These results show that the extent of branching for these star-shaped materials *increases* with reaction time and that the maximum extent of branching is not reached until the later stages of the reaction. In other words, the star formation process involves, at least in part, the addition of unlinked (or perhaps coupled) parent material to branched polymer. These observations are in accord with the divinylbenzene linking reaction characteristics discussed previously.

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References and Notes

(1) The Department of Chemistry. The University of Sheffield, Sheffield, S3 7HF, England.

(2) L.-K. Bi and L. J. Fetters, *Macromolecules*, **9**, 732 (1976).

(3) L. J. Fetters and L.-K. Bi, U.S. Patent, 3985830 (Oct. 12, 1976); British Patent 1502800 (Mar. 1, 1978).

(4) G. Quack and L. J. Fetters, *Polym. Prepr., Am. Chem. Soc., Div. Polym. Chem.*, **18**, 558 (1977).

(5) G. Quack, N. Hadjichristidis, and L. J. Fetters, to be published.

(6) R. Milkovich, Canadian Patent 716645 (Aug. 24, 1965).

(7) D. Decker and P. Rempp, *C. R. Hebd. Sceances Acad. Sci.*, **261**, 1977 (1965).

(8) J. G. Zilliox, D. Decker, and P. Rempp, *C. R. Hebd. Sceances Acad. Sci., Ser C*, **262**, 726 (1966).

(9) D. J. Worsfold, J. G. Zilliox, and P. Rempp, *Can. J. Chem.*, **47**, 3379 (1969).

(10) P. Rempp and E. Franta, *Pure Appl. Chem.*, **30**, 229 (1972).

(11) A. Kohler, J. G. Zilliox, P. Rempp, J. Polacek, and T. Koessler, *Eur. Polym. J.*, **8**, 627 (1972).

(12) J. G. Zilliox, P. Rempp, and J. Parrod, *J. Polym. Sci., Part C*, **22**, 145 (1968).

(13) K. Ziegler, *Angew. Chem.*, **49**, 499 (1936).

(14) M. Morton and L. J. Fetters, *Rubber Rev.*, **48**, 359 (1975).

(15) D. Rubenstein and J. R. Snyder, U. S. Patent 3217051 (Nov. 9, 1965).

(16) P. D. Meek and D. Rubenstein, U.S. Patent 3217052 (Nov. 9, 1965).

(17) D. Rubenstein and E. R. Winiarczyk, *Ind. Eng. Chem.*, **50**, 1553 (1968).

(18) R. H. Wiley, J. I. Jin, and Y. Kamath, *J. Polym. Sci. Part A*-1, **6**, 1065 (1968).

(19) D. J. Worsfold, *Macromolecules*, **3**, 514 (1970).

(20) A. Gourdenne and P. Sigwalt, *Eur. Polym. J.*, **3**, 481 (1967).

(21) J. E. L. Roovers and S. Bywater, *Trans. Faraday Soc.*, **62**, 701 (1966).

(22) G. V. Schulz, L. L. Böhm, M. Chmelir, G. Löhr, and B. J. Schmitt, "Kinetics and Mechanism of Polyreactions", IUPAC International Symposium on Macromolecular Chemistry, Akademiai Kido, Budapest, 1971, p 223; *Adv. Polym. Sci.*, **9**, 1 (1972).

(23) B. J. Schmitt, *Makromol. Chem.*, **156**, 243 (1972).

(24) J. Comyn and M. D. Glasse, *Makromol. Chem.*, **175**, 695 (1974).

(25) L. J. Fetters and E. Firer, *Polymer*, **18**, 306 (1977).

(26) F. L. McCrackin, *J. Appl. Polym. Sci.*, **21**, 191 (1977).

(27) M. R. Ambler, L. J. Fetters, and Y. Kesten, *J. Appl. Polym. Sci.*, **21**, 2439 (1977).

(28) D. J. Worsfold and S. Bywater, *Can. J. Chem.*, **38**, 1891 (1960).

(29) P. E. Black and D. J. Worsfold, *J. Appl. Polym. Sci.*, **14**, 1671 (1970).

(30) M. Morton and L. J. Fetters, *J. Polym. Sci., Part A2*, 3311 (1964).

(31) M. Morton, L. J. Fetters, R. A. Pett, and J. F. Meier, *Macromolecules*, **3**, 333 (1970).

(32) W. H. Glaze, J. E. Hanicak, M. L. Moore, and J. Chandhuri, *J. Organomet. Chem.*, **44**, 39 (1972).

