

# POLYELECTROLYTES

A. KATCHALSKY

*Weizmann Institute of Science, Rehovot, Israel*

## ABSTRACT

A general review of the behaviour of polyelectrolyte systems is presented. It deals primarily with the colligative properties of aqueous solutions of polyelectrolytes in salt-free and salt-containing mixtures, as well as with several transport phenomena (such as conductance, transference and diffusion). The theoretical model underlying this account is that of charged macromolecules surrounded by a dense cylindrical counterion atmosphere. This model proves a quantitative description of osmotic pressure, of activity coefficients, of the heat of dilution and of Donnan distribution between polyelectrolyte solutions (or gels) and an external salt solution.

For the analysis of transport phenomena a non-equilibrium thermodynamic treatment was introduced which allows the identification of straight and coupling phenomena in polyelectrolyte systems. This analysis led to an explicit consideration of relaxation and electrophoretic effects in polyelectrolytes.

Some attention was paid to the biophysical significance of polyelectrolyte interactions, and in particular to the combination of polyelectrolytes with divalent ions and to the complex formation of polyelectrolytes with other charged polyions. These interactions play an important role in the organization of nucleic acids and in their combination with basic proteins. Since the double- or triple-stranded complexes of some polynucleotides exhibit hysteresis—related to the conformation of the complex—the last part of the paper is devoted to the discussion of molecular hysteresis and its significance as a biomolecular memory imprint.

---

## 1. INTRODUCTION

1.1 Polyelectrolyte research is concerned with the study of the electrochemical properties of charged polymers; it may be regarded as a direct descendant of the classical colloid chemistry of hydrophilic biocolloids. It is therefore both an honour and a pleasure to present a lecture at this Mecca of colloid chemistry, where major advances have been made in the quantitative evaluation of equilibrium and non-equilibrium properties of colloidal systems. The Dutch school has been for many years a leading centre, in the study of electrical double layers and in the elucidation of colloid stability. It is this school which introduced rigorous thermodynamic methods into the phenomenological description of equilibria, and employed non-equilibrium thermodynamics in the treatment of colloid transport phenomena. Here, complexes of natural polyelectrolytes have been extensively investigated and their significance in biological organization brought to the attention of the world of science<sup>1</sup>. Hence whatever will be said during this

lecture owes an inestimable debt to the ideas and to the achievements of our host country.

1.2 The transition from colloid chemistry to the present day study of polyelectrolytes may be attributed to the pioneering observations of Staudinger's school—and in particular to the work of Kern—on the behaviour of synthetic polyacids. These man-made polyelectrolytes provided an opportunity to study colloidal particles having a stable polymeric backbone of known molecular weight and the ionizable groups of which could be varied at will. During the last two decades, the investigation of synthetic polyelectrolytes grew in importance, both from an applied point of view (as the basis for ion exchangers and permselective membranes) and as convenient models for the theoretical interpretation of the behaviour of biocolloids.

For many years the students of biological macromolecules were engrossed in the elucidation of biopolymer structure and could not pay attention to their electrochemical properties. Recently there is, however, increasing interest in the role of the electrical properties of the nucleic acids, of the polyuronic acids and of certain proteins. It is gradually being realized that many organizational and regulatory properties of the biopolymers are due to their electrical influence on their neighbourhood, so that polyelectrolyte research begins to be significant also in molecular and cellular biology.

1.3 Historically, some of the demonstrable properties of polyelectrolytes were discovered by measuring their specific viscosity and its dependence on polymer concentration, on the degree of ionization and on ionic strength. The data indicated that polyelectrolyte molecules are endowed with a powerful electrical field which suffices to overcome the Brownian randomization of intramolecular movements and which at low concentrations and high charge densities, may stretch appreciably the macromolecular chains. Corroborative evidence for the high potentials and strong fields of polyelectrolytes was obtained by analysing the results of pH titrations and electrophoretic mobility. A fuller discussion of these interesting findings requires, however, more sophisticated mastery of the topic of polyelectrolyte behaviour and will not be considered in this lecture. Since we are not bound to a *chronological* treatment, we shall instead follow a *logical* order and start our discussion with the consideration of the equilibrium properties of the binary system of salt-free polyelectrolyte in aqueous solution<sup>2</sup>.

## 2. COLLIGATIVE PROPERTIES OF SALT-FREE POLYELECTROLYTE SOLUTIONS<sup>3</sup>

### 2.1 Osmotic pressure coefficient of polyelectrolyte solutions

Consider a solution of  $N_p$  polyelectrolyte molecules, of a degree of polymerization  $P$ , dissolved in  $V$  ml water so that the molecular concentration of the polymers is

$$n_p = N_p/V \quad (1)$$

and its monomolar concentration

$$n_m = n_p P \quad (2)$$

POLYELECTROLYTES

Let each of the monomeric units carry an ionizable group, such as the carboxylic groups of a polyacrylic or a polymethacrylic acid, and let  $\nu$  of the  $P$  ionizable groups be ionized so that the degree of ionization is

$$\alpha = \nu/P \quad (3)$$

If the ionized groups are neutralized by monovalent counterions the concentration of the counterions is

$$n_c = n_p \nu = n_m \alpha \quad (4)$$

From a theoretical point of view, the simplest property of the solution is the osmotic pressure ( $\pi$ ) and were the polyelectrolyte solution to obey ideal laws, we would expect the osmotic pressure to be due to that of the polymeric and counterion contribution, i.e.

$$\pi_{\text{ideal}} = (n_p + n_p \nu) kT = n_m (1/P + \alpha) kT \quad (5)$$

For high molecular polymers and non-vanishing degrees of ionization  $\alpha \gg 1/P$ , hence

$$\pi_{\text{ideal}} \simeq \alpha n_m kT \quad (6)$$

In reality, polyelectrolyte solutions are strongly non-ideal, so that it is convenient to introduce an osmotic coefficient  $\phi_p$  to account for the deviation of the counterion contribution from ideality. We therefore write

$$\pi = (n_p + n_p \nu \phi_p) kT = n_m (1/P + \alpha \phi_p) kT \quad (7)$$

Although  $\phi_p$  is always smaller than unity,  $\phi_p \alpha$  is generally much larger than  $1/P$  so that usually

$$\pi \simeq \alpha \phi_p n_m kT \quad (8)$$

Comparing equations 6 and 8 we observe that the osmotic coefficient is the ratio of real to ideal osmotic pressures, or

$$\phi_p = \pi/\pi_{\text{ideal}} \quad (9)$$

It is often found that at higher degrees of ionization  $\alpha \phi_p$  is approximately constant, or that the osmotic coefficient decreases to the same extent that the degree of ionization increases. This fact can be deduced also from theoretical considerations and hence the product  $\alpha \phi_p$  deserves a special denotation. Since it represents the fraction of the counterions which contributes to the osmotic pressure, it is generally known as the 'effective' degree of ionization  $i$ , or

$$i = \alpha \phi_p \quad (10)$$

Inserting equation 10 into 7 or 8 we obtain

$$\pi = n_m (1/P + i) kT \quad \text{or} \quad \pi \simeq n_m i kT \quad (11)$$

The relative constancy of the effective degree of ionization is significant from a biophysical point of view. Since metabolic processes produce or take up protons, it might be expected that the osmotic pressure of biological media would undergo strong fluctuations. The protons bind, however, to

natural polyelectrolytes and change their degree of ionization—changing simultaneously the osmotic coefficient of the counterions. As the ‘effective’ degree of ionization remains practically constant the osmotic pressure will not change, or, the solution will behave as ‘osmotically buffered’.

