta, V}.$$

Using the equation (10) we obtain

$$\rho_m = \frac{c}{\beta \tilde{\Phi}(0)} - \frac{N c^2}{2\pi(1 + cN)} \int_0^\infty \frac{u^2(q) q^2 dq}{[1 + cu(q)]^2}, \quad (15)$$

where $u(q) = G(q^2) \tilde{\Phi}(q) / \tilde{\Phi}(0)$, and $\rho_m = \langle \rho \rangle N$ - average monomer densities.

The analytic expression for osmotic pressure has the following form:

$$\frac{P}{k_B T} = \frac{2c + N c^2}{2N \tilde{\Phi}(0)} + \frac{1}{12\pi^2} \int_0^\infty dq \frac{du(q)}{dq} \frac{c^2 q^3 u(q)}{[1 + cu(q)]^2}, \quad (16)$$

and the pair distribution function for monomers can be written as

$$g(r) = \exp[-\beta D(r)],$$

where

$$D(r) = \int \frac{d^3q}{(2\pi)^3} \widetilde{D}(p) e^{i\vec{p}\vec{r}} \quad (17)$$

is potential of mean force¹³ within our approximation ($W_2 = 0$).

The equations (15-16) determine the equation of state for polymer solution. The osmotic pressure as the function of the density, temperature, polymerization index and the parameters of the pair potential of interaction among monomers can be explored via equations (15-16). There are different approaches for the development of an equation of state for polymer systems described in literature^{14,25}, however, in our work the equation of state is derived by GER from basic conception of statistical mechanics - Gibbs distribution for grand canonical ensemble.

III. MODEL OF POLYELECTROLYTE SOLUTION

In this section we consider a model of polyelectrolyte solution to investigate the aqueous solution of HA with added salts. A polymer chain is described by the continuous Gaussian thread model.

The monomers interact via a Yukawa-type of pair potential¹⁴

$$\Phi(r; \kappa_D) = z_m^2 A(\kappa_D, a) \frac{\lambda_B}{\beta} \frac{e^{-\kappa_D r}}{r}, \quad (18)$$

where

$$A(\kappa_D, a) = \left(\frac{\exp(\kappa_D a/2)}{1 + \kappa_D a/2} \right)^2 \quad (19)$$

is factor, which depends from the size and form of monomer. The parameter a is characteristic size for excluded volume of monomer. The solvent is taken into account implicitly via Bjerrum length $\lambda_B = z_m^2 e^2 \beta / (4\pi \epsilon_0 \epsilon)$ and Debye screening parameter κ_D . Hydrophobic and attractive van der Waals non-bonded interactions are ignored because electrostatic effects have been shown to dominate in determining HA conformational and thermodynamic properties at the ionic strengths considered ($\sim 1M$)²².

The Fourier transformation for the potential of mean-force (11) has a form

$$\beta\widetilde{D}(q^2) = \frac{4\pi z_m^2 A(\kappa, a)\lambda_B}{q^2 + \kappa_D^2[1 + cG(q^2)]}, \quad (20)$$

where $G(q^2)$ is defined by equation (12).

The Debye screening parameter, taking into account an electroneutrality, it is possible to write down in the form

$$\kappa_D = \sqrt{4\pi\lambda_B(2z_m^2\rho_m + 2z_{s1}^2\rho_{s1} + 2z_{s2}^2\rho_{s2})} \quad (\text{\AA}^{-1}). \quad (21)$$

Here the following designations are used:

$$z_{s1} = z_{Na} = -z_{Cl} = 1, \quad z_{s2} = z_{Ca} = -2z_{Cl} = 2,$$

$\rho_m = N_A C_m 10^{-27} \text{ (\AA}^{-3}\text{)}$ or $\rho_m = \phi 10^3 / (v_{HA} M) \simeq 6\phi / (\pi a^3) \text{ (\AA}^{-3}\text{)}$ - number density for monomers and $\rho_s = N_A C_s 10^{-27} \text{ (\AA}^{-3}\text{)}$ - number density of counterions, C_m is concentration of monomers (monomol/l), ϕ is volume fraction of polyelectrolyte, $v_{HA} = 0.59 \text{ (cm}^3\text{/g)}$ - specific volume of HA²¹, C_s is concentration (M) of added salt, and N_A is Avogadro constant, $\lambda_B \simeq 7 \text{ \AA}$ (water at 298K). We choose the parameter a for HA as $a = 10 \text{ \AA}^{21,22}$. The parameter $c = c(\beta, \rho_m, \rho_s, \kappa_D, b)$ is calculated via the equation (15), where

$$u(q^2) = \frac{G(q^2)}{q^2 + \kappa_D^2}. \quad (22)$$

The osmotic pressure is defined by equation (16). The potential of mean force can be calculated as inverse Fourier-transformation:

$$D(r) = \frac{1}{2\pi^2} \int_0^\infty \widetilde{D}(q^2) \frac{\sin(qr)}{r} q dq, \quad (23)$$

and pair distribution function for monomers is defined as $g(r) = \exp[-\beta D(r)]$.