(33) L. J. Fetters and M. Morton, *Macromolecules*, **7**, 552 (1974).

(34) M. M. Al-Jarrah and R. N. Young, to be published.

(35) In an indirect fashion, the results of Fraenkel (G. Fraenkel, W. E. Beckenbaugh, and P. P. Yang, *J. Am. Chem. Soc.*, **98**, 6878 (1976)) support the dimeric association state of the styryl and dienyllithium species.

(36) S. Bywater and D. Worsfold, *J. Organomet. Chem.*, **10**, 1 (1967).

(37) J. E. L. Roovers and S. Bywater, *Macromolecules*, **8**, 251 (1975).

(38) J. W. Burley and R. N. Young, *Chem. Commun.*, 991 (1970).

(39) H. Hirohara and N, Ise, *Macromol. Rev.*, **6**, 295 (1972).

Interactions of Small Ions with Heparin and Dextran Sulfate by Self-Diffusion Measurements

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ABSTRACT: Self-diffusion coefficients for Na^+, Ca^{2+}, Cl^-, and SO_4^{2-} were determined in aqueous solutions of sodium heparin and sodium dextran sulfate at 25 °C. Simple salt concentrations of 0.00050, 0.0010, 0.0050, and 0.010 N were used and the equivalent concentrations of the ionic polysaccharides were in the range of 0.000050 to 0.10 N. The sodium ion diffusion coefficients were found to decrease initially as X increased from zero to approximately six, X being the ratio of the equivalent concentration of ionic polysaccharide to the molar concentration of simple salt, followed by a leveling off as X increased. The calcium ion diffusion coefficients showed a greater initial decrease at low X values prior to its leveling off than did the values for sodium ions. For both ionic polysaccharides, the diffusion coefficients for Cl^- ion were close in value to those for SO_4^{2-} ion at corresponding X values. The experimentally obtained diffusion coefficients are correlated with those predicted from the Manning theory of polyelectrolyte solutions.

Ionic polysaccharides are of paramount importance because of their roles in biological phenomena. Their importance in the chemistry of connective tissue, of the eye, and of blood is evidenced by the growing number of publications in the field. A principal aspect of elucidating the behavior of ionic polysaccharides is a study of the interactions of small ions with these biological polyelectrolytes in aqueous salt solutions. Biological systems involving polyelectrolytes could be better understood with the aid of a good model and the model could act as a guide to further studies. The Manning theory[1-6] has proven most useful in our research endeavors because of its success in describing both transport and equilibrium properties.

Counterions and coion interactions with polyelectrolytes in aqueous salt solutions have been investigated by determining the tracer diffusion cofficients of the desired small ion. From studies on such polyelectrolytes as sodium poly(styrene sulfonate),[7] sodium polyacrylate,[8,9] sodium, calcium, strontium chondroitin sulfate,[10,11] sodium carboxymethylcellulose,[12] sodium alginate,[13] and sodium ι-

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carrageenan,[13] it has been shown that the counterions interact with the polyion to a greater degree than do the coions, as one might expect; divalent counterions interact to a greater extent than do the monovalent counterions. Also, the multivalent coions were found to interact with the polyions to approximately the same degree at very low simple salt concentration or at infinite dilution. These results have been correlated with the modern theory of polyelectrolytes by Manning.

With our continuing investigation of electrolytic properties of ionic polysaccharides, we determined the self-diffusion coefficients of Na^+, Ca^{2+}, Cl^-, and SO_4^{2-} ions in aqueous salt solutions containing sodium heparin (NaH) and sodium dextran sulfate (NaDS) over a large range of concentrations. It was desirous to compare the interactions of these ions with the ionic polysaccharides of similar linear charge densities, i.e., the average charge per unit length of chain. Sodium heparin has a tetrasaccharide as its basic unit.[14] It is thought to be a helix of 1 → 4 linked α-D-glucosamine, β-D-glucuronate, and iduronate containing three types of pendant ionic groups, *O*-sulfate, *N*-sulfate, and carboxyl, with an average distance between ionic groups of 0.24 nm, as determined from X-ray fiber diffraction studies.[15] Sodium dextran sulfate, a sulfonated derivative of dextran, was found to have an average of 2.07 sulfate groups per glycosyl residue. This gives an average distance between ionic groups of 0.25 nm.[16] Also, the Manning theory predicts that the interactions governing the tracer diffusion coefficients should be independent of the nature of the repeating monovalent charge groups in the polyion. While this was shown to be valid for polyelectrolytes containing one type of charge group, it was never tested with a polyelectrolyte such as NaH which contains three different charge groups incorporated in its repeating unit.