2.2 The osmotic pressure of various polyelectrolyte solutions has been determined by many classical methods. Since the concentration of the counterions is high, it is possible to use cryoscopic methods, regular osmometry or isopiestic equilibration of a polyelectrolyte solution with solutions of known vapour pressure<sup>4</sup>. From the results thus obtained on various polyacids—such as polyacrylic and polymethacrylic acids, on polyphosphates and polysulphonates, on carboxymethyl cellulose and the alginates—a well defined pattern emerges which can be summarized in a few sentences:

(a) It is generally found that the values of  $\phi_p$  are by and large determined by the linear charge density of the polyelectrolyte and do not vary appreciably with the chemical nature of the polymeric chain.

Stiffer polymers are characterized, however, by larger  $\phi_p$ s than the more flexible polyelectrolytes.

(b) The osmotic coefficients for sufficiently high polymers—of a degree of polymerization say higher than fifty—are independent of the molecular weight and are fully determined by the local electrochemical properties.

(c) It is rather remarkable that the dilution of a polyelectrolyte solution does not lead to a stronger ‘dissociation’ of the counterions and does not increase the osmotic coefficient. Often the opposite effect is observed—namely upon diluting the solution, a decrease in  $\phi_p$  is recorded, indicating a ‘stronger binding’ of the counterions to the polyion, with a concomitant diminution in their osmotic activity.

(d) The nature of the ‘non-specific’ counterions, such as  $\text{Li}^+$ ,  $\text{Na}^+$  or  $\text{K}^+$ , has generally little effect on the osmotic activity—although the bulkier ions such as tetra-alkyl ammonium increase significantly the value of  $\phi_p$ .

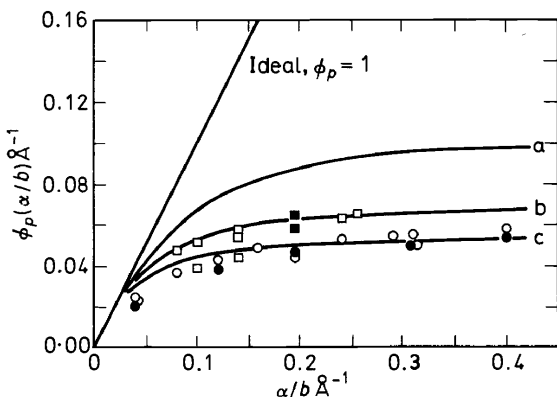


Figure 1. The osmotic coefficient  $\phi_p$  of salt-free solutions, multiplied by reciprocal interchange distance  $(\alpha/b)$ , versus the latter. (—) values calculated from equation 60 using (a)  $\lambda_{\text{effective}} = \lambda_{\text{structural}}$ ; (b)  $\lambda_{\text{effective}} = 1.5 \lambda_{\text{structural}}$ ; (c)  $\lambda_{\text{effective}} = 2 \lambda_{\text{structural}}$ . Experimental results for (○) polyacrylates and phosphate; (●) polymethacrylates; (□) carboxymethyl cellulose; (■) alginates. (From ref. 3, Figure 1)

(e) Finally, as stated before, increasing the degree of ionization ( $\alpha$ ) decreases the osmotic coefficient. It is more convenient to compare different polyelectrolytes if the linear charge densities are considered instead of the degrees of ionization. Denoting the distance between *ionizable groups* by  $b$ , the real distance between *charged groups* is  $b/\alpha$ , and the linear, numerical charge density is  $\alpha/b$ . A plausible effective ionization is now  $\phi_p(\alpha/b)$  instead of  $i = \phi_p\alpha$ ; the dependence of  $\phi_p(\alpha/b)$  on the charge density  $\alpha/b$  is shown in *Figure 1*.

2.3 The fundamental importance of the osmotic coefficients of polyelectrolyte solutions, as well as the difficulty involved in the direct measurement of osmotic pressures, led several workers to the search for an alternative determination of  $\phi_p$ . The method of choice was light scattering, which is widely used in polymer chemistry<sup>5</sup>. It is known that Einstein's theory of fluctuations led to a relation between the turbidity ratio at  $90^\circ$ ,  $R^{90}$ , and the change in the chemical potential of the solute with concentration. In our case this relation is

$$\partial\mu_p/\partial n_m = K'/R^{90} \quad (12)$$

where  $\mu_p$  is the chemical potential of the polyelectrolyte and  $K'$  is a characteristic constant given by

$$K' = (2\pi^2 n_0^2/N_A \lambda^4) (\partial n/\partial n_m)^2 \quad (13)$$

( $n_0$  is the refractive index of the solute and  $n$  the refractive index of the solution;  $N_A$  is Avogadro's number and  $\lambda$ , the wavelength of the incident light.) In equation 13,  $\partial n/\partial n_m$  is assumed to be practically constant. Now by the equation of Gibbs and Duhem

$$n_m \partial\mu_p/\partial n_m + n_w \partial\mu_w/\partial n_m = 0 \quad (14)$$

where  $n_w$  is the number of molecules of solvent per unit volume and  $\mu_w$  the chemical potential of the solvent (in our case **always** water). Further the thermodynamic relation between osmotic pressure and the chemical potential of the solvent is

$$\partial\mu_w/\partial n_m = -\bar{V}_w \partial\pi/\partial n_m \quad (15)$$

where  $\bar{V}_w$  is the partial molecular volume. Inserting 15 into 14 we obtain

$$n_m \partial\mu_p/\partial n_m = n_w \bar{V}_w \partial\pi/\partial n_m \quad (16)$$

A unit volume of a binary solution is related to the solute and solvent concentrations by  $n_m \bar{V}_m + n_w \bar{V}_w = 1$ , but since we deal with dilute solutions for which  $n_w \bar{V}_w \gg n_m \bar{V}_m$ , we may write  $n_w \bar{V}_w \simeq 1$  and hence  $\partial\mu_p/\partial n_m$  in equation 12 is practically equal to

$$\partial\mu_p/\partial n_m = (1/n_m) (\partial\pi/\partial n_m) \quad (17)$$

Upon differentiating equation 7 and assuming that  $\phi_p$  is only slightly dependent on  $n_m$ , we obtain from equation 12

$$\partial\mu_p/\partial n_m = [\{(1/P) + \alpha\phi_p\}/n_m]kT$$

or

$$\frac{1}{P} + \alpha\phi_p = \frac{K'n_m}{kTR^{90}} = \frac{Kn_m}{R^{90}} \quad (18)$$

Thus for given  $K$ , the measurement of  $R^{90}$ , at known  $n_m$ , suffices for the evaluation of  $\phi_p$ .

The first experimental results were, however, discouraging, since the values of  $R^{90}$  were many times larger than expected. A closer inspection of equation 18 clarifies the source of the experimental difficulties: for very low degrees of ionization  $\alpha \rightarrow 0$  the polyelectrolyte behaves as an uncharged polymer  $Kn_m/R^{90} \simeq 1/P$  and for high degrees of polymerization large turbidity ratios are obtained. This is of course the basis for the turbidimetric determination of polymer molecular weights. On the other hand, at finite degrees of ionization, when  $\alpha\phi_p \gg 1/P$ , the turbidity decreases appreciably and becomes independent of the degree of polymerization. Ultimately,  $Kn_m/R^{90} \rightarrow \alpha\phi_p = i$ , so that light scattering becomes a direct measure for the effective degree of ionization; but since  $i$  is of the order of magnitude of 0.1 while for synthetic polyelectrolytes  $1/P$  is  $\sim 0.001$  (and for biopolymers may be still lower) it is clear that the turbidity of polyelectrolytes is many hundred times smaller than that of uncharged polymers—and hence only a

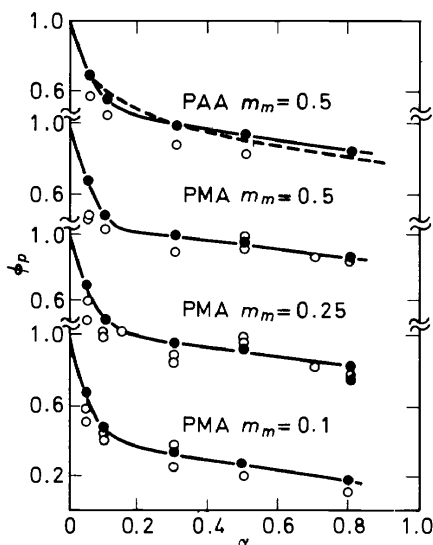


Figure 2. Osmotic coefficient  $\phi_p$  as a function of  $\alpha$ . Results are given for PMA at three monomolal concentrations,  $m_m = 0.5, 0.25$  and  $0.1$ , and for PAA at  $m_m = 0.5$ . (○) values determined by light scattering; (●) values determined by the 'osmotic concentration' method (the solid line is drawn through the latter, while the broken line represents osmotic results. (From ref. 3, Figure 2)

painstaking optical purification of the solution will provide reliable results. Such a purification, including careful sterilization, was carried out by Alexandrowicz<sup>6</sup>, who finally obtained results in good agreement with direct osmotic measurements. These are given in Figure 2.

### 3. THE CELL MODEL AND THE COLLIGATIVE PROPERTIES OF POLYELECTROLYTE SOLUTIONS<sup>7</sup>

3.1 Aqueous solutions of polyelectrolytes resemble mixtures of low molecular multivalent electrolytes with water, and it is tempting to treat their behaviour by the classical method of Debye and Hückel. The fact, however, that there is a pronounced asymmetry between the behaviour of the polyions and the low molecular counter- and co-ions, makes the simple model of spherical ionic atmospheres inapplicable to the present case. In polyelectrolyte solutions, the charged macromolecule is always the centre of its own atmosphere and cannot be assumed to participate in the atmosphere of other ions. It is therefore advantageous to divide the solution *a priori* into 'cells', the centres of which are the polymeric ions, each one surrounded by an atmosphere of small ions. This cell model of statistical mechanics is well known in colloid chemistry, but should not be used for the description of low molecular weight electrolytes where it may lead to erroneous results.

For the sake of concreteness it is assumed that the electrostatic repulsion between the polyions leads to a homogeneous distribution of the macromolecular centres so that an average interpolymer distance  $2R$  may be attributed to the cell structure. Moreover, there is good evidence that the macromolecules are sufficiently stretched by electrostatic repulsion as to be ascribed a cylindrical symmetry, at least on a segmental scale\*. Finally, following the consideration of Onsager<sup>8</sup> the state of minimum energy is that of an array of parallel rods, so that the solution cells are taken to be parallel cylindrical units of a height  $h$ —equal to that of the average, partially stretched, length of the polyions and a cross section of  $R^2$ . For a solution containing  $n_p$  macromolecules per unit volume

$$n_p \pi R^2 h = 1 \quad (19)$$

If the average cross section of a macromolecule is  $\pi a^2$  the volume occupied

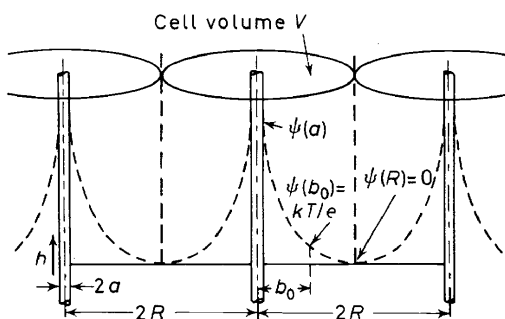


Figure 3. A schematic drawing to illustrate the cell model for polyelectrolytes. (From ref. 3, Figure 4)

\* NOTE: In this paper we shall consider only the cylindrical model, which seems at present to be favoured by most workers. The attempts to use a spherical model of permeable macromolecules<sup>9</sup> were criticized from several points of view, and this model will not be discussed here.

by the polymeric component in unit volume will be  $n_p \pi a^2 h$  and hence the volume fraction of the polymer in solution is

$$V_p = n_p \pi a^2 h / n_p \pi R^2 h = a^2 / R^2 \quad (20)$$

For reasons of symmetry it is clear that the electrostatic potential assumes an extremal value at  $R$ , or

$$(\partial \psi / \partial r)_R = 0 \quad (21)$$

Since in the 'intercellular region', close to  $r = R$ , the electrical forces vanish, it is convenient also to make the reference potential at  $R$  equal to zero, i.e.

$$\psi_R = 0 \quad (22)$$

3.2 We now assign local electrochemical potentials to the  $i$ th small ion at a distance  $r$  from the centre of the polyion

$$\tilde{\mu}_i^r = \mu_i^0 + kT \ln n_i^r + z_i \varepsilon \psi^r \quad (23)$$

where  $n_i^r$  is the numerical concentration of the  $i$ th ion at  $r$ ,  $z_i$  is the electrical valency of the ion and  $\psi^r$  the potential at  $r$ . The basic requirement of equilibrium is that

$$\nabla \tilde{\mu}_i^r = 0 \quad (24)$$

or that  $\tilde{\mu}_i^r$  has the same value throughout the cell. As the value of  $\tilde{\mu}_i^r$  at  $R$  is, in view of equation 22,

$$\tilde{\mu}_i^R = \mu_i^0 + kT \ln n_i^R \quad (25)$$

we find that

$$\mu_i^0 + kT \ln n_i^r + z_i \varepsilon \psi^r = \mu_i^0 + kT \ln n_i^R \quad (26)$$

where we have assumed  $\mu_i^0$  to be independent of position. Equation 26 is another form of Boltzmann's equation

$$n_i^r = n_i^R \exp(-z_i \varepsilon \psi^r / kT) \quad (27)$$

and is a useful starting point for calculating the local potential at  $r$ . For the present purpose it is, however, sufficient to compare the local electrochemical potential with the average thermodynamic potential

$$\bar{\mu}_i = \mu_i^0 + kT \ln \bar{n}_i f_i \quad (28)$$

where  $\bar{n}_i$  is the average concentration in solution and  $f_i$  the activity coefficient of the  $i$ th co-ion. From the equivalence of 25 and 28 we find immediately that

$$f_i = n_i^R / \bar{n}_i \quad (29)$$

3.3 With these notions about the colligative properties attributable to a cell model, we may now proceed to the evaluation of a straightforward relation between the osmotic pressure and the activities of the ionic constituents:

Let us consider the region of solution close to  $r = R$  and assume that it is large enough to allow the assignment of thermodynamic parameters to



POLYELECTROLYTES

the component ions. Since at  $R$  the potential  $\psi_R$  is zero, all the electrochemical potentials at  $R$  become chemical potentials and assume ideal properties. Now the application of the Gibbs–Duhem equation to this region is in the form

$$\sum n_i^R d\mu_i^R = -n_w^R d\mu_w^R \quad (30)$$

but for an ideal system

$$n_i^R d\mu_i^R = n_i^R kT (dn_i^R/n_i^R)$$

while  $-d\mu_w^R = \bar{V}_w d\pi$  where  $\pi$  measures the overall osmotic pressure of the system. Inserting into equation 30 we obtain the important equation

$$kT \sum dn_i^R = n_w^R \bar{V}_w d\pi \quad (31)$$

and again, since  $n_w^R \bar{V}_w \simeq 1$ ,

$$kT \sum dn_i^R = d\pi \quad (32)$$

Equation 32 can be integrated directly; when all  $n_i^R = 0$ ,  $\pi$  vanishes and hence the integration constant is also zero, so that

$$kT \sum n_i^R = \pi \quad (33)$$

or, inserting the experimental values from equation 29, we obtain

$$kT \sum f_i \bar{n}_i = \pi \quad (34)$$

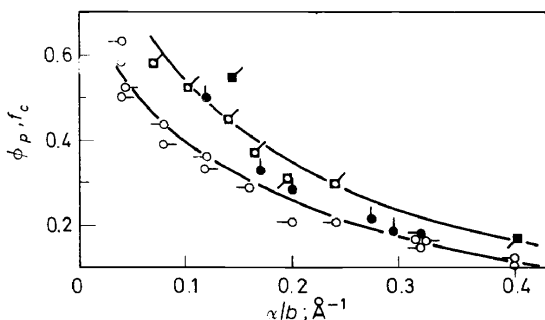


Figure 4. The osmotic coefficient  $\phi_p$  and the activity coefficient  $f_c$  of the counterions in salt-free polyelectrolyte solutions, as functions of the reciprocal intercharge distance  $(b/a)^{-1}$ . Results obtained for flexible polymer are: (—○) Na polyacrylates; (○—) Na polymethacrylates; (●) Na polyvinyl sulphate and (—●) Na polyphosphate. Results for cellulose derivatives: (■) Na carboxymethylcellulose; (■) Na cellulose sulphate and (□) Na alginate. Open and filled in symbols are used to designate  $\phi_p$  and  $f_c$  respectively. (From ref. 3, Figure 3)

This important relation was first derived by Langmuir<sup>10</sup> and considered in a more general way by Verwey and Overbeek<sup>11</sup>; the application of this equation to polyelectrolyte systems is due to the study of Marcus<sup>12</sup>.

A particularly simple consequence of equation 34 is the relation between osmotic pressure and activity of the counterions in salt-free solutions. In this case, the cell model requires only the osmotic contribution of the

counterions the concentration of which is  $n_i = vn_p = \alpha n_m$ ; the osmotic pressure under the same condition is (from equation 11)  $\pi = n_p v \phi_p kT$ , which upon insertion into 34 gives

$$kT \cdot f \cdot vn_p = n_p v \phi_p kT \quad \text{or} \quad f = \phi_p! \quad (35)$$

With the conventional assumption on the nature of liquid junctions, involved in the determination of single ion activities, equation 35 has been tested by Kern<sup>13</sup>, by Nagasawa and Kagawa<sup>14</sup> and by Liquori *et al.*<sup>15</sup>; excellent agreement was found between the electrode determination of  $f$  and direct measurement of  $\phi_p$ .

During the last few years, several workers felt that a sounder approach from a thermodynamic point of view would be the determination of the mean activity coefficient  $f_{\pm}$  of a polyelectrolyte with its counterions. The first measurements on the mean activity of numerous polyelectrolytes were carried out by Ise and Okubo<sup>16</sup>. The experimental method is based on the

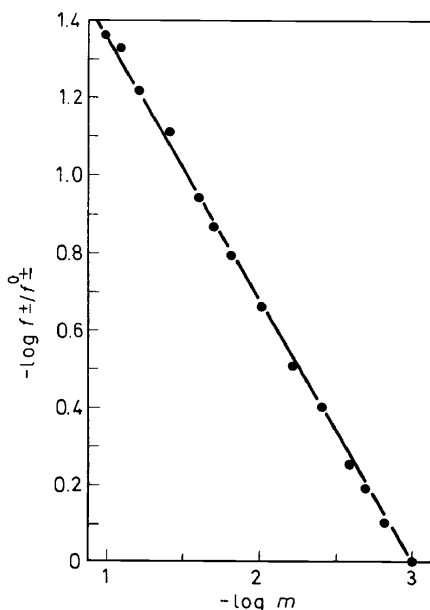
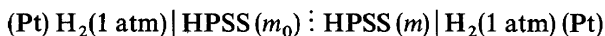


Figure 5. Data from ref. 17

measurement of the e.m.f. of cells. Since cells *without* transference require electrodes reversible to both the *polyion* and the counterions, such measurements are excluded and one has to use the less advantageous system, namely cells with transference. Thus Dolar<sup>17</sup> measured the e.m.f. in cells of polystyrene sulphonic acid (HPSS) according to the following scheme:



Since even at the highest dilutions, amenable to experimental determination,  $f_{\pm}$  does not go to unity, the only possibility is to consider a ratio of the mean

activity coefficient at a given polyelectrolyte concentration to that of a reference polyelectrolyte solution ( $f_{\pm}^0$ ). The results of Dolar's measurements are represented in *Figure 5* as a plot of  $-\log f_{\pm}/f_{\pm}^0$  versus  $-\log m$ . The reference solution was at  $m_0 = 10^{-3}$  monomole/kg and the concentration range extends from 0.001 to 0.1 monomolar polystyrene sulphuric acid.

It is rather interesting that the logarithm of the mean activity coefficient is exactly linear with the logarithm of concentration over the whole experimental range. An interpretation of this finding can be provided by again applying the equation of Gibbs–Duhem according to equation 17. Now, by the classical definition of the mean activity of a polyvalent electrolyte<sup>18</sup>, the chemical potential of the electroneutral polyelectrolyte is given by

$$\mu_p = \mu_p^0 + (v + 1)kT \ln(f_{\pm} n_p) + vkT \ln v$$

and hence  $n_p d\mu_p/dn_p$  at constant  $v$  is

$$n_p d\mu_p/dn_p = kT(v + 1) [1 + d \ln f_{\pm}/d \ln n_p] = d\pi/dn_p \quad (36)$$

From equation 7,  $d\pi/dn_p = kT(1 + v\phi_p + v d\phi_p/d \ln n_p)$  and hence

$$\frac{d \ln f_{\pm}}{d \ln n_p} = -\frac{v}{v + 1}(1 - \phi_p) + \frac{v}{(v + 1)} \frac{d\phi_p}{d \ln n_p}$$

In the present case  $v \gg 1$  so that  $v/(v + 1) \simeq 1$ . Further, since  $d \ln n_p$  is independent of the concentration units, we may write  $d \ln n_p = d \ln m$ , and finally if we consider only the range of dilute solutions in which  $d\phi_p/d \ln n_p \simeq 0$ , we obtain

$$d \log f_{\pm}/d \log m = -(1 - \phi_p) \quad (37)$$

As pointed out above, in many cases  $\phi_p$  is independent of concentration so that  $1 - \phi_p$  will be a constant, equal in *Figure 5* to 0.675, so that  $\phi_p = 0.325$ , which is of the right order of magnitude.

To make further progress in our discussion, we cannot remain within the framework of thermodynamics and our next step will be an explicit evaluation of the potentials and free energies of the polyion system.

#### 4. THEORETICAL EVALUATION OF THE ELECTROSTATIC POTENTIAL AND OSMOTIC COEFFICIENT IN SALT-FREE SOLUTIONS

4.1 We have seen that the ionic distribution in the field of the polyelectrolyte follows the Boltzmann equation 27. The next step is usually the correlation of the local charge density  $\rho_r$ , at a distance  $r$  from the centre of the polyion,

$$\rho_r = \sum n_i^r z_i \varepsilon \quad (38)$$

(where  $\varepsilon$  is the charge of a proton) with the local potential through Poisson's equation

$$\nabla^2 \psi^r = -4\pi \rho_r/D \quad (39)$$

$D$  being the local dielectric constant. In the case of a salt-free polyelectrolyte solution, which we shall consider for the sake of correctness to be a solution of a polyacid neutralized by monovalent cations, the distribution according to equation 27 is

$$n^r = n^R \exp(-\varepsilon\psi^r/kT) \quad (40)$$

For the cylindrical symmetry which governs our cell model, the Laplacian of  $\psi$  is given by  $\nabla^2\psi = (1/r) d/dr (r d\psi/dr)$  so that the Poisson-Boltzmann equation is

$$\frac{1}{r} \frac{d}{dr} \left( r \frac{d\psi}{dr} \right) = - \frac{4\pi n^R \varepsilon}{D} \exp(-\varepsilon\psi/kT) \quad (41)$$

There is no need to review again all the weighty arguments which were adduced against the applicability of the non-linearized Poisson-Boltzmann equation in the case of electrolyte solutions. It suffices to mention that in the case of a strong central field, the use of equation 41 provides a better approximation than in the case of low molecular symmetrical electrolytes—and it is rather gratifying that the recent attempt to evaluate polyelectrolyte behaviour by the cluster expansion method of statistical mechanics led Manning<sup>19</sup> to the conclusion that in dilute solutions this method leads to the same results as obtained by applying equation 41.

4.2 We shall not discuss here the mathematical treatment of the Poisson-Boltzmann equation<sup>20</sup>, but will consider in some detail the consequences of its exact solution, following the work of Lifson and myself. Multiplying both sides of equation 41 by  $\varepsilon/kT$  and denoting  $\varepsilon\psi/kT$  by  $\phi$ , equation 41 assumes the suggestive form

$$\frac{1}{r} \frac{d}{dr} \left( r \frac{d\phi}{dr} \right) = -\kappa^2 \exp(-\phi) \quad (42)$$

where

$$\kappa^2 \equiv 4\pi n^R \varepsilon^2 / DkT \quad (42')$$

is the square of a reciprocal distance, related to the thickness of the ionic atmosphere. The solution of equation 42 is

$$\phi \equiv \varepsilon\psi/kT = \ln \{ \kappa^2 r^2 \sinh^2(\beta \ln Ar) / 2\beta^2 \} \quad (43)$$

where  $A$  and  $\beta$  are integration constants.

For the evaluation of  $A$  and  $\beta$  we use the following conditions. From equation 43 we find that in general

$$\partial\psi/\partial r = (2kT/\varepsilon r) (1 + \beta \coth \beta \ln Ar) \quad (44)$$

but since  $(\partial\psi/\partial r)_R = 0$  we obtain as the first relation

$$1 + \beta \coth \beta \ln AR = 0 \quad (45)$$

## POLYELECTROLYTES

The other condition is based on the electroneutrality of each solution cell: The number of counterions in a cylindrical volume element  $dV$  lying between  $r$  and  $r + dr$  is  $n' dV = n' 2\pi r h dr$ , and hence the total number of counterions

in the cell is  $\int_{r=a}^{r=R} n' dV$ , where  $r = a$  is the surface of the polyion and  $r = R$  the

boundary of the cell. This number must be equal to the total number of charged groups  $v$  carried by the polyion, or

$$v = 2\pi h \int_a^R n' r dr \quad (46)$$

Upon insertion of  $n'$  from equations 40 and 43 we find that

$$v = (DkTh/\epsilon^2) (\beta \coth \beta \ln Aa - \beta \coth \beta \ln AR)$$

or, in view of equation 45, we may write for the second condition in the evaluation of  $A$  and  $\beta$ :

$$v\epsilon^2/DkTh = 1 + \beta \coth \beta \ln Aa \quad (47)$$

The interesting term  $v\epsilon^2/DkTh$  appearing on the LHS of equation 47 is a pure number which expresses the relation of the electrostatic energy  $v\epsilon^2/Dh$  to the thermal energy  $kT$ . It deserves a special symbol and has been christened the 'charging parameter'

$$\lambda \equiv v\epsilon^2/DkTh \quad (48)$$

For a polyelectrolyte of known dimension,  $h$ , and of known charge number,  $\lambda$  is a known quantity which plays a prominent role in the theoretical characterization of polyelectrolyte solutions. It is worth noting that even if  $h$  and  $\lambda$  are unknown but the linear density of the charges is given as  $v/h = \alpha/b$  (cf. para. 2.2c) then  $\lambda$  is still determinable by

$$\lambda \equiv \alpha\epsilon^2/DbkT \quad (49)$$

Introducing  $\lambda$  into 47 we obtain the useful form

$$\lambda = 1 + \beta \coth \beta \ln Aa \quad (50)$$

4.3 Our next aim is to evaluate from the theoretical expressions the observable colligative properties, and in particular the osmotic coefficient  $\phi_p$ . In our previous discussion on the equilibria in a polyelectrolyte system, we imposed only the condition 24 for the electrochemical potentials. A resting solution, or even a system in a slow flow process, is however generally in a state of mechanical equilibrium which is characterized by the balancing of the mechanical volume forces. The volume force due to hydrostatic pressure in an isotropic liquid is  $\nabla p$ , while that due to the existence of a local electrical field  $E = -\nabla\psi$  is  $\rho E = -\rho\nabla\psi$  where  $\rho$  is as before given by equation 38.

Thus local mechanical equilibrium is expressed by

$$-\rho\nabla\psi + \nabla p = 0 \quad (51)$$

Inserting  $\rho$  from Poisson's equation we may rewrite 51 as follows

$$(D/4\pi)\nabla^2\psi + \nabla p = 0 \quad \text{or} \quad \nabla[p + (D/8\pi)E^2] = 0 \quad (52)$$

The integration of 52 gives

$$p + (D/8\pi) E^2 = \text{const.} \quad (53)$$

showing that the total pressure acting in the solution is composed of a mechanical pressure  $p$  and an electrical pressure  $(D/8\pi) E^2$ , the sum of which is constant. Thus the value of the constant may be obtained from the magnitude of  $p$  at the point where  $E = 0$ . But this is the point  $R$  [at which  $(\partial\psi/\partial r)_R = 0$ ] so that  $p^R$  is the overall constant equilibrium pressure of the solution. This pressure is evidently the osmotic pressure which we evaluated thermodynamically in para. 3 (equation 33), or

$$p^R = p^r + (D/8\pi) E_r^2 = \pi \quad (54)$$

From equations 33 and 8 the osmotic pressure of a salt-free solution is

$$\pi = n^R kT = n_p \nu \phi_p kT \quad (55)$$

hence the theoretical evaluation of  $n^R$  will provide the overall pressure prevailing in the solution cell.

4.4 A convenient way to evaluate  $n^R$  is from equations 43 and 45. At  $r = R$  when  $\psi_R = 0$ , equation 43 yields

$$\kappa^2 R^2 \sinh^2 \beta \ln AR/2\beta^2 = 1 \quad (56)$$

Equation 45 may be rewritten as

$$\coth^2 \beta \ln AR - 1 = 1/(\sinh^2 \beta \ln AR) = (1 - \beta^2)/\beta^2 \quad (57)$$

and hence

$$\frac{1}{2} \kappa^2 R^2 = 1 - \beta^2 \quad (58)$$

or, inserting  $\kappa^2$  from equation 42', we find

$$n^R = (1 - \beta^2) DkT/2\pi R^2 \epsilon^2 \quad (59)$$

Recalling that the volume of each solution cell is  $\pi R^2 h = 1/n_p$ , equation 59 can be rewritten as  $n^R = n_p (1 - \beta^2)/2(\epsilon^2/DhkT)$  which when inserted into equation 55 gives for the osmotic coefficient the value

$$\phi_p = (1 - \beta^2)/2(\nu\epsilon^2/DhkT) = (1 - \beta^2)/2\lambda \quad (60)$$

It is convenient to write for  $\lambda$

$$\lambda = \alpha\lambda_0 \quad (61)$$

where the constant  $\lambda_0$  is by equation 49

$$\lambda_0 = \epsilon^2/DbkT \quad (62)$$

Inserting 61 into 60 we find that the effective degree of ionization is given by

$$i = \alpha\phi_p = (1 - \beta^2)/2\lambda_0 \quad (63)$$

Thus the constancy of the effective degree of ionization, considered in para. 2.1, depends on the constancy of the integration constant  $\beta$ . The value of  $\beta$  is determined by the charging parameter  $\lambda$  and by the volume concentra-

POLYELECTROLYTES

tion of the polyelectrolyte ( $V_p = a^2/R^2$ ). By straightforward manipulation of equations 45 and 50 we find the transcendental equation

$$\lambda = (1 - \beta^2)/(1 + \beta \coth [\beta \ln (R/a)]) \tag{64}$$

where, according to equation 20,  $\gamma \equiv \ln R/a = -\frac{1}{2} \ln V_p$ . The dependence of  $\beta$  on  $\lambda$  for different  $\gamma$ s is represented in Figure 6.

It will be observed that for all values of  $V_p$ , the value of  $\beta$  for  $\lambda = 0$  is  $\beta = 1$ . The magnitude of  $\beta$  decreases with increasing  $\lambda$  and reaches a value of

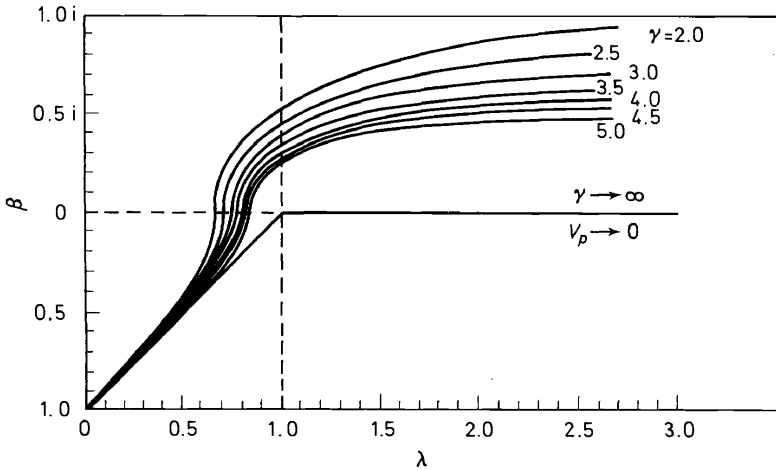


Figure 6. Integration constant  $\beta$  as a function of the charge density parameter for a series of values of the concentration parameter  $\gamma$ . (S. Lifson and A. Katchalsky, ref. 20)

zero at  $\lambda \leq 1$ . A further increase in  $\lambda$  causes a dramatic change in  $\beta$ , for it passes from the realm of real numbers into the realm of imaginary numbers. This transition will be considered in the following paragraph; here it is important to notice that for  $\lambda$ s larger than 1,  $\beta$  approaches a constant limiting value—depending on the concentration. Since the constancy of  $\beta$  means also constant  $i$ , we have here the theoretical interpretation of the fact that at higher degrees of ionization the effective degree of ionization is practically constant.

4.4 The constancy of  $\alpha\phi_p$  at higher values of  $\lambda$  has, however, a deeper significance. According to equations 25, 28 and 35 for a salt-free solution, the chemical potential of the counterion is given by

$$\tilde{\mu}_c = \mu_c^0 + kT \ln(n_p \nu \phi_p) = \mu_c^0 + kT \ln(n_m \alpha \phi_p) = \mu_c^0 + kT \ln(n_m i) \tag{65}$$

If we now increase the degree of ionization, keeping the polymer concentration constant, we find that the chemical potential does not change, or

$$(\partial \tilde{\mu}_c / \partial \alpha)_{n_m} = 0 \tag{66}$$

This can be interpreted as if the counterions are distributed between two phases which maintain an equilibrium—a ‘condensed phase’ of the atmosphere surrounding the polyion, and a ‘vapour phase’ of ions floating

freely in solution. Increasing the number of counterions per macromolecule does not increase their chemical potential, but causes an additional condensation around the polyelectrolyte molecules.

To reach the point of condensation a critical value of  $\lambda$  is required which can be identified with the values of *Figure 6* at which  $\beta$  reaches constancy. As pointed out by Manning<sup>19</sup>, following the suggestion of Onsager, the critical point can best be demonstrated in a clearer manner in the case of 'infinitely dilute' solutions. In this case  $V_p \rightarrow 0$  and  $\gamma \rightarrow \infty$  for which equation 64 has two physically meaningful solutions: for  $\lambda < 1$ ,  $\beta = 1 - \lambda$ , while for  $\lambda > 1$ ,  $\beta = 0$ . Thus for  $\lambda < 1$ ,

$$\phi_p = [1 - (1 - \lambda)^2]/2\lambda = 1 - \frac{1}{2}\lambda \quad (67)$$

while for  $\lambda > 1$ ,

$$\phi_p = \frac{1}{2}\lambda \quad (68)$$

At  $\lambda = 1$  both solutions coincide and  $\phi_p = \frac{1}{2}$ .

It will be observed that for  $\lambda < 1$ ,  $\phi_p$  decreases linearly with  $\lambda$  so that the effective ionization is not constant. On the other hand at  $\lambda > 1$  even for 'infinite' dilution

$$i = \alpha\phi_p = 1/2\lambda_0 = \text{const.} \quad (69)$$

If we insert 67 and 68 into equation 69 we find that, as above, in the range of  $\lambda < 1$ ,  $(\partial\tilde{\mu}_c/\partial\alpha)_{n_m} > 0$  which proves that the system is thermodynamically stable, while for  $\lambda > 1$ ,  $(\partial\tilde{\mu}_c/\partial\alpha)_{n_m} = 0$  so that condensation is expected also at very high dilutions—although in reality neither theory nor its consequences should be driven to 'infinite dilution'.

To conclude this paragraph let us evaluate numerically the constant value of  $i$  at high dilutions. By equation 63,  $i = 1/2\lambda_0$  and since  $\lambda_0 = \epsilon^2/DbkT$ , we shall use the following values  $-\epsilon = 4.8 \times 10^{-10}$  e.s.u.; at room temperature,  $kT = 4.1 \times 10^{-10}$  ergs; for the interchange distance we shall take the value for vinylic polymers  $b = 2.55 \times 10^{-8}$  cm, and for the dielectric constant  $D$  we shall assume the value of the pure solvent, i.e.  $D = 78$ . Inserting all the constants we find  $\lambda_0 = 2.8(>1)$  and hence  $i = \alpha\phi_p = 0.177$ , which fits closely many of the experimental data.

## 5. EXPERIMENTAL VERIFICATION OF THE THEORY OF SALT-FREE POLYELECTROLYTE SOLUTIONS

5.1 To obtain a quantitative test of the theory outlined above it is important to test the behaviour of a rodlike polyelectrolyte, the dimensions of which are independent of the degree of ionization. Such an example was provided by Auer and Alexandrowicz in sonicated native DNA<sup>21</sup>. It was found that the osmotic coefficient of highly purified and fractionated, salt-free DNA solutions is  $\phi_p = 0.17-0.20$ . This value closely corresponds to the calculated value in the concentration range  $(2-10) \times 10^{-3}$  M,  $\phi_p = 0.17-0.19$ .

A wealth of measurements is available for different synthetic and natural polyelectrolytes, some of which are given in *Figures 1* and *2*. In all cases, the theoretical approach provides a useful guide to the evaluation of osmotic



properties. Generally, however, the polyions are not fully stretched and their precise dimension unknown. It may be stated *a priori* that  $b_{\text{effective}}$  of the polymer is smaller than the fully stretched value  $b_{\text{stretch}}$ , and hence the magnitude of  $\lambda_0$  to be used for the evaluation should be larger than that calculated for rigid molecules. A good fit was obtained by assuming that  $\lambda_0^{\text{effective}}$  for vinylic polymers is twice that of the fully stretched, while  $\lambda_0^{\text{effective}}$  for cellulose derivatives is about 1.5 that of the rigid polyions.

Recently some attention was paid to the dependence of the osmotic coefficient  $\phi_p$  on polyelectrolyte concentration. In particular, Ise and Okubo<sup>16</sup>, as well as Chu and Marinsky<sup>22</sup> carried out an extensive series of measurements on different polyelectrolytes to elucidate this point. It was found that in all cases  $\phi_p$  increases with concentration and its magnitude at, say, molar concentration, may be almost twice that at a hundredth molar. This is to be expected on theoretical grounds, since according to equation 60,  $\phi_p = (1 - \beta^2)/2\lambda$ , and, as shown in *Figure 6* for larger values of  $\lambda$ ,  $\beta$  becomes imaginary or  $\beta = i|\beta|$  so that  $\phi_p = (1 + |\beta|^2)/2\lambda$ . Moreover,  $|\beta|$  increases with concentration and hence the predicted dependence pattern of  $\phi_p$  with concentration for a constant value of  $\lambda = \lambda_0$  compares satisfactorily with that found by Chu and Marinsky for H-polystyrene sulphonate.

The data indicate, however, that the concentration dependence is also influenced by the nature of the counterion. Although the values extrapolated to zero concentration seem to converge to the same magnitude [e.g. in ref. 22 the  $\text{H}^+$ ,  $\text{Li}^+$ ,  $\text{Na}^+$ ,  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Cs}^+$ ,  $\text{TMA}^+$ ,  $\text{TEA}^+$  forms of polystyrene sulphonic acid all extrapolate to  $\phi_p = 0.21\text{--}0.23$  (at  $m = 0$ )] the slope of  $\phi_p$  versus  $c$  is ion-specific. An attempt to include the finite radius of the counterion, in a procedure similar to that of Debye and Hückel, was made by Kagawa and Gregor<sup>23</sup> and leads to a corrected value of  $\phi_p$

$$\phi_p = \{(1 - \beta^2)/2\lambda\} [1 - (1 + r_h/a)^2 V_p] \quad (70)$$

in which  $r_h/a$  is the ratio of the radius of the counterion to that of the polymeric cylinder, and  $V_p$ , as before, is the volume fraction of the polyelectrolyte in solution. Equation 70 shows that the effect of the finite size of the counterions indeed becomes more prominent with increasing  $V_p$  and vanishes when  $V_p \rightarrow 0$ . The order of the effect for the alkali cations does not, however, fit that predicted from their hydrated radii and there is little doubt that a more detailed theory of the excluded volume in the 'condensation layer' is required.

5.2 A very important field of study is that of the interaction of charged polymers with divalent ions. From the point of view of electrostatic theory, the stronger attraction of polyvalent small ions to the polyion should reduce the concentration of 'free' counterions and the  $\phi_p$  of divalent ions should be about half its value for monovalent counterions. Indeed, our measurements<sup>24</sup> on the  $\phi_p$  of  $\text{Mg}^{2+}$  alginate gave a value of  $\phi_p = 0.15$  as compared with  $\phi_p = 0.4$  for  $\text{Na}^+$  alginate or  $\phi_p = 0.35$  for  $\text{K}^+$  alginate. Similar measurements on the activity coefficients of salt-free polymethylstyrene sulphonates<sup>25</sup> gave for the Zn salt  $f_c = 0.11\text{--}0.15$  and for the Cs salt,  $f_c = 0.09\text{--}0.15$ ; the corresponding values for the monovalent salts were for the hydrogen form of polyvinylsulphonic acid  $f_c = 0.47\text{--}0.50$ , for the sodium salt  $f_c = 0.31\text{--}0.43$  and for the silver salt  $f_c = 0.44\text{--}0.46$ .

The rodlike model can be extended also for the evaluation of the osmotic coefficients of mixed polyelectrolyte salts in which the polyion is neutralized by both mono- and di-valent ions. The theoretical evaluation was carried out by Dolar and Peterlin<sup>26</sup>, and the predictions of the numerical calculation were tested by Dolar and Kozak<sup>27</sup>. Denoting the concentration of the monovalent ions at the distance  $R$  from the polyion by  $n_1^R$  and those of the divalent ions by  $n_2^R$ , and assuming as before that  $\psi(R) = 0$  and  $(\partial\psi/\partial r)_R = 0$ , the Poisson-Boltzmann equation becomes

$$\frac{1}{r} \frac{d}{dr} \left( r \frac{d\psi}{dr} \right) = \frac{4\pi\epsilon}{D} [n_1^R \exp(-\epsilon\psi/kT) + 2n_2^R \exp(-2\epsilon\psi/kT)] \quad (71)$$

If the average concentration of the monovalent ions is  $\bar{n}_1$  and that of the divalent  $\bar{n}_2$ , then the osmotic coefficient is given by

$$\phi_p = (n_1^R + n_2^R)/(\bar{n}_1 + \bar{n}_2) \quad (72)$$

while the total charge on the polyelectrolyte molecule is

$$v = (\bar{n}_1 + 2\bar{n}_2) V \quad (73)$$

Both theory and experiment indicate the existence of a maximal value of  $\phi_p$  at a certain ratio of monovalent to divalent ions. Denoting the fraction of the polymeric charge neutralized by monovalent counterions by  $\bar{N}_1 = \bar{n}_1/(\bar{n}_1 + 2\bar{n}_2)$  the following behaviour was observed with a mixed  $\text{Na}^+ - \text{Mg}^{2+}$  salt of polystyrenesulphonic acid.

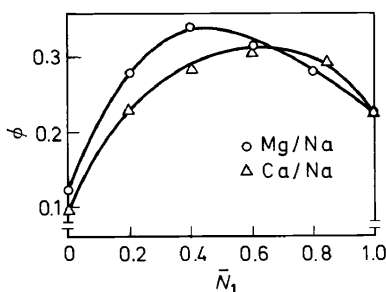


Figure 7. Osmotic coefficient  $\phi$  as a function of the equivalent fraction  $\bar{N}_1$  of the monovalent counterion component. Mixed solutions of NaPSS and MgPSS ( $\circ$ ), and of NaPSS and CaPSS ( $\Delta$ ) at the constant concentration of the polyion (0.012 monomolal). (Ref. 27)

5.3 The basic assumption of the theoretical picture developed hitherto is that despite the striking deviation of the colligative properties from the ideal pattern no appreciable 'true' binding of counterions to specific sites on the polyion has to be considered and all behaviour may be attributed to electrostatic field effects. There is little doubt that this model holds well for all monovalent counterions studied hitherto and for many divalent counterions. A direct proof that whatever be the distribution of the counterions, whether in a loose Debye-type of atmosphere or largely as a condensed Bjerrum layer, all the counterions are fully ionized, and thus free in a chemical sense, was supplied by the important study of Kotin and Nagasawa<sup>28</sup>. These authors

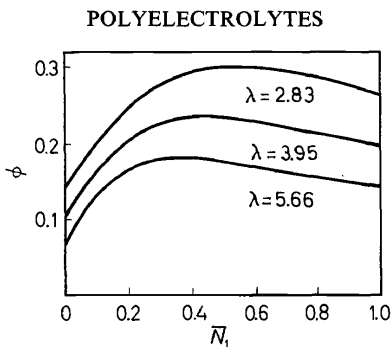


Figure 8. Theoretical curves calculated for the indicated values of the charging parameter  $\lambda$  and for  $\ln(R/a) = 3.00$ . (Ref. 28)

measured the proton magnetic resