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SITE BINDING AS A LOCAL EQUILIBRIUM

Mn^{2+} BINDING TO POLY(ACRYLIC ACID) AS A FUNCTION OF THE DEGREE OF IONIZATION

B. MEURER, P. SPEGT and G. WEILL

Centre de Recherches sur les Macromolécules, C.N.R.S., and Université Louis Pasteur, Strasbourg, France

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Mn^{2+} binding to poly(acrylic acid) at different degrees of ionization, α, has been studied from the frequency dependence of the water protons' relaxation rates T_1^{-1} and T_2^{-1}. Site binding is treated as an equilibrium with the concentration of free ions at the immediate vicinity (CIV) of the polyion. The CIV is calculated as the solution of the Poisson-Boltzmann equation at the surface of the cylindrical polyion. A single value of K is shown to fit the results at all values of α. The amount of site binding is higher than the total amount of condensed divalent counterions predicted for a finite polyion concentration in the presence of monovalent counterions by Manning's theory.

1. Introduction

In a series of preceding papers [1–4], we have shown that magnetic resonance techniques can distinguish between three types of divalent counterions in solutions of highly charged polyelectrolytes. The first one experiences a disorganization of its hydration shell and a loss of its mobility. These simultaneous changes are taken as a definition for 'site binding'. The second one retains its hydration shell but can be distinguished from the third one, the 'free counterions', on the basis of NMR relaxation measurements which suggest a still restricted mobility. We have called them 'atmospherically bound' and Manning [5] speaks of 'territorial binding' or 'delocalized binding'. It has been repeatedly suggested [6–8] that site binding could be treated as an equilibrium between the ionized groups carried by the polyion and the high local concentration in its immediate vicinity (CIV). Since the models currently used for the calculation of the counterion distribution (either from the solution of the P.B. equation or from Manning's condensation model including the evaluation of the condensation volume [9]) treat the polyion as a continuous distribution of charge and the counterion as a point charge, the local equilibrium hypothesis takes care of all the chemical specificity originating from the nature of the two ionic species in a simple equilibrium constant K. A test of the model requires, therefore, demonstration that the same value of K can be used to interpret the degree of site binding for a series of polyions carrying the same ionized group at different mean distances along or from the chain. Copolymers with a non-ionizable monomer, polymers carrying the same acid group at the end of side chains of variable length, or weak polyacids at variable degrees of ionization α can be used for that purpose. We have chosen the last case and we report in this paper the experimental and theoretical study of Mn^{2+} site binding to poly(acrylic acid) (PAA) at

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different α. The NMR relaxation of water in this system has been studied by Westra and Leyte [10], but for ratios r of Mn^{2+} to the monomolar concentration in ionized groups which did not permit using their results as a basis for our calculations (see below). Our results establish the validity of the local equilibrium hypothesis in this system.

2. Formulation of the site binding hypothesis

We assume that the formation of an ion pair between the divalent counterions and the acid groups does not require the chelation of Mn^{2+} by two COO^-. The formation of a $(COO^- Mn^{2+})$ ion pair decreases, however, the charge of the polyion by two elementary charges. One can therefore define formally the binding site as two neighbouring COO^- and write:

$$\mathrm{Pol(COO^-)_2 + Mn^{2+} \rightleftharpoons Pol(COO)_2Mn}$$

Denoting by x the fraction of 'site bound' acid groups and taking for the concentration in divalent ions the local concentration of the territorially bound Mn^{2+} at the surface of the polyion $[CIV_2]$, the competitive aspects of metal binding as compared to protonation are treated approximately by the equilibrium relationship:

$$K = \frac{x}{(1-x)[CIV_2]} \qquad (1)$$

Since the accumulation of divalent counterions in the strong electrostatic potential of the polyion is a function of the effective surface charge, $[CIV_2]$ depends on x as well as on the polyion diameter, the composition of the solution in monovalent and divalent counterions and the polyion concentration. It can be calculated from solutions of the Poisson-Boltzmann equation or from the latest version of Manning's condensation model.

We know from previous work [2,3] that for the Mn^{2+}-PAA system at $\alpha = 1$, site binding is complete up to $r \approx 0.4$. In this work our purpose is to measure the value of x for the same system at variable α. Deriving a value of K from the first piece of information, we want to show that this value of K can be used to predict satisfactorily changes of x with α.