Sodium heparin and sodium dextran have antithrombotic activity, the former being more potent. Since both NaH and NaDS are polyelectrolytes, their antithrombotic action must be in some way due to the linear charge density of the polyelectrolyte and its interaction with the small ions about it. It should be of use to understand the electrolytic characteristics of ionic polysaccharides so as to help elucidate their biological functioning. One would, of course, want to know the degree of dissociation (and of binding) of small ions from the ionic polysaccharides.

Experimental Section

A sample of purified sodium heparin derived from hog mucosa was kindly provided in powder form by the Inolex Co. The equivalent weight of this sample was determined by titration to be 183.5 g/equiv.

A sample of purified dextran sulfate was kindly supplied by Pharmacia Fine Chemicals. Its experimental equivalent weight of 181 ± 1 g is in accord with an average of 2.07 sulfate groups per glucosyl residue, which is in excellent agreement with that found by Wells.[16] Hence, we assumed an average distance between charged groups of 0.25 nm, the value used by Wells. Prior to its use, a sample of dextran sulfate was dissolved in water and the phosphate buffer was removed by anion exchange. A portion of the purified sample was then converted to the acid form and titrated with standard NaOH to determine its concentration.

The radioactive salts $^{22}NaCl$ and $Na^{36}Cl$ were supplied by ICN Pharmaceuticals, Inc., and $Na_2{}^{35}SO_4$ and $^{45}CaCl_2$ were supplied by New England Nuclear Co. Stock solutions of radioactivity labeled salts were prepared by diluting 0.1 mCi of $Na^{36}Cl$ to 270 mL, 0.1 mCi of $^{22}NaCl$ to 100 mL, 1 mCi of $Na_2{}^{35}SO_4$ to 250 mL, and 1.0 mCi of $^{45}CaCl_2$ to 100 mL. The tracer diffusion coefficients of SO_4^{2-}, Cl^-, Na^+, and Ca^{2+} ions were determined in aqueous NaCl, Na_2SO_4, and $CaCl_2$ solutions as a function of X, the ratio of the normality of the polyelectrolyte, n_p, to the molarity of the simple salt, n_s, added. Diffusion coefficients were measured at four concentrations of each simple salt, 0.0100, 0.00500, 0.00100, and 0.000500 N with polymer concentrations ranging from 0.10 N to 0.000050 N; therefore X varied between 0.1 and 10.

The radioactive solutions were prepared according to the following procedure. One or two milliliters of the appropriate radioactive salt stock solution were placed in a 10-mL volumetric and evaporated in a vacuum oven. Then a radioactively inert solution of a given X value was added to the flask. If the amount of labeled salt was in excess of 0.3% of the total simple salt concentration of the solution appropriate adjustments were made on the concentration of the nonradioactive solution to make both radioactive and inert solutions of identical simple salt and polyelectrolyte concentration.

Self-diffusion measurements were performed in a constant temperature bath at 25.00 ± 0.01 °C by the open-end capillary method. Precision bore capillaries of 1.00 ± 0.005 mm diameter and 2.82 ± 0.005 cm length were filled with the solution containing a small quantity of radioactively labeled salt. After cleaning the outside surfaces of the filled capillaries, each capillary was placed into individual test tubes (100 mm × 13 mm) which were subsequently filled with the radioactive-inert solution of identical composition. Since both solutions inside and outside of the capillaries were equivalent in simple salt and polymer concentrations no concentration gradient was present. This permitted a study of the interaction of the simple ion with the polyelectrolyte by measuring the rate of self-diffusion of the labeled simple salt out of the capillary.

Self-diffusion coefficients were calculated from[17]

$$D = (\pi/4)(1 - C/C_0)^2 L^2/t \tag{1}$$

where t is the time elapsed during diffusion in seconds, C_0 is the radioactive content of the capillary at time zero (prior to diffusion), C is the radioactive content at time t, and L corresponds to the length of the capillary in centimeters. The validity of this equation for values of C/C_0 greater than 0.5 was demonstrated. After sufficient time was allowed for diffusion the outer solution was withdrawn with an aspirator and the capillary was removed from the test tube. The outsides of the capillaries were washed, dried, and placed open-end down into a vial containing 5 mL of scintillation fluid (Insta Gel, Packard Instrument Co.). In order to remove the radioactive solution from the capillary and to ensure thorough mixing with the scintillation fluid, each vial was centrifuged for 5 min, followed by shaking for 30 s on a Vortex Genie, Scientific Industries, Inc. The radioactive content of each capillary, C, was determined by using a Packard Tri Carb liquid scintillation spectrometer Model 3385. The initial radioactive content of the capillaries, C_0, was determined by using capillaries of identical bore and length by the same procedure; however, no diffusion was allowed to occur. Each capillary was counted to a minimum of 15000 counts, which results in an error of 0.8%. The ratio C/C_0 was kept between 0.6 and 0.7 in order to minimize the error in the experimental results. Linearity between the radioactive concentrations in the capillaries and the counts was demonstrated.

Each reported value of a diffusion coefficient was based on at least six individual determinations of C and at least five values of C_0. The average standard error of each of the reported diffusion coefficients was found to be ±3%.

Results and Discussion

Tracer diffusion coefficients for Na^+, Ca^{2+}, Cl^-, and SO_4^{2-} were determined at 25 °C. These are shown in Figures 1–8 as D_i/D_i^0, where D_i and D_i^0 are the tracer diffusion coefficients of ion i in the presence and absence of polyelectrolyte, respectively, as a function of X, where $X = n_p/n_s$ with n_p the equivalent concentration of polyelectrolyte and n_s the molar concentration of simple salt added. (Previous papers of ours have defined X as the equivalent concentration of polyelectrolyte to the equivalent concentration of simple salt added.) The values used for D_i^0 are 1.33×10^{-5} for Na^+, 0.792×10^{-5} for Ca^{2+}, 2.03×10^{-5} for Cl^-, and 1.06×10^{-5} cm^2/s for SO_4^{2-}.[18,19]

Comparing $D_{Na^+}/D_{Na^+}^0$ values for NaH and NaDS, it is noted from Figures 1 and 2 that the Na^+ ion diffusion

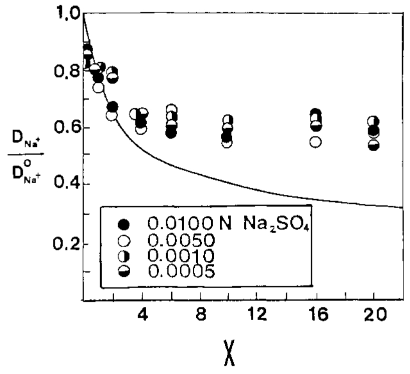

Figure 1. The dependence of the self-diffusion ratio for Na^+ ions on X in NaH solutions. The solid line is predicted from the Manning theory.

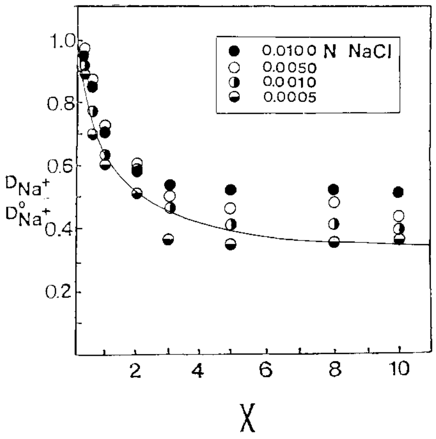

Figure 2. The dependence of the self-diffusion ratio for Na^+ ions on X in NaDS solutions. The solid line is predicted from the Manning theory.

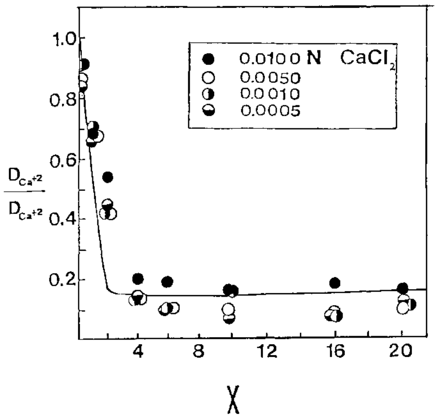

Figure 3. The dependence of the self-diffusion ratio for Ca^{2+} ions on X in NaH solutions. The solid line is predicted from the Manning theory.

ratios decrease initially as X increases and level off at approximately $X > 4$ with diffusion ratios of 0.6 and 0.4 for NaH and NaDS, respectively. While D_{Na^+} was determined with Cl^- as the coion in NaDS solutions and with SO_4^{2-} as the coion in NaH solutions, it was shown[7] that $D_{Na^+}/D_{Na^+}{}^0$ for sodium poly(styrene sulfonate) solutions was independent of the valence of the coion for Cl^-, SO_4^{2-}, and $Fe(CN)_6^{4-}$, when X was less than 12.

