Biophysical Chemistry 17 (1983) 119–124
Elsevier Biomedical Press

ION BINDING OR CONDENSATION TO POLYELECTROLYTES?

A DETAILED COMPARISON, ON THE EXAMPLE OF SODIUM HEPARINATE *

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Received 13th July 1982
Revised manuscript received 1st October 1982
Accepted 3rd November 1982

Key words: Ion binding; Polyelectrolyte; Poisson-Boltzmann treatment

A modified Poisson-Boltzmann treatment is applied to aqueous solutions of sodium heparinate, in the presence of added salts (NaCl, LiCl, KCl, $CaCl_2$). The results show that ^{23}Na-NMR line widths for the counterions are determined nearly exclusively by Na^+ in the immediate vicinity of the linear polyelectrolyte. Hence, two-state treatments, despite their crude character, can reproduce such experimental data rather well.

1. Introduction

Two rival and seemingly incompatible approaches can be and have been applied to the study of the interaction between ions and polyelectrolytes. One can solve the Poisson-Boltzmann equation, or a modified form thereof [1], to study the atmospheric condensation [2] of fully solvated counterions. Or one can use the altogether different formalism of a chemical equilibrium to determine binding at the various equivalent sites [2] by partly desolvated counterions. We have recently applied the former treatment to the quantitative understanding of the activity coefficients and of the ^{23}Na-NMR line widths for sodium polystyrenesulfonate (NaPSS) in the presence of various added salts [1]. Previously, we had used the alternate, site-binding formalism to account for the ^{23}Na-NMR line widths for sodium heparinate (NaHpr), also in the presence of added salts [3]. In this paper, we compare these two treatments for NaHpr, showing that they converge, rather surprisingly, to rather similar conclusions. More generally, we are able to show that a local property, such as the NMR relaxation rates for the counterion, is determined predominantly by those ions present in the first shell around the polyelectrolyte. Hence, simplified two-state models, whether of the Manning [4] or Guéron and Weisbuch [5] type (condensation [2]) or of the chemical type [3] (site binding [2]) will provide qualitatively equivalent results to those from the more complete and intellectually more satisfying full Poisson-Boltzmann treatment.

2. Experimental

2.1. Materials

Sodium heparinate from Roche (148 U.S.P./mg) is used. The procedures for recording the ^{23}Na-NMR spectra have been detailed in our preceding paper [3].

2.2. Methods

The numerical solution of the modified Poisson-Boltzmann equation is performed [1], after expressing the local concentration of the counter-

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* This paper is dedicated to Professor Edgar Lederer on the occasion of his 75th birthday.

ions as their Boltzmann activities divided by a corrective activity term, expressing the dependence upon the ionic strength of the solution according to ref. 6. A cell model is used, composed of N ($N = 30$–40) coaxial cylindrical shells around the polyelectrolyte, taken as a linear rod with uniform charge density. The thickness of the first shell is taken as the diameter of the solvated counterions [7], a choice, which, while intuitively satisfying, does not have an overwhelming influence upon the results. The electrostatic potential is calculated from the Poisson equation, using as input digitalized concentrations. The electrostatic field gradient is calculated as the second derivative of the potential ($q = \frac{\partial^2 V}{\partial r^2}$) in ref. 1. In order to have a flawless procedure, since in a cell model the electrostatic field gradient tensor is non-diagonal, one should first calculate it in the referential of the counterion: one then finds a diagonalized tensor in which $|V_{zz}| = |V_{yy}| \neq 0$ and $V_{xx} = 0$. The components V_{zz} and V_{yy} are equal to $\frac{\partial^2 V}{\partial r^2}$ and $\frac{1}{r} \cdot \frac{\partial V}{\partial r}$, because at the site of the ion the charge density is formally zero: and $\frac{\partial^2 V}{\partial r^2} + \frac{1}{r} \cdot \frac{\partial V}{\partial r} = -\rho/\varepsilon_0\varepsilon_r = 0$, from the Poisson equation [8]. In our earlier work [1] we calculated

$$\langle e^2q^2\rangle = \sum_{i=1}^{N} \bar{C}_i/C_t \cdot \left(\frac{2}{R^2 - a^2}\right) \int_{r_i}^{r_{i+1}} \left(\frac{\partial^2 V}{\partial r^2}\right)^2 r\,\mathrm{d}r \tag{1}$$