3. Material and methods

Two stock solutions of PAA (Polysciences $M_r \approx 250000$) of the same concentrations in water have been prepared, one being neutralized by the appropriate quantity of tetramethylammonium (TMA) hydroxide. They are mixed in suitable proportions for each required value of α. The concentrations to be used in our experiments have been chosen according to the following constraints. In order to measure with enough accuracy the frequency dependence of the change in the water protons' relaxation rates produced by Mn^{2+} counterions in the presence and absence of polyions, concentrations of $Mn^{2+} > 10^{-3}$ M, giving $T_1 < 10^{-2}$ s, are required. A concentration of Mn^{2+} of 3.25×10^{-3} M has been chosen and the required quantity of $MnCl_2$ added to the stock solutions. Due to the difference in the titration curves for the hydroxides of monovalent and divalent metal ions [11], this concentration has to be kept small in comparison with the monomolar concentration of COO^- groups at small α in order not to displace the actual value of α. Since our lowest α value was 0.1, a total concentration in monomer units of 0.6 M has been used. Thus, the values of $r = 0.0108\ \alpha^{-1}$ are limited to rather low values. The work of Westra and Leyte [10] has been performed with the same Mn^{2+} concentrations but with concentration in monomer units of 2.5×10^{-2} M. Under these conditions, $r \approx 0.26$

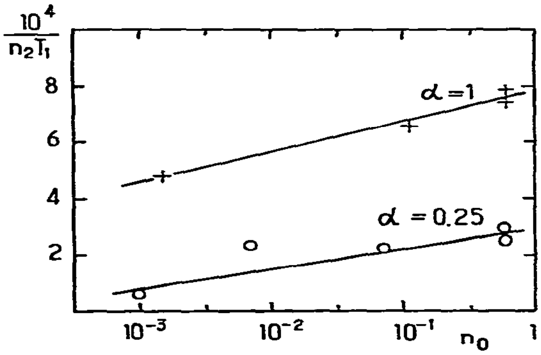

Fig. 1. Dependence of the relaxation rates at 40 MHz normalized to the Mn^{2+} concentration with the polymer concentration for two ionization degrees, $\alpha = 0.25$ and $\alpha = 1$.

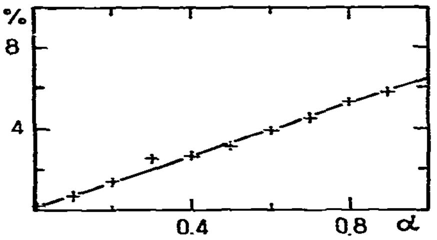

Fig. 2. Relative amplitude of the slow component of the transverse relaxation as a function of α.

α^{-1}. There is consequently a large uncertainty in the low values of α and the necessary presence of large amounts of free Mn^{2+} up to $\alpha \approx 0.4$ makes the determination of site binding very imprecise. We shall see, however, that their interpretation of their data at $\alpha > 0.6$ is in good agreement with our results. In order to check that there is no special effect attached to the large polymer concentration, we have performed a series of measurements at 40 mHz, on diluted solutions for $\alpha = 1$ and 0.25. One sees in fig. 1 that the increase in relaxation rate per mole of added Mn^{2+} is a smooth linear function of the log of concentration.

Our proton relaxation measurements have been carried out using for T_1 the inversion recovery method and for T_2 the Carr-Purcell-Meiboom-Gill method.

Data acquisition and treatment have been fully automated, time constants being extracted from the relaxation curve using a non-linear iterative least-squares curve fitting [12] to the expression:

$$M(t) = A\exp(-R_{\text{fast}}\cdot t) + B\exp(-R_{\text{slow}}\cdot t) + C$$

A one-exponential fit was always sufficient for the longitudinal relaxation while the faster transverse relaxation presented a two-exponential behaviour due to the TMA^+ protons. This appears clearly in fig. 2 where the ratio $B/(A+B)$ is shown to be a linear function of α with a value at $\alpha = 1$ corresponding exactly to the mole ratio of the TMA and H_2O protons ($12 \times 6\ 10^{-1}/2 \times 55 = 0.065$).