The Manning model for the polyelectrolyte is an infinite line charge with a stoichiometric linear charge density ξ given by

$$\xi = e^2/\epsilon kTb \qquad (2)$$

where e is the protonic charge, ϵ is the dielectric constant

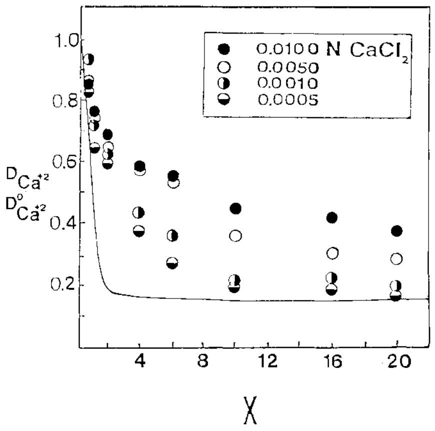

Figure 4. The dependence of the self-diffusion ratio for Ca^{2+} ions on X in NaDS solutions. The solid line is predicted from the Manning theory.

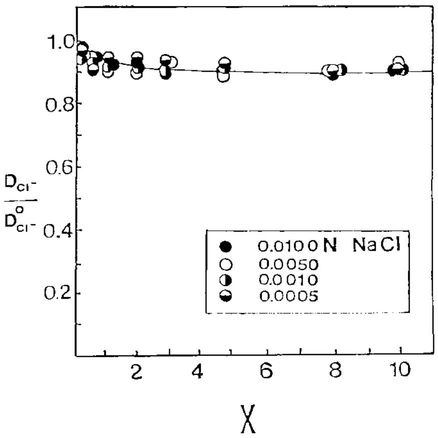

Figure 5. The dependence of the self-diffusion ratio for Cl^- ions on X in NaH solutions. The solid line is predicted from the Manning theory.

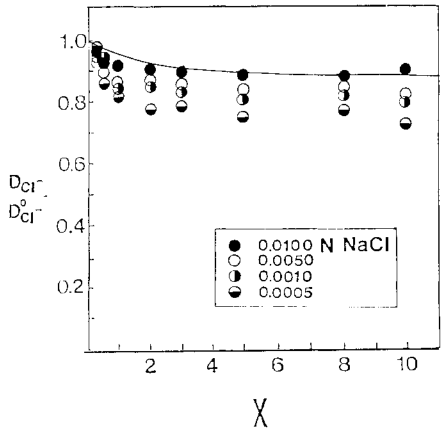

Figure 6. The dependence of the self-diffusion ratio for Cl^- ions on X in NaDS solutions. The solid line is predicted from the Manning theory.

of the solvent, T is the temperature, b is the average distance between charge groups, and k is the Boltzmann constant. If ξ is greater than its critical value ξ_c of $|z_1|^{-1}$, where z_1 is the valence of the counterion, counterion condensation occurs so as to reduce ξ to its effective value ξ_c. When the simple salt and the polyelectrolyte have a *common counterion*, the concentration of the dissociated counterions is $(\xi z_1)^{-1}n_p$, which together with the counterions and the coions of the simple salt interact with the polyion of Debye–Hückel forces. If $\xi < \xi_c$, no counterion condensation occurs and all small ions interact with the

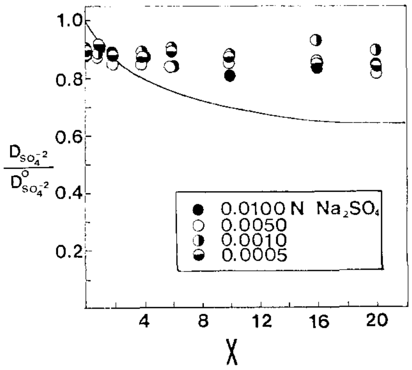

Figure 7. The dependence of the self-diffusion ratio for SO_4^{2-} ions on X in NaH solutions. The solid line is predicted from the Manning theory.