Our unique free parameter is b ($b \simeq a$), which defines the harmonic stretching energy (2) and the linear charge density of polymer chain. We use the parameter b to approximate the experimental measurements of osmotic pressure by equation (16).

IV. DISCUSSION

Here we shall investigate the thermodynamic and structure properties of aqueous HA solution with added salts, using the experimental measurements of osmotic pressure from

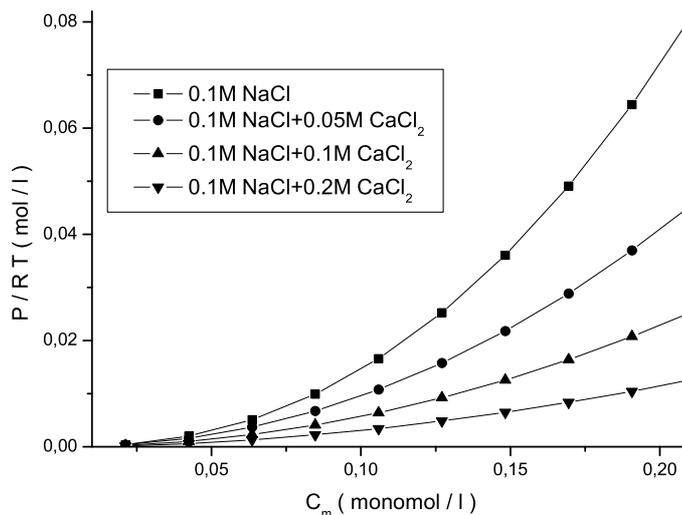


FIG. 1. Osmotic pressure for aqueous HA solution as a function of monomer concentration at various concentrations of calcium chloride ($R=8.31 \text{ J}/(\text{K mol})$, $T=298\text{K}$). Lines - approximation of experimental data by equation $P = A\phi^n$ from Ref.¹⁶, symbols - our results, Eq.16.

work by Horkay et al.¹⁶, where HA with weight-average molecular weight $M_W = 1.2 \times 10^6 \text{ Da}$ was utilized. Polymerization index is defined as $N = M_W/m = 3 \times 10^3$, where $m = 400 \text{ Da}$ is mass of monomer unit. To obtain numerical experimental data we used A and n from Table 1 of Ref.¹⁶.

In Fig.1 we show the osmotic pressure as a function of monomer concentration at different concentrations of CaCl_2 , obtained by adjusting the GER expressions in Eqs.(15)-(16) onto the experimental through varying the parameter b . As we can see the Eq.(16) correctly describes the experimental data. Corresponding graphs for parameter $\xi = \lambda_B/b$ are presented in Fig.2. Parameter ξ is less than unity and slightly depends from CaCl_2 concentration, therefore we are inclined to believe that counterions condensation is not observed. In our opinion, the deviations of b from a are caused by the nonspherical form of HA monomers. These may be related too to changes in the intrinsic properties of the polyelectrolyte molecule, such as rigidity, due monovalent-divalent ion exchange¹⁶.

Now we can calculate the potential of mean-force, pair distribution function and entropy using the obtained values of parameter b . In Fig.3 we show the potential of mean-force as a function r/a for different polymerization indexes N . Function $D(r)$ describes both intra-