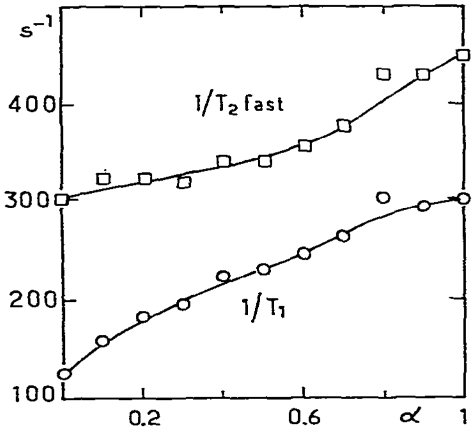

Fig. 3. Longitudinal and transverse relaxation rates of the water protons at 60 MHz as a function of α.

4. Results

A plot of R_1 and $R_{2,\text{fast}}$ as a function of α at 60 MHz is shown in fig. 3. It is clear that although R_1 seems to be constant above $\alpha = 0.8$ and R_2 seems to be constant below $\alpha = 0.3$, there is no clear discontinuity which could be used as an indication of the change of the counterion state. Care must

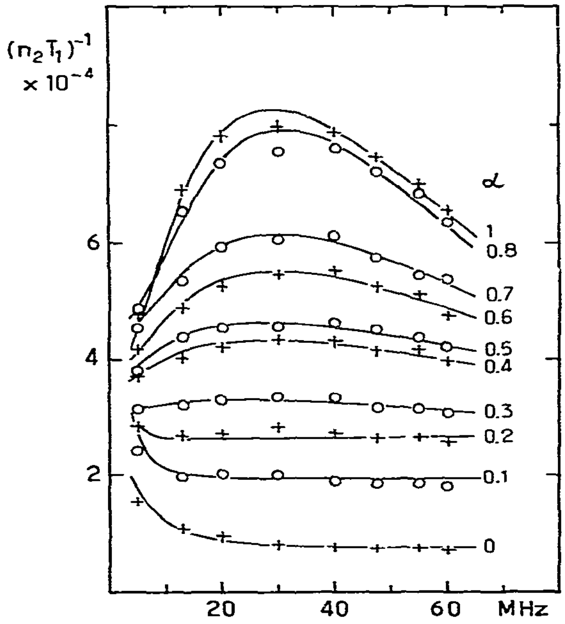

Fig. 4. Frequency dependence of the longitudinal relaxation rate for different α. The calculated curves are best fits to the Bloemberger-Morgan equations.

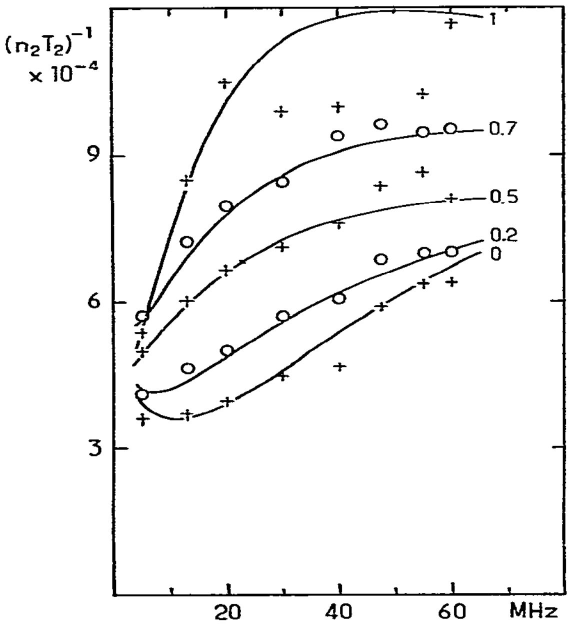

Fig. 5. Frequency dependence of the transverse relaxations rate for different α.