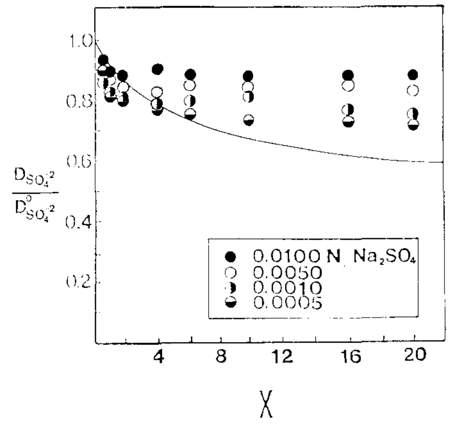

Figure 8. The dependence of the self-diffusion ratio for SO_4^{2-} ions on X in NaDS solutions. The solid line is predicted from the Manning theory.

fully charged polyion by Debye–Hückel forces.

For the ionic polysaccharides studied here, the ξ values are 3.0 and 2.85 for NaH and NaDS, respectively. If the self-diffusion coefficients of the condensed counterions are much smaller than for the free ones, the theoretical equation for Na^+ ions is

$$\frac{D_{Na^+}}{D_{Na^+}{}^0} = \frac{(\xi^{-1}X + 1)}{(X + 1)}\left[1 - \frac{A_1}{3}\right] \tag{3}$$

where the lead term is the contribution due to counterion condensation and A_1 is given by

$$A_1 = \sum_{(m,n\neq 0)} [\pi(m^2 + n^2) + |z_1| + (|z_1| + |z_2|)\xi X^{-1}]^{-2} \tag{4}$$

where z_1 and z_2 are the counterion and coion charges, respectively, and A_1 is the contribution of the Debye–Hückel interactions of all uncondensed small ions with the polyion, which is a function of ξ_c and the effective value of X. Since eq 2 is a limiting law, it is more fitting to correlate the theoretical values with the experimental ones at the lowest simple salt concentration or at an average value if the points at an X value are bunched together. With this in mind, the theoretical line in Figure 2 for NaDS is in excellent agreement with the experimental points over the whole X range. In Figure 1, the theoretical line for NaH is in excellent accord with the experimental points for $X < 3$, but for $X > 3$ the experimental points are constant with $D_{Na^+}/D_{Na^+}{}^0$ approximating 0.6, while the theoretical line decreases continuously in the X range studied. This may be due to the contribution of the condensed Na^+ ions to D_{Na^+} because of its lower molecular weight (<30 000)[20] as compared to that of NaDS (500 000). This problem is currently under investigation. It is interesting that a recent ^{23}Na NMR study of the binding of Na^+ ion to heparin gave results that were entirely consistent with the Manning model.[21]

In Figures 3 and 4 it is seen that for NaH and NaDS, respectively, $D_{Ca^{2+}}/D_{Ca^{2+}}{}^0$ initially decreases more rapidly at low X than was evident for $D_{Na^+}/D_{Na^+}{}^0$. The $D_{Ca^{2+}}/D_{Ca^{2+}}{}^0$ ratios achieve constant values of 0.1 and 0.2 for NaH and NaDS, respectively, for approximately $X > 5$ at the lowest simple salt concentration. This is expected since divalent Ca^{2+} would be expected to bind to the polyion to a greater extent than monovalent ions.

Upon adding Ca^{2+} ions to solutions containing the sodium salts of heparin and of dextran sulfate, the composition of the solutions changes. Solutions of polyelectrolytes containing monovalent and divalent counterions have been discussed.[6,11,22] Simply, in salt-free solutions, NaH and NaDS have an effective $\xi_c = 1$ and a charge fraction, i.e., the fraction of structural polyelectrolyte charge uncompensated by associated counterions, of $(z_1\xi)^{-1}$. Gradually adding Ca^{2+} ion to the polyelectrolyte solutions at a constant effective $\xi_c = 1$ results in a continuous replacement of the condensed monovalent Na^+ ions by divalent Ca^{2+} ions. When all the condensed Na^+ ions are replaced by the added Ca^{2+} ions the effective ξ_c value becomes $\xi_c = z_1^{-1} = {}^1/_2$ and further addition of Ca^{2+} ions results in condensation of Ca^{2+} ions onto the polyion until this effective ξ value is reached. At this critical point the value of X is $4\xi/(2\xi - 1)$ and the charge fraction of the polyelectrolyte is $(\xi z_1)^{-1}$ or $\xi^{-1}/2$. Further addition of Ca^{2+} ions results only in uncondensed Ca^{2+}, Na^+, and coions interacting with the polyion by Debye–Hückel forces. At X values lower than this critical point of $4\xi/(2\xi - 1)$, $D_{Ca^{2+}}$ is that of the uncondensed Ca^{2+} ions, while above this value the diffusion coefficient is constant and equal to that of the salt-free calcium polyelectrolyte polyion itself, since all the Ca^{2+} ions are assumed to be condensed on it. For $X < 4\xi/(2\xi - 1)$, Manning's theoretical relationship is[10]

$$D_{Ca^{2+}}/D_{Ca^{2+}}{}^0 = (1 - X/2 + \xi^{-1}X/4)(1 - 4A_2/3) \tag{5}$$

where

$$A_2 = \sum_{(m,n\neq 0)}\sum [2\pi(m^2 + n^2) + 2 - 2\xi + 12\xi X^{-1}]^{-2} \tag{6}$$

The series in eq 6 converges rapidly and contributes insignificantly to eq 3 and the values for $D_{Ca^{2+}}/D_{Ca^{2+}}{}^0$ are determined by the condensation term. Comparing the theoretical values, given by the lines in Figures 3 and 4, with the experimental points, good agreement is evident for NaH over the whole range of X and for NaDS at low and high X values. (Recall that the lowest salt concentration points should be compared with theory if an ionic strength dependence at a given X value is present.) This good agreement is, of course, due to the important condensation contribution of divalent counterions and the large charge densities of NaH and NaDS. Just above and below the critical X value (2.4) for NaDS there is poor accord with the theory. Such was also noted for chondroitin sulfate ($\xi = 1.40$),[11] which seems to indicate that upon adding Ca^{2+} ions at this transition point, the Ca^{2+} ion condensation is incomplete until a sufficient excess is added. The study with chondroitin sulfate showed that as the simple electrolyte concentration is increased at constant polyelectrolyte concentration, the ion self-diffusion coefficient approaches its value in the absence of chondroitin sulfate and that Ca^{2+} and Sr^{2+} ions gave similar results. This coupled with the good agreement with the theoretical values at high electrolyte concentration gives credibility to the Manning theory for counterion–polyelectrolyte interaction. This seems to indicate that

Ca^{2+} ions do not bind to heparin in a highly specific manner. The results of investigations of divalent counterion binding to heparin by equilibrium dialysis[23,24] and by circular dichroism[25] indicate a similar type of binding for Mg^{2+}, Ca^{2+}, and Ba^{2+} ions and a highly specific binding for Cu^{2+} ions.

Since the coions can interact with the polyion only by Debye–Hückel forces, the Manning theory for the coions gives

$$D_2/D_2^0 = 1 - z_2^2 A_2/3 \qquad (7)$$

where the subscript 2 refers to the coions. The experimental $D_{Cl^-}/D_{Cl^-}^0$ points for NaH and NaDS are shown in Figures 5 and 6, respectively, along with the lines calculated using eq 6. For the NaH solutions, the $D_{Cl^-}/D_{Cl^-}^0$ experimental points appear to be fairly independent of X and of simple salt concentration. The excellent agreement with the theoretical model is evident. For the NaDS solutions, the experimental $D_{Cl^-}/D_{Cl^-}^0$ points in Figure 6 decrease from $X = 0$ to $X = 1$, followed by a leveling off for $X > 1$. An ionic strength dependence of $D_{Cl^-}/D_{Cl^-}^0$ is noted and upon comparing the $D_{Cl^-}/D_{Cl^-}^0$ at the lowest simple salt concentration with the theoretical line, agreement is not good. It is interesting that the experimental points for $D_{SO_4^{2-}}/D_{SO_4^{2-}}^0$ for NaH and NaDS which are shown in Figures 7 and 8, respectively, are close in value to their corresponding points for $D_{Cl^-}/D_{Cl^-}^0$. Similar findings have been reported for sodium poly(styrene sulfonate),[7] sodium ι-carrageenan,[13] and sodium alginate.[13] It appears that the Cl^- and SO_4^{2-} ions interact with each polyelectrolyte to the same extent. It is evident from Figures 7 and 8 that the diffusion ratios for SO_4^{2-} ions do not appear to approach unity as X tends to zero. This also is evident for SO_4^{2-} and $Fe(CN)_6^{4-}$ ions in sodium carrageenan and sodium alginate solutions,[13] but it was not found for these coions in sodium poly(styrene sulfonate) solutions at low simple salt concentrations. It is tempting to attribute this to greater experimental error at X less than unity because of low polyelectrolyte concentration. Because this region is of particular importance theoretically, experiments are in progress to evaluate the initial slopes of the curves. The Manning theory appears to be a good representation for monovalent–polyelectrolyte interactions but overemphasizes the interactions of multivalent coions with the polyelectrolyte.

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References and Notes

(1) G. S. Manning, *Annu. Rev. Phys. Chem.*, **23**, 117 (1972).
(2) G. S. Manning, *Biophys. Chem.*, **7**, 95 (1977).
(3) G. S. Manning, *J. Chem. Phys.*, **51**, 924, 934 (1969).
(4) G. S. Manning, *J. Phys. Chem.*, **79**, 262 (1975).
(5) G. S. Manning and D. I. Devore, *J. Phys. Chem.*, **78**, 1242 (1974).
(6) G. S. Manning, "Polyelectrolytes", E. Selegny, Ed., D. Reidel Publishing Co., Dordrecht, Holland, 1974.
(7) M. Kowblansky and P. Ander, *J. Phys. Chem.*, **80**, 297 (1976).
(8) D. Dixler and P. Ander, *J. Phys. Chem.*, **77**, 2684 (1973).
(9) S. Menezes-Affonso and P. Ander, *J. Phys. Chem.*, **78**, 1756 (1974).
(10) H. Magdelenat, P. Turq, M. Chemla, and B. Para, *Biopolymers*, **15**, 175 (1976).
(11) H. Magdelenat, P. Turq, M. Chemla, and B. Para, *Biopolymers*, **13**, 1535 (1974).
(12) M. Milas, Ph.D. Thesis, University of Grenoble, Grenoble, France, 1974.
(13) A. Kowblansky, R. Sasso, V. Spagnuola, and P. Ander, *Macromolecules*, **10**, 78 (1977).
(14) M. E. Silva and C. P. Dietrich, *J. Biol. Chem.*, **250**, 6841 (1975).
(15) E. D. Atkins and I. A. Nieduszynski in "Advances in Experimental Medicine and Biology", R. A. Bradshaw and S. Wessler, Ed., Plenum Press, New York, N.Y., 1974.
(16) J. D. Wells, *Proc. R. Soc. London, Ser B*, **183**, 399 (1973).
(17) J. C. Anderson and K. Addington, *J. Chem. Soc.*, S381 (1949).
(18) P. Turq, Ph.D. Thesis, University of Paris, Paris, France, 1970.
(19) R. A. Robinson and R. H. Stokes, "Electrolytic Solutions", 2nd ed, Butterworths, London, 1959.
(20) S. Stivala, *Fed. Proc., Fed. Am. Soc. Exp. Biol.*, **32**, 561 (1973).
(21) L. Herwats, P. Laszlo, and P. Genard, *Nouv. J. Chim.*, **1**, 173 (1977).
(22) J. C. T. Kwak, N. J. Morrison, E. J. Spiro, and K. Iwasa, *J. Phys. Chem.*, **80**, 2753 (1976).
(23) B. Lagos and S. S. Stivala, *Biopolymers*, **12**, 127, 961 (1973).
(24) S. S. Stivala, *Fed. Proc., Fed. Am. Soc. Exp. Biol.*, **36**, 83 (1977).
(25) M. C. M. Chung and N. F. Ellerton, *Biopolymers*, **15**, 1409 (1976).

Calculation of Three-Dimensional Elastic Constants of Polymer Crystals. 1. Method of Calculation

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ABSTRACT: A new set of equations for calculating three-dimensional elastic constants of polymer crystals has been derived by using the **B** matrix in the normal coordinate treatments. Contrary to the methods by previous workers which need the parameters concerning all atoms in one unit cell, the number of parameters in the present method is reduced to that necessary to describe one crystallographic asymmetric unit by employing the space group symmetry of the crystal. This saves a great amount of memory size of computer and therefore becomes applicable to the cases with complicated crystal structure. Derivation of the equations has been illustrated by the case of orthorhombic polyethylene.

The crystallite moduli and compressibilities have been measured by many investigators for a wide variety of crystalline polymers.[1-3] It is very important to interpret these experimental results theoretically from the standpoint of the crystal structures determined by X-ray analysis and to clarify the relationship among crystal structures, intra- and intermolecular forces, and mechanical properties.

In the polymer crystal the chain molecules, the atoms which are linked tightly by the covalent bonds, are as-

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