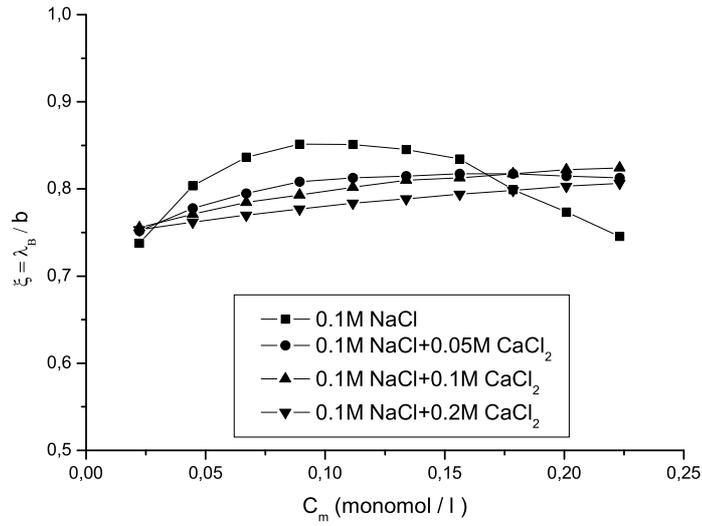


FIG. 2. Parameter $\xi = \lambda_B/b$ as a function of monomer concentration at various concentrations of calcium chloride.

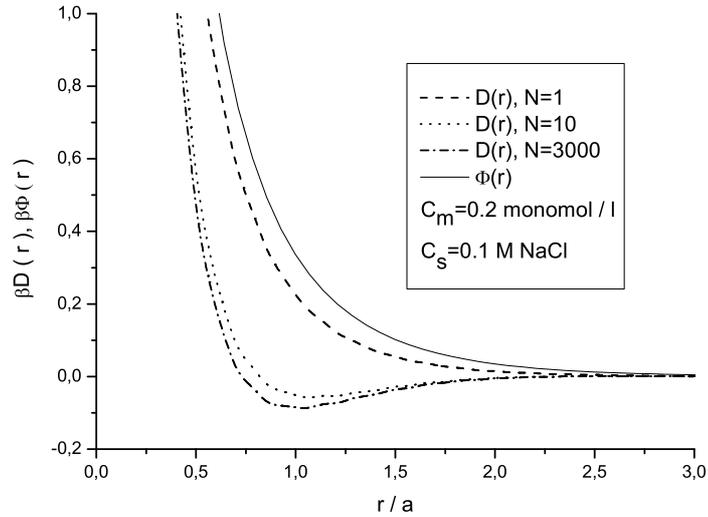


FIG. 3. Potential of mean-force $D(r)$ as a function r/a .

molecular and inter-molecular interactions among monomers in solution and defines the behavior of pair distribution functions.

The monomer-monomer pair distribution functions $g(r) = \exp[-\beta D(r)]$ for various HA and $CaCl_2$ concentrations are presented in the Fig.4-6. As we can see, the increases of the

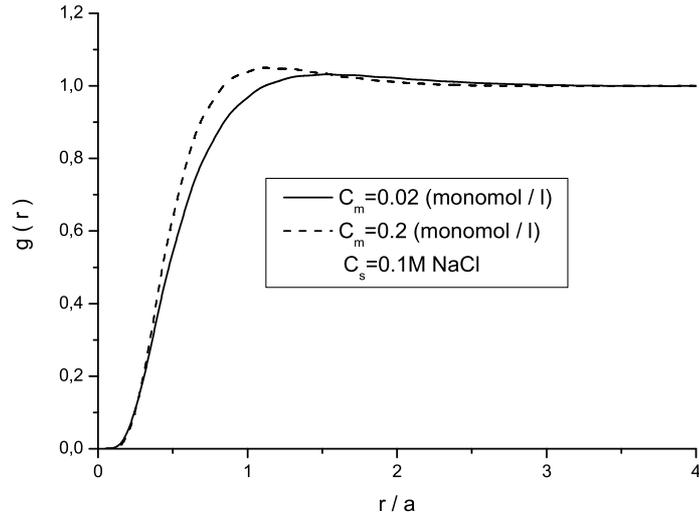


FIG. 4. The pair distribution functions of monomers without calcium chloride.

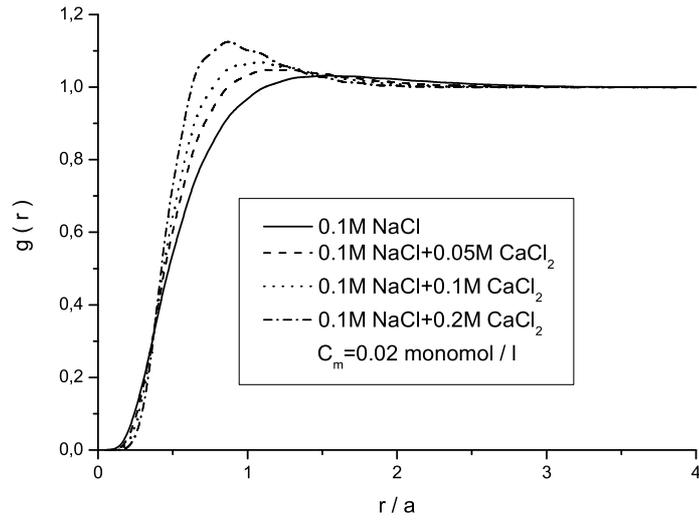


FIG. 5. Pair distribution functions of monomers at HA concentration $C_m = 0.02$ monomol/l and various concentration of calcium chloride.

concentration of both HA and salt displace the maximum of the pair distribution function to the left. The reduction of distance to the first maximum can be explained by the increase of a screening parameter (21) and by the decrease of the repulsion among the segments. At that, as was noted in Ref.¹⁹, HA chains interfere with each other at even low concentrations

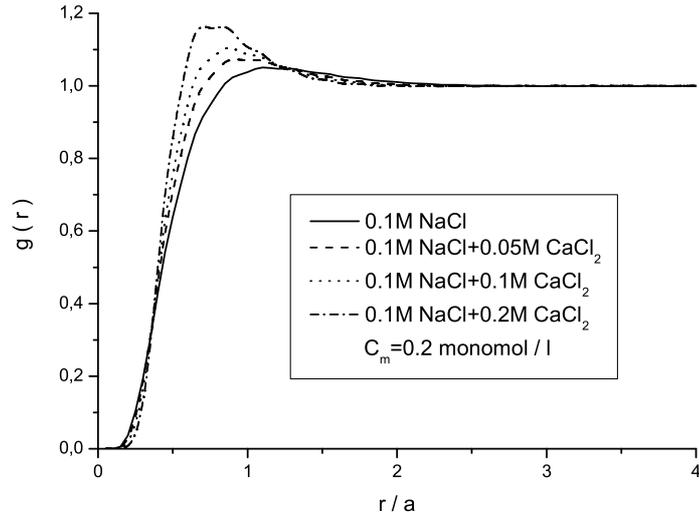


FIG. 6. Pair distribution functions of monomers at HA concentration $C_m = 0.2$ monomol/l and various concentration of calcium chloride.

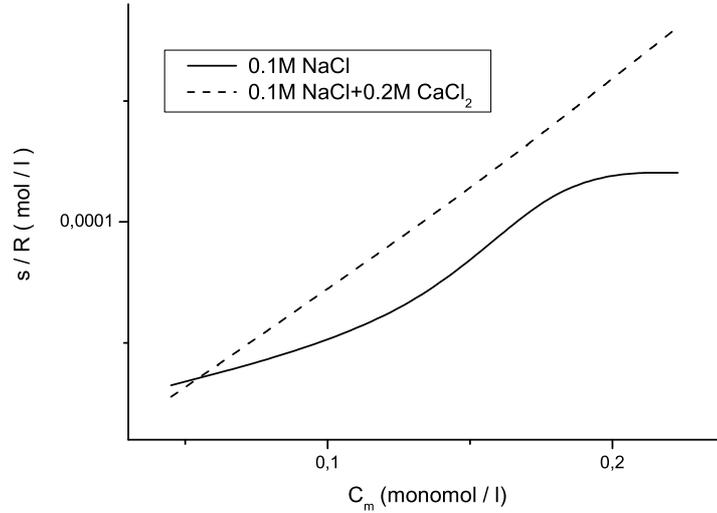


FIG. 7. Entropy as a function of monomer concentration ($T=298K$).

and their long chains show evident coiling. This is corroborated by increase of entropy with the adds of salt. The graphs for entropy density $s = \left(\frac{\partial P}{\partial T}\right)_\mu$ at $T = 298K$ are shown in the Fig.7. As we can see, the adds of $CaCl_2$ increase the entropy of the system.

At the small concentration of salt (0.1M NaCl) we can see the significant changes in

behaviors of parameter b and entropy at polyelectrolyte concentration 0.1-0.2 monomol/l (Fig.2 and Fig.7). In our opinion, it reflects the some reconstruction of the polymer chains in solution.

V. CONCLUSION

In this work we have presented the field-theoretic model of polyelectrolyte solution. The essential formulas for calculations of thermodynamic and structural functions have been derived. As example we have considered the aqueous solution of HA with added $NaCl$ and $CaCl_2$. Our conclusion is accordant to conclusion of the work¹⁶, that the changes in thermodynamic and structural functions with increasing ionic strength is due essentially to the change in electrostatic screening, and the valence of the counterion exerts no specific effect on the HA molecule except through its contribution to the ionic strength.

Indeed, the full insight into the structure of polyelectrolyte system is possible via explicit multiscale examination of all presented interactions in solution^{4,26-28}. However, the simple analytic formulas and correlations, which derived from the basic principles of statistical mechanics, can be very useful for the successful investigation too. We have tried to demonstrate by comparing our theoretical approach to experimental measurements that it provides reliable results of the thermodynamic osmotic pressure and pair distribution function for monomers.

Appendix A: Solving the equation (9)

By using Fourier transformation we can rewrite the second term in the Eq.(9) as

$$\begin{aligned}
& \int_0^1 ds_1 \int_0^1 ds_2 \int d\sigma[\vec{r}] \phi(\vec{r}(s_1)) \phi(\vec{r}(s_2)) = \\
& = \int_0^1 ds_1 \int_0^1 ds_2 \int \frac{d^3q}{(2\pi)^3} \int \frac{d^3p}{(2\pi)^3} \tilde{\phi}(q) \tilde{\phi}(p) \int d\sigma[\vec{r}] e^{iqr(s_1)+ipr(s_2)} = \\
& = \int_0^1 ds_1 \int_0^1 ds_2 \int \frac{d^3q}{(2\pi)^3} \int \frac{d^3p}{(2\pi)^3} \tilde{\phi}(q) \tilde{\phi}(p) \times \\
& \times \int \frac{\delta\vec{r}}{C_r} \exp \left[-\frac{3}{2Nb^2} \int_0^1 d\tau \left(\frac{d\vec{r}(\tau)}{d\tau} \right)^2 + i\vec{q}\vec{r}(s_1) + i\vec{p}\vec{r}(s_2) \right], \tag{A1}
\end{aligned}$$

where

$$C_r = \int \delta \vec{r} \exp \left[-\frac{3}{2Nb^2} \int_0^1 d\tau \left(\frac{d\vec{r}(\tau)}{d\tau} \right)^2 \right].$$

Now let us perform a following displacement

$$\vec{r}(s) \rightarrow \vec{r}(s) + \vec{y} s + \vec{x} (1 - s), \quad \vec{r}(0) = \vec{r}(1) = 0.$$

The chain is beginning in the point \vec{x} and ending in the point \vec{y} . The integrations over \vec{x} and \vec{y} should be executed.

$$\begin{aligned} & \int_0^1 ds_1 \int_0^1 ds_2 \int d\sigma[\vec{r}] \phi(\vec{r}(s_1)) \phi(\vec{r}(s_2)) = \\ & = \int_0^1 ds_1 \int_0^1 ds_2 \int \frac{d^3 q}{(2\pi)^3} \int \frac{d^3 p}{(2\pi)^3} \tilde{\phi}(q) \tilde{\phi}(p) \int d^3 x \int d^3 y \times \\ & \times \int \frac{\delta \vec{r}}{C_r} e^{-\frac{3}{2Nb^2} \int_0^1 d\tau \left(\frac{d\vec{r}(\tau)}{d\tau} + \vec{y} - \vec{x} \right)^2 + i\vec{q}\vec{r}_{s_1} + i\vec{p}\vec{r}_{s_2} + i(\vec{y} - \vec{x})(\vec{q}s_1 + \vec{p}s_2) + i\vec{x}(\vec{q} + \vec{p})}. \end{aligned}$$

Now the following displacement should be done:

$$\vec{y} \rightarrow \vec{y} + \vec{x}.$$

Further we had performed the integrations over \vec{x} and \vec{p} :

$$\begin{aligned} & \int_0^1 ds_1 \int_0^1 ds_2 \int d\sigma[\vec{r}] \phi(\vec{r}(s_1)) \phi(\vec{r}(s_2)) = \\ & = \int_0^1 ds_1 \int_0^1 ds_2 \int \frac{d^3 q}{(2\pi)^3} \tilde{\phi}^2(\vec{q}) \int d^3 y e^{-\frac{3}{2Nb^2} \vec{y}^2 + i\vec{y}\vec{q}(s_1 + s_2)} \times \\ & \times \int \frac{\delta \vec{r}}{C_r} \exp \left[-\frac{3}{2Nb^2} \int_0^1 d\tau \left(\frac{d\vec{r}(\tau)}{d\tau} \right)^2 + i\vec{q}(\vec{r}(s_1) + \vec{r}(s_2)) \right], \end{aligned} \quad (\text{A2})$$

here

$$\int d^3 y e^{-\frac{3}{2Nb^2} \vec{y}^2 + i\vec{y}\vec{q}(s_1 + s_2)} = \frac{2}{9} \sqrt{6\pi} N^{\frac{3}{2}} b^3 e^{-\frac{1}{6} q^2 (s_1 + s_2)^2 Nb^2}. \quad (\text{A3})$$

The integrations over all \vec{x} and \vec{y} should be executed in the denominator (C_r) too.

Now, let us consider the functional integral

$$\int \frac{\delta \vec{r}}{C_r} \exp \left[-\frac{3}{2Nb^2} \int_0^1 d\tau \left(\frac{d\vec{r}(\tau)}{d\tau} \right)^2 + i\vec{q}(\vec{r}(s_1) + \vec{r}(s_2)) \right] =$$

$$= \int \frac{\delta \vec{r}}{C_r} \exp \left[-\frac{1}{2} (\vec{r} A^{-1} \vec{r}) + (\vec{J} \vec{r}) \right] = \frac{e^{\frac{1}{2}(\vec{J} A \vec{J})}}{\frac{2}{9} \sqrt{6\pi} N^{\frac{3}{2}} b^3}, \quad (\text{A4})$$

where $\vec{r}(0) = \vec{r}(1) = 0$,

$$\begin{aligned} (\vec{r} A^{-1} \vec{r}) &= \int_0^1 d\tau_1 \int_0^1 d\tau_2 \vec{r}(\tau_1) A^{-1}(\tau_1, \tau_2) \vec{r}(\tau_2), \\ (\vec{J} \vec{r}) &= \int_0^1 d\tau \vec{J}(\tau) \vec{r}(\tau), \\ A^{-1}(\tau_1, \tau_2) &= -\frac{3}{2Nb^2} \frac{d^2}{d\tau_1^2} \delta(\tau_1 - \tau_2), \end{aligned} \quad (\text{A5})$$

$$\vec{J}(\tau) = i\vec{q} [\delta(\tau - s_1) + \delta(\tau - s_2)], \quad (\text{A6})$$

$$A(\tau_1, \tau_2) = \frac{2Nb^2}{3} \left[-\frac{|\tau_1 - \tau_2|}{2} + \frac{\tau_1 + \tau_2}{2} - \tau_1 \tau_2 \right], \quad (\text{A7})$$

$$\int_0^1 d\tau' A^{-1}(\tau_1, \tau') A(\tau', \tau_2) = \delta(\tau_1 - \tau_2). \quad (\text{A8})$$

So, we obtain

$$e^{\frac{1}{2}(\vec{J} A \vec{J})} = e^{-\frac{q^2 N b^2}{6} [|s_1 - s_2| - (s_1 - s_2)^2]}. \quad (\text{A9})$$

The second term of the equation (9) gets the following form:

$$\begin{aligned} &\int_0^1 ds_1 \int_0^1 ds_2 \int d\sigma [\vec{r} \phi(\vec{r}(s_1)) \phi(\vec{r}(s_2))] = \\ &= \int \frac{d^3 q}{(2\pi)^3} |\tilde{\phi}(q)|^2 \int_0^1 ds_1 \int_0^1 ds_2 e^{-\frac{Nq^2 b^2}{6} |s_1 - s_2|} = \\ &= \frac{1}{N} \int \frac{d^3 q}{(2\pi)^3} |\tilde{\phi}(q)|^2 G(q^2), \end{aligned} \quad (\text{A10})$$

where $G(q^2)$ is so-called Debye's structure-factor:

$$G(q^2) = \frac{12}{q^2 b^2} + \frac{72}{Nb^4 q^4} (e^{-\frac{Nb^2 q^2}{6}} - 1). \quad (\text{A11})$$

On the other hand the first term in Eq.(9) can be written in the form

$$(\phi [D^{-1} - \Phi^{-1}] \phi) = \int \frac{d^3 q}{(2\pi)^3} |\tilde{\phi}(q)|^2 [\tilde{D}^{-1}(q^2) - \tilde{\Phi}^{-1}(q^2)], \quad (\text{A12})$$

Thus, the solution of the equation(9) can be expressed for Fourier transformation of the function D as

$$\tilde{D}(q^2) = \frac{\tilde{\Phi}(q^2)}{1 + cG(q^2) \frac{\tilde{\Phi}(q^2)}{\tilde{\Phi}(0)}}. \quad (\text{A13})$$

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