be exercized in the interpretation of data at a single frequency and the full frequency dependence must be analyzed at each value of α. The results are displayed in figs. 4 and 5. Characteristic qualitative changes, already associated with the presence of site binding [3], appear around $\alpha = 0.3$. For R_1, the curvature changes sign, while the flattening or upward curvature of R_2 at low frequencies is replaced by a steep decrease and downward curvature. The quantitative analysis of the frequency dependence using the Bloembergen-Morgan formulae [13,14] involves six parameters for each type of counterion. Three correlation times τ_R, τ_M and τ_V are associated with the rotation of the counterion, the exchange time of water and the modulation of the electron spin zero-field splitting B. Besides B, there are two other coupling constants associated with the dipolar D and scalar C interaction between the electronic and nuclear spins. There is no hope of extracting so many parameters from the experiments, and our previous [3] results indicate that the correlation times of territorially bound counterions may be significantly lengthened. For that reason, we have not tried to postulate in a first approach the existence of two types of counterions and have analyzed our results in terms of a single population of counterions, using a procedure similar to that described in ref. 3. The resulting parameters, the value of C and D being given as their ratio to that of C_0 and D_0 of free Mn^{2+}, are given in table 1. The corresponding calculated frequency dependences are given as full lines in figs. 4 and 5. The values of D/D_0 and τ_R as a function of α are plotted in fig. 6. From the steep

Table 1

Parameters for the curve fitting of the frequency dependence of the proton relaxation rates in the presence of Mn^{2+} and PAA at different α

α	$D/D_0 = C/C_0$ [a]	τ_R (s) ($\times 10^{11}$)	τ_M (s) ($\times 10^8$)	B (Hz) ($\times 10^{-18}$)	τ_V (s) ($\times 10^{12}$)
0	1	3.8	2	7	5.3
0.1	1	10	2	10	6
0.2	0.66	21	1.7	7	9
0.3	0.21	83.5	3	2	15
0.4	0.23	105	3	4.2	15
0.5	0.23	115	3	4.5	16
0.6	0.23	145	3	7	14
0.7	0.23	173	3	8	11
0.8	0.27	245	2.5	12	9
0.9	0.25	250	2.5	12	11
1	0.26	235	2.5	11	17
$MnCl_2$	1	2.8	2	8	13

[a] This ratio has been taken to be equal for D and C in the curve-fitting procedure.

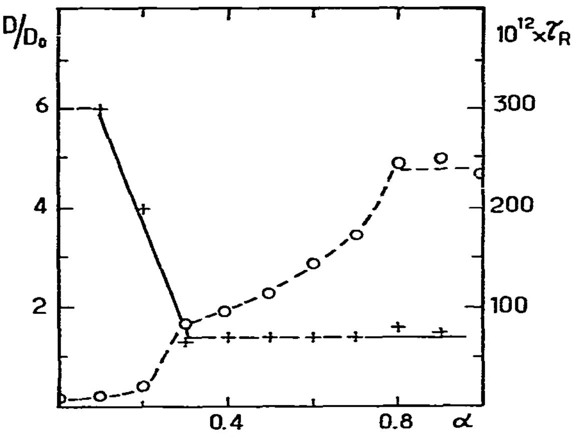

Fig. 6. Variation of the parameters $D/D_0(+)$ and $\tau_R(0)$ as a function of α.

change at $\alpha \approx 0.2$ and the constancy of D/D_0 above $\alpha = 0.3$ one can conclude that Mn^{2+} is then completely site bound, the change in relaxation rates above $\alpha = 0.3$ being essentially associated with the change in τ_R. In order to confirm this result, we have carried out experiments at $\alpha = 0.25$ at several very low values of r and a lower concentration of PAA (7.2×10^{-2} M). The relaxation rates are a linear function of r and the frequency dependence of R_1 shows no indication of a sizeable fraction of free ions. We therefore conclude that below $\alpha = 0.1$ with $r > 0.108$, all the counterions are free and above $\alpha = 0.3$ with $r < 0.036$ they are all bound.