Here, we improve this calculation by taking

$$\langle e^2q^2\rangle = \frac{(1-\eta)^2}{C_t(R^2 - a^2)} \sum_{i=1}^{N} \int_{r_i}^{r_{i+1}} C(r) \left(\frac{\partial^2 V}{\partial r^2}\right)^2 r\,\mathrm{d}r \tag{2}$$

where the asymmetry parameter $\eta = 1$ (axial symmetry), $C(r)$ is the local concentration at a radial distance r from the polymer, $\bar{C}_i$ the average concentration in the i-th shell, and C_t the total concentration of Na^+. Let us also remind the reader of the extreme narrowing ^{23}Na-NMR relaxation rate:

$$1/T_1 = 1/T_2 = \frac{3}{40} \cdot \frac{2I+3}{I^2(2I-1)} \left(\frac{eQ}{h}\right)^2$$
$$\times \langle e^2q^2\rangle (1+\gamma_\infty)^2 \left(\frac{2\varepsilon+3}{5}\right)^2 \tau_C \tag{3}$$

where $I = 3/2$, $(1+\gamma_\infty)$ is the Sternheimer antishielding factor, $(\frac{2\varepsilon+3}{5})$ the polarization factor [9], and τ_C a correlation time descriptive of fluctuations of the electrostatic field gradient. Eq. 3 is fully equivalent with the formalism used in ref. 10.

Eq. 3 is applied to the line broadenings $\Delta\Delta\nu_{1/2}^{\mathrm{exp}} = \Delta\nu_{1/2}(\mathrm{obs}) - \Delta\nu_{1/2}\ (\mathrm{lim})$ [1], with $\Delta\nu_{1/2} = 1/\pi T_2$.

3. Results and discussion

In fig. 1, the ^{23}Na-NMR line broadenings for NaPSS (+HCl, NaCl, $MgCl_2$, and $MnCl_2$) are plotted vs. $\langle e^2q^2\rangle$, calculated using the above formalism (in practice, the results are qualitatively identical to those in ref. 1). The characteristic parameters for NaPSS are a radius $a = 4.8$ Å and a distance between adjacent anionic sites $H = 2.46$ Å [1]. For NaHpr, with $a = 3$ Å [13] and $H = 2.9$ Å [14], the ^{23}Na-NMR line broadenings, in the presence of added LiCl, NaCl, KCl, and $CaCl_2$ [3], are likewise plotted vs. the calculated (Poisson-Boltzmann) $\langle e^2q^2\rangle$ (fig. 2).

Naively, one might have expected entirely different behavior with the two polyelectrolytes, since with NaPSS only slight (≈ 8 Hz) line broadenings are observed [1], as against considerable (> 50 Hz) line broadenings with NaHpr [3]. However, the modified Poisson-Boltzmann treatment is entirely successful, since both sets of results (figs. 1 and 2)

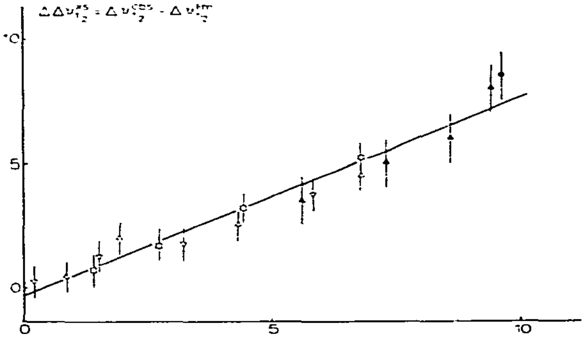

Fig. 1. Experimental line broadenings vs. calculated mean square electrostatic field gradient for NaPSS. Data from ref. 1. (●) NaPSS, (▲)+NaCl, (▽)+HCl, (□)+$MgCl_2$, (△)+$MnCl_2$.

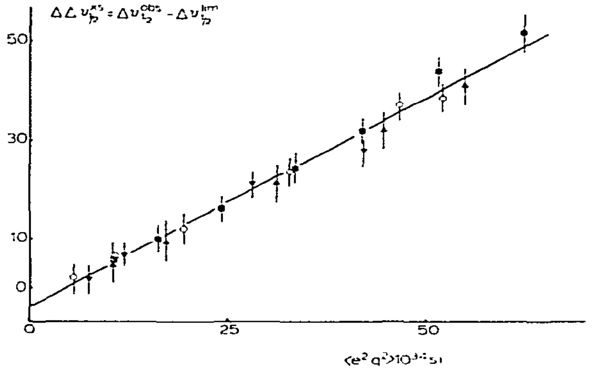

Fig. 2. Same as in fig. 1: data from ref. 3. (●) NaHpr, (▲) + NaCl, (■) + KCl, (▼) + LiCl, (○) + $CaCl_2$.

reduce to the same slope $(0.79 \pm 0.05) \times 10^{-34}$ (NaPSS) and $(0.85 \pm 0.02) \times 10^{-34}$ (NaHpr) as implied by eq. 3.

This success, to some degree, might result from a favorable coincidence. Firstly, the linear regression obtained with each of the two polyelectrolytes implies invariance of its geometry with ionic strength of the solution. Yet, it is known for conformational changes to occur with increasing salt content [11]. What should be emphasized, though, is that the results are determined predominantly by the mean polymeric radius and intercharge separation, whose variations are not likely to be large. Such a result is in full accord with earlier conclusions by Leyte et al. [12]. Secondly, we have deliberately refrained from any adjustment procedure: the geometrical parameters a and H quoted above are standard values from the literature, and we have not attempted to select other values which might have provided an improved fit with the results.

However conforting this success is, we would still like to explain the physical origin of the much larger ^{23}Na-NMR line broadenings observed with NaHpr. For this purpose, let us first consider the relative importance of successive terms in eq. 2: $\langle e^2q^2\rangle$ has values of 62.042, 0.049, 0.016, 0.005, and 0.002×10^{34} SI (J M^{-5}) for shell Nos. 1–5, respectively; and a total value of 62.116×10^{34} SI for NaHpr in aqueous solution, without added salt. Clearly, only the contribution from the first shell is important. Furthermore, detailed examination of the numerical results shows that the first shell contribution to $\langle e^2q^2\rangle$ originates predominantly, to the extent of 80–90%, from the polymer. To a good first approximation, one can neglect the influence of other ions. In other terms, the line width characteristic of counterions in the immediate vicinity of the polymer is much greater than the line width of ions further away and is only polymer dependent: this is exactly what would have been predicted, intuitively.

Indeed, since the $\langle e^2q^2\rangle$ term is proportional to ρ^2, where ρ is the charge density on the polyelectrolyte, it is not surprising that NaHpr should give rise to the largest line broadenings:

$$\rho^2 \propto (\text{volume})^{-2} = \pi^{-2}H^{-2}a^{-4} \tag{4}$$

From eq. 4, $\rho^2(\text{NaHpr})/\rho^2(\text{NaPSS}) = 6$ with the values of a and H given earlier, and a sulfonation degree of 0.89 for NaPSS. In comparison, the ratio of the line broadenings is approx. 6.5.

Hence, experimental results are determined mostly by the dimensions of the polyelectrolyte: line broadenings induced by NaHpr are much larger because the polyelectrolyte has a significantly smaller radius than that of NaPSS.