5. Poisson-Boltzmann approach to the calculation of CIV_2

The calculation of CIV_2 in the presence of a mixture of monovalent and divalent counterions at a finite concentration can be carried out in the framework of the cell model [15]. The concentration in ionized groups n_0 (in mol l^{-1}) separated along the chain by a distance b, fixes the radius of the cell

$$R^2 = \frac{10}{6\pi n_0 b} = \frac{10\xi}{6\pi n_0 l_B} \text{Å}^2$$

where l_B is the Bjerrum length and $\xi = l_B/b$ the classical linear charge parameter of the polyelectrolyte theories. Taking the polyion as a charged cylinder of radius a, the concentration in counterions of valence 1 and 2 being given by n_1 and n_2, the Poisson-Boltzmann equation can be written in terms of the reduced variable $x = \rho/a$ (ρ distance from the polyion axis) and $\phi = e\psi/kT$ (ψ electrostatic potential)

$$\frac{d^2\phi}{dx^2} + \frac{1}{x}\frac{d\phi}{dx} = \frac{\epsilon^2}{2+\beta}[(1-\beta)\exp\phi + \beta\exp(2\phi) - \exp(-\phi)] \tag{2}$$

with the boundary conditions

$$\left(\frac{d\phi}{dx}\right)_{(x=1)} = -2\xi$$

$$\left(\frac{d\phi}{dx}\right)_{(x=R/a)} = 0$$

In a dilute solution the electrostatic potential is a rapidly decreasing function from the surface of the cylinder to the boundary of the cell. We suppose that the cell radius R is sufficiently large ($R/a > 5$) for the potential to reach a zero value at the boundary. This implies that the concentration of counterions at R is equal to that in Donnan equilibrium with the polyelectrolyte solution n_1^0 and n_2^0. Under these conditions:

$$\beta = 2n_2^0/(n_1^0 + 2n_2^0)$$

and

$$\epsilon^2 = 2.4 l_B(n_1^0 + 2n_2^0)a^2$$

The solutions have been calculated using two schemes developed by Gueron and Weisbuch [7,16]. The first one is an iterative scheme initially developed for infinitely diluted solutions which we have adapted to the case of concentrations which are not too high. n_1^0 and n_2^0 are fixed at the cell boundary, a value of $\phi(1)$ is guessed and $\phi(x)$ is calculated by stepwise integration. The value of $\phi(1)$ is then modified in such a way that ϕ remains positive and its derivative negative. The convergence is fast. Within the numerical accuracy of the computer, the value of ϕ at the cell boundary has always been found to be equal to zero up to $n_1^0 = 0.1$ N. From the final value of $\phi(x)$ the CIV is calculated from:

$$\mathrm{CIV}_i = n_i^\circ \exp[z_i \phi(1)] \tag{3}$$

and the total concentration in counterions from:

$$n_t = \frac{n_i \int_1^{R/a} 2\pi x \exp[z_i \phi(x)]}{\int_1^{R/a} 2\pi x \, \mathrm{d}x} \tag{4}$$

The calculation have been performed on a Hewlett-Packard 9830 mini-computer, using a Runge-Kutta method of order 2. The results have been checked against those obtained by Westra and Leyte [17] using the procedure proposed by Dolar and Peterlin [18] which does not suppose a zero potential at the cell boundary. β and ϵ are then a function of the solution $\phi(x)$ which makes the procedure of numerical integration much more complex. The agreement is good.

The methods breaks down, however, at high polymer concentration (≈ 0.1 N) where R/a becomes too small. We have therefore also used the heuristic method of Weisbuch and Gueron [16] which derives the CIV for a cylinder from the exact solution for a plane of the same surface charge density σ. For the planar case there exists an invariant S proportional to σ^2 and related to the potential ω at the surface of the plane and to the differences between the CIV values and the concentrations far from the surface by:

$$S = 2\pi l_\mathrm{B}\left(\frac{\sigma}{e}\right)^2 = \sum_i (\mathrm{CIV}_i - n_i^\circ) = \sum_i n_i^\circ |\exp(z_i\omega) - 1| \tag{5}$$

The surface potential ω is then easily computed by solution of this algebraic equation. A correction to ω for the cylindrical case of the same charge density gives the surface potential for the given values n_i°:

$$\phi(1) = \omega - \frac{2}{z'}\left(\xi z' - 0.45 + 1.8\sqrt{\epsilon/\xi z'} + 3\epsilon^2/\xi z'\right)^{-1} \tag{6}$$

with

$$z' = \frac{1 + (2\alpha_2/3\alpha_1)\exp\omega}{1 + (\alpha_2/3\alpha_1)\exp\omega}$$

where

$$\alpha_i = n_i^\circ / \sum n_i^\circ z_i^2$$

One calculates then from eq. 3 the CIV_i for the cylindrical case, using for n_i° the value at the cell boundary. The fraction of ions i per unit charge of polymer N_{it} is obtained from:

$$N_{it} = \mathrm{CIV}_i / (3S \exp(-2\phi_i) - \mathrm{CIV}_i) \tag{7}$$

where ϕ_i is the value of ϕ_1 calculated from eq. 6 with only one type of counterion i. One has directly $r = 2N_{2t}$.

We have checked that in the domain of concentration where both algorithms can be used, extending from 10^{-3} to 10^{-1} N, the values of CIV_2 are in close agreement. A typical plot of CIV_2 as a function of r is given in fig. 7 for $a = 4$ Å and different values of the charge parameter ξ. It is seen that for values of $r < 0.4$ the variation is linear. The slope CIV_2/r has been calculated in a range of values of $\xi (0 < \xi < 3)$ corresponding to a variation of ionization degree α of PAA between 0 and 1. As shown in fig. 8 it is practically independent of the polymer concentration in a broad range corresponding to our different EPR and NMR experiments. It is well represented by the relationship:

$$\frac{\mathrm{CIV}_2}{r} \simeq 3\left(\xi - \frac{1}{2}\right)^2 \tag{8}$$

The treshold value $\xi = 1/2$ for the rise of CIV_2 is reminiscent of the condensation hypothesis and can be compared with an equivalent result derived

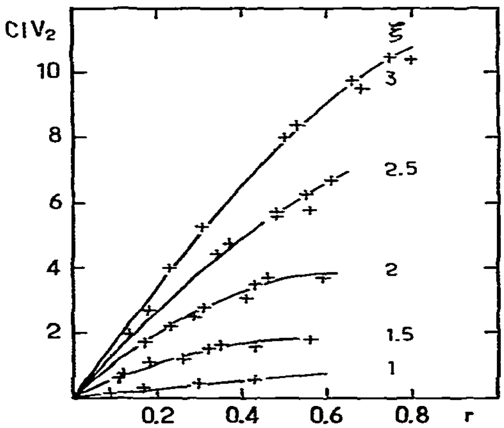

Fig. 7. Variation of the CIV_2 with the fraction of divalent counterions r for different charge parameters (concentration of monomer $n_0 = 0.082$ M, polyion radius 0.4 nm).

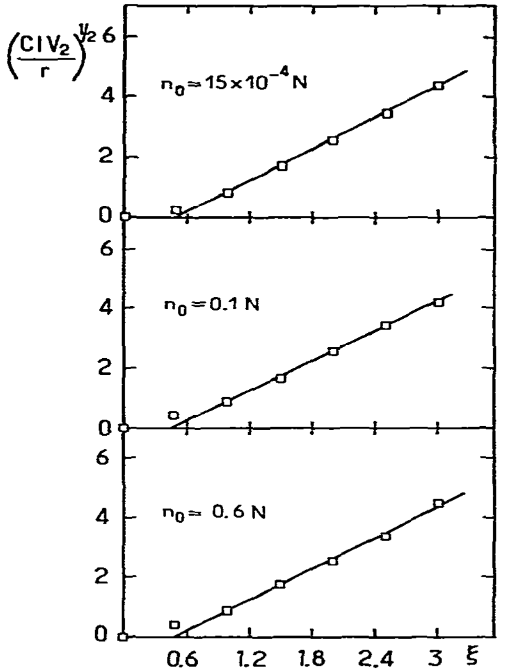

Fig. 8. Square root of the ratio CIV_2/r as a function of the charge parameter ξ for different concentrations of monomer n_0.

recently by Anderson and Record [19] for polyelectrolytes with only monovalent counterions.

$$CIV_1 = \frac{37.0}{a^2}(\xi-1)^2 \tag{9}$$

6. Calculation of the binding curves

Taking into account the fact that site binding reduces the apparent charge of the polyion from ξ to $(1-x)\xi$ and the fraction of free divalent counterions from r to $r-x$, one obtains from eqs. 1 and 8 the equation:

$$3K(1-x)[a\xi(1-x)-\tfrac{1}{2}]^2(r-x)+x=0 \tag{10}$$

We have first calculated x as a function of r for different values of K, taking for the fully ionized PAA $\xi=3$, $a=4$ Å. It can be seen in fig. 9 that in order to explain an essentially complete binding up to $r=0.4$, which corresponds grossly to our analysis of the relaxation measurements, one can use $K=6.4$. Despite the difference between Mn^{2+} and Co^{2+}, it was tempting to use the same equilibrium constant to recalculate the r dependence of the water chemical shift obtained with the latter counterion [2]. In these experiments the chemical shift of the water protons is related to the hyperfine coupling constant C of the magnetic counterion with the protons of its water hydration shell. It is likely to be different for site-bound and free counterions. We have therefore calculated the curve

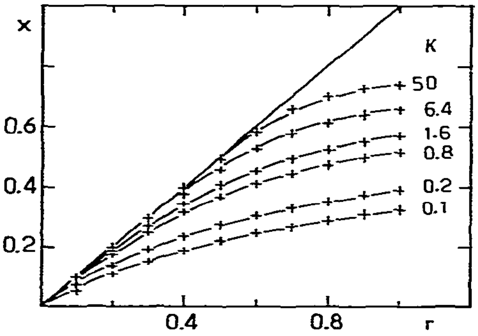

Fig. 9. Dependence of the site-bound fraction of the polyion charges on the total fraction of divalent counterions r for different values of the binding constant K.

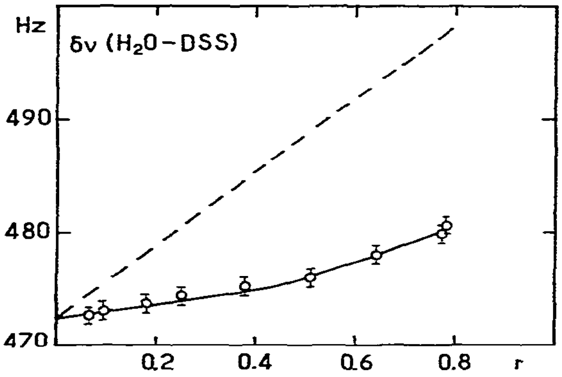

Fig. 10. Calculated dependence of the chemical shift of the water protons in the presence of Co^{2+} and PAA, $\alpha=1$. The dotted line corresponds to Co^{2+} in the absence of PAA. The experimental points are taken from ref. 2. DSS, 2,2-dimethyl-2-silapentane 5-sodium sulfonate.

of fig. 10 according to:

$$\delta\nu = \delta\nu_0 + \beta\frac{C}{C_0}x + \beta(r - x)$$

where $\delta\nu_0$ is the chemical shift of water in the absence of Co^{2+}, C/C_0 the ratio of the hyperfine constants for site-bound and free counterions, and β the value of $\delta\nu$ measured for a concentration of Co^{2+} corresponding to $r = 1$ in the absence of polyelectrolyte.

The curve of fig. 10 has been calculated using for each value of r the corresponding value of x for $K = 6.4$ and $C/C_0 = 0.15$, close to that obtained from the frequency dependence of the relaxation rates (table 1 and ref. 4). The agreement with the experimental data is very good.

We have then calculated for the same value of K the binding curves $x(r)$ for different values of α. The result is given in fig. 11. As already seen the CIV is negligible for $\alpha\xi < 0.5$, i.e., $\alpha < 0.17$. Above this value $x(r)$ is a linear function of r at low r. The ratio x/r has been plotted as a function of α in fig. 12. It clearly shows that the site binding is essentially complete for $\alpha > 0.3$ in excellent agreement with our experimental results. We have not tried to reinterpret the results of Westra and Leyte [10] which deal with very high values of r at low α, because, as already mentioned, the values of α are ill defined under these conditions and our eq. 8 is only valid up to $r = 0.4$. It is, however, interesting to recall that they find a complete binding above $\alpha = 0.6$ with $r < 0.4$.

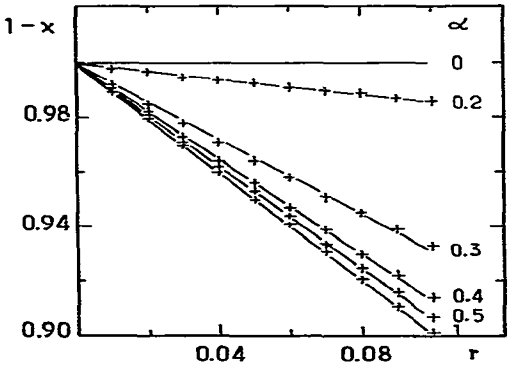

Fig. 11. Dependence of the site-bound fraction of polyion charges on the total fraction of divalent counterions r calculated for different α with $K = 6.4$ l mol^{-1}.

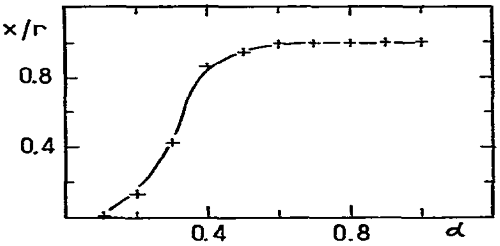

Fig. 12. Fraction of site-bound divalent ions as a function of α taken from fig. 11 for the r values of our experiments.

7. Comparison with the condensation model

In its initial formulation as a limiting law, the condensation model predicts a preferential condensation of the divalent counterions as soon as $\alpha\xi = 0.5$. This fits with the steep increase of CIV_2 above this value. In the case of a mixture of monovalent and divalent counterions, the condensation of the divalent counterion can be formally put in the form of a chemical equilibrium:

$$\frac{2\theta_2}{(r - 2\theta_2)} = K_2$$

where θ_2 is the total fraction of condensed counterions, i.e., site bound and territorially bound. But K_2 is itself a function of θ_2, since its calculation involves the interaction of the polyion, partially neutralized by the condensed counterions, with the uncondensed counterions. It involves also the value of the condensation volume V_p which is a function of the concentration C_1 of monovalent ions. The relevant expressions have been given by Wilson et al. [20]. For $\theta_2 < \frac{1}{2}(1 - \xi^{-1})$, the relationship between r and θ_2 can be written:

$$r = 2\theta_2\left[1 + 5.5C_1(\xi - 1)\xi^{-3}(1 - \xi^{-1} - 2\theta_2)^{-2}\right]$$

Our initial goal was to calculate θ_2 as a function of r and to try to extract from θ_2 the site-bound fraction using as a local concentration the fraction of territorially bound condensed counterions uniformly distributed in the condensation volume V_p.

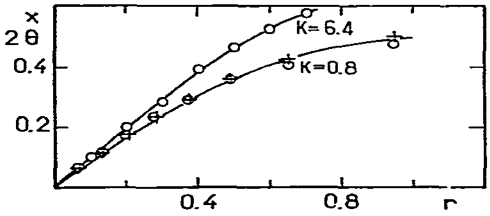

Fig. 13. Total fraction of condensed counterions $2\theta(+)$ calculated from Manning's model [7] and fraction x of site-bound charges on the polyion calculated with the Poisson-Boltzmann model for $K=6.4$ and $K=0.8$ l mol^{-1}.

The values of θ_2 as a function of r are given in fig. 13. They coincide with those predicted by the PB treatment for a value of $K=0.8$. Therefore, the predicted total fraction of condensed divalent counterions is always smaller than the experimental site-bound fraction. It seems that the condensation model is inedequate for the description of the details of the counterion distribution in the immediate vicinity of the polyion.

8. Discussion

The agreement between our calculation and our experimental determination of site binding at varying α and r leads to the conclusion that the local equilibrium hypothesis permits predicting the degree of site binding with the local concentration computed from the Poisson-Boltzmann equation at the surface of the charged cylinder. The value of K we have found is of the right order of magnitude for ion pair formation with monobasic acids. However, one can wonder about the use of the same value of K for the whole range of α, since the free energy of binding should depend on the change of hydration of both the counterion and polyion, and the structure of water near the latter should depend on the charge density. The flexibility offered by polyions to build systems with varying ionized groups and charge densities may be helpful for the study of binding constants which can be then transfered to micelles, vesicles and membranes.

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