Let us now proceed to write the observed $\Delta\nu_{1/2}$ by the highly simplified expression:

$$\Delta\nu_{1/2} = p_{\mathrm{Na_C}}(\Delta\nu_{1/2})_{\mathrm{C}} + p_{\mathrm{Na_F}}(\Delta\nu_{1/2})_{\mathrm{F}} \tag{5}$$

where $p_{\mathrm{Na_C}} = 1 - p_{\mathrm{Na_F}}$ is the mole fraction of condensed Na^+, and $(\Delta\nu_{1/2})_{\mathrm{F}}$ and $(\Delta\nu_{1/2})_{\mathrm{C}}$ the limiting line widths for the free and for the condensed (first shell) ions, respectively. Let us then define:

$$\Delta\nu_{1/2} - (\Delta\nu_{1/2})_{\mathrm{F}} = \Delta\Delta\nu_{1/2}^{\mathrm{ex}} = (\Delta\Delta\nu_{1/2})_{\mathrm{C}} \cdot p_{\mathrm{Na_C}} \tag{6}$$

(since $(\Delta\Delta\nu_{1/2})_{\mathrm{F}} = 0$). Application of eq. 6 to the experimental results yields the limiting value of $(\Delta\Delta\nu_{1/2})_{\mathrm{C}}$, i.e., the line broadening characteristic of Na^+ condensed in the first shell around the polyelectrolyte (tables 1 and 2).

As the radius a of the polyelectrolyte tends towards zero, the Poisson-Boltzmann and Manning models converge to the same answers [16], as is apparent from comparison of the NaHpr (A) and NaPSS (B) entries in table 1: with the thinner

Table 1

Molar fractions of condensed Na^+ in the first shell, line broadenings from figs. 1 and 2, and limiting line broadenings in the first shell, according to the Poisson-Boltzmann and Manning treatments

Results for NaHpr ($(\Delta\nu_{1/2})_F = 4.2$ Hz) (A), and for NaPSS ($(\Delta\nu_{1/2})_F = 6.3$ Hz) (B), with NaCl added, from refs. 1 and 3. Note the constancy of $(\Delta\Delta\nu_{1/2})_C$ in the Poisson-Boltzmann (P-B) approach. $\overline{(\Delta\Delta\nu_{1/2})_C} = (98 \pm 10)$ Hz for sodium heparinate and (16 ± 3) Hz for NaPSS.

	C_p (M)	C_t (M)	1st-shell		$\Delta\Delta\nu^{\rm exp}_{1/2}$ (Hz)	$(\Delta\Delta\nu_{1/2})_C$ (Hz)	
			p_{Na} (P-B)	p_{Na} (Manning)		P-B	Manning
(A)	0.0648	0.0648	0.502	0.584	58.8	117	101
	0.0648	0.0768	0.442	0.492	43.8	99	89
	0.0648	0.102	0.356	0.372	34.8	98	94
	0.648	0.158	0.252	0.240	23.8	94	99
	0.648	0.340	0.139	0.111	12.3	88	110
	0.0648	0.667	0.086	0.057	7.8	90	137
(B)	0.0221	0.0221	0.416	0.596	7.9	19	13
	0.021	0.0215	0.409	0.583	7.5	18	13
	0.0169	0.0192	0.372	0.525	5.5	15	11
	0.0122	0.0166	0.314	0.438	4.5	14	10
	0.0078	0.0138	0.244	0.337	3.0	13	9

Table 2

Molar fractions of condensed Na^+ in the first shell, line broadenings from fig. 2, and limiting line broadenings in the first shell, according to the Poisson-Boltzmann treatment for NaHpr (0.0648 M) in the presence of various added salts

Note the constancy of $(\Delta\Delta\nu_{1/2})_C$ in the Poisson-Boltzmann (P-B) approach. $\overline{(\Delta\Delta\nu_{1/2})_C} = (100 \pm 8)$ Hz for this set of data. $C_p = 0.0648$ M; $(\Delta\nu_{1/2})_F = 10.3$ Hz for LiCl, 4.2 Hz for KCl, 5.2 Hz for $CaCl_2$.

[Salt] (M)	1st-shell p_M		1st-shell p_{Na}		$\Delta\Delta\nu^{\rm exp}_{1/2}$ (Hz)	$(\Delta\Delta\nu_{1/2})_C$ (Hz)	
	P-B	Manning	P-B	Manning		P-B	Manning
LiCl							
0.0401	0.408	0.359	0.342	0.359	31.6	92	88
0.106	0.273	0.221	0.229	0.221	24.8	108	112
0.461	0.111	0.071	0.098	0.071	10.3	105	145
0.935	0.072	0.038	0.062	0.038	5.6	90	146
KCl							
0.0201	0.436	0.444	0.415	0.444	45.8	110	103
0.0472	0.356	0.337	0.339	0.337	33.8	100	100
0.0873	0.283	0.248	0.269	0.248	25.8	96	104
0.169	0.157	0.161	0.195	0.161	17.8	91	110
0.343	0.138	0.092	0.129	0.092	11.8	92	128
0.686	0.0915	0.051	0.0855	0.051	7.6	88	140
$CaCl_2$							
0.00451	0.995	1	0.420	0.444	41.3	98	93
0.00702	0.988	1	0.376	0.367	39.8	106	108
0.014	0.962	1	0.265	0	26.3	99	–
0.0241	0.860	1	0.157	0	14.8	94	–
0.0441	0.609	0.582	0.0878	0	9.3	106	–
0.853	0.364	0.300	0.0593	0	6.8	115	–
0.168	0.212	0.153	0.0447	0	4.8	107	–

NaHpr rod, the discrepancy between the Poisson-Boltzmann results and the Manning approximation is less than with the thicker NaPSS macroion. For NaHpr and NaPSS, in the presence of added NaCl (table 1), the invariance (98 ± 10 Hz) with total sodium concentration C_t of the limiting line broadening in the first shell $(\Delta\Delta\nu_{1/2})_C$ is a good index of the validity of a two-state model as a simplification of the Poisson-Boltzmann approach (eq. 5). By contrast, the Manning approximation is less successful, leading to variable limiting line broadenings ranging between 90 and 140 Hz (table 1). The ratio of $(\Delta\Delta\nu_{1/2})_C$ values for the two systems, i.e., 98/16 = 6.13, is that predicted on the basis of the geometrical factors summarized above.

In table 2, we provide another comparison between the results of the Poisson-Boltzmann approach and those from the Manning theory, for the competition between Na^+ and Li^+, K^+ or Ca^{2+}. Again, we find a very satisfactory constancy of the $(\Delta\Delta\nu_{1/2})_C$ values (100 ± 8 Hz). It is indeed remarkable to find such nicely consistent $(\Delta\Delta\nu_{1/2})_C$ values for the two sets of experiments, differing in the nature of the added salts which include both univalent (Na^+, K^+, Li^+) and divalent (Ca^{2+}) cations. As for the Manning model, it is unable to account for competition between ions having the same charge: it will treat them in a purely statistical manner without distinguishing between differing chemical species – which our modified Poisson-Boltzmann approach tells apart from the size of the first shell, that of the hydrated ion. Furthermore, the Manning model introduces considerable bias in competition between uni- and divalent ions, to the exclusive benefit of the former (see the first four entries for $CaCl_2$ in table 2).

With the applicability of eq. 5 thus borne out by the $(\Delta\Delta\nu_{1/2})_C$ values (tables 1 and 2), we can now exploit the corresponding two-state model for a chemical equilibrium treatment of competition between the various cations for condensation (not binding!) to the polyelectrolyte. For this purpose, denoting by A_F^- an anionic site (-COO$^-$ or -OSO_3^-) on heparin of total concentration equal to the equivalent polymer concentration C_p:

$$Na_F^- + A_F^- \overset{K_{Na}}{\rightleftarrows} Na_C$$

$$M_F^+ + A_F^- \overset{K_M}{\rightleftarrows} M_C$$

we use the values from table 1 to yield $K_{Na} = 25 \pm 3$ M^{-1}. Then, the values from table 2 provide K_M values, with an interesting possibility for an internal check, since they can be derived from either the mole fractions p_{M_C} or p_{Na_C}, using a Newton-Raphson least-squares minimization, or a Simplex (Ca^{2+}) procedure [17]: $K_{Li} = 27 \pm 2$ (p_{Li_C}); 40 ± 7 (p_{Na_C}); $K_K = 23 \pm 2$ (p_{Li_C}); 33 ± 3 (p_{Na_C}); $K_{Ca} = 73\,000$ (p_{Ca_C}); $139\,000$ (p_{Na_C}). In the latter case, a 1 : 1 stoichiometry does not work; a 1 : 2 stoichiometry:

$$Ca_F^{2+} + 2A_F^- \rightleftarrows Ca_C$$

has to be used in order for self-consistency to be achieved. Such a stoichiometry is not surprising, because of the requirement for electroneutrality, and has already been documented elsewhere in the atmospheric-condensation concept [15], or in the site-binding concept[3]. That there is only order-of-magnitude agreement between the K (or ΔG) values obtained from p_{M_C} or from p_{Na} data points to the limits of a two-state model. One should not forget that it is merely an approximation to the 'true' amount of condensation given by the full modified Poisson-Boltzmann treatment.

Also, there are the approximations and limitations inherent in the Poisson-Boltzmann treatment [1], which we shall merely list here since they have been discussed in detail [1]: the polyelectrolyte is assumed to have a rigid rod geometry; the cell model demands that there be no interpenetration between adjoining cells; interactions between polyions are neglected; the excess ΔG is purely electrostatic; the solvent is treated as a continuum, in which are imbedded point charges (ions) and a homogeneous charge distribution(the polyion); ion-ion correlation is neglected. In our modified Poisson-Boltzmann treatment, we introduce an empirical correction to account for ion-ion correlation as a function of the ionic strength. We set the thickness of the first shell commensurate with the size of the solvated ions. And the digitalization used in the calculation is equivalent to analytical resolution of the Poisson-Boltzmann equation [1].

Finally, we should detail the assumptions made in calculating the electrostatic field gradient: rather than the point charges demanded by the formalism [8], we have to carve holes in the dielectric continuum at the position of each counterion; we have to assume fast chemical exchange, in order to write eq. 5, between ions condensed in the first shell and those in subsequent shells; and, strictly speaking, a point charge in the first shell of our cell model does not undergo any reorientational time fluctuation of the electrostatic field gradient: hence, the significance of τ_C (eq. 3) must be sought either in the deficiencies of the model, e.g., in distortions from the full O_h symmetry of $Na^+ \cdot 6H_2O$, in the finite size of the counterions, or in the deformation modes of the polymer; or, within the framework of the model, as Leyte et al. [10] have shown, in diffusional excursions of the probe ion along equipotential surfaces.

However, lest these reservations throw doubt upon the validity of eq. 3, we want to stress again [1] that figs. 1 and 2 provide a unique confrontation between experiment and theory (eq. 3), thus vindicating completely the use of eq. 3. The analysis can only give a value of the product $(1+\gamma_\infty)^2\tau_C$. In order to go further, one has to make assumptions: with a realistic upper limit for τ_C of 10^{-10} s, the Sternheimer anti-shielding factor $(1+\gamma_\infty) \approx 7.0$). This value applies to the fully solvated Na^+ in the first shell, and compares well with the value of approx. 5 appropriate for the isolated Na^+.

In conclusion, two-state treatments of various sorts [3–5] of the polyelectrolyte-counterion interaction, as studied by NMR, are appropriate and successful for the reasons outlined above. A condensation model, as applied here to the NaHpr system, is superior to a site-binding model [3] in completeness: not only NMR observables but also activity coefficients can be predicted [1], and it does not require any parametrization. A choice between the two models amounts to determination of the structure of first-shell counterions: if these are solvated, then a modified Poisson-Boltzmann approach should be applied; if these are partly desolvated, a chemical equilibrium becomes legitimate. However, such an experimental test, in the state of the art, is still a pending (and nagging) question [1,18,19]!

Acknowledgments

We thank Professor J.C. Leyte, University of Leiden, for clarification of the electrostatic field gradient tensor calculation, and Fonds de la Recherche Fondamentale Collective, Brussels, for grants 2.4504.78 and 21420.D which made possible purchase of the Bruker WP-80 spectrometer used in this study. The calculations were performed with the IBM 4341 computer, of the S.E.G.I., Université de Liège. We thank Dr. Eric Enwall of the University of Oklahoma for the use of his generalized least-squares program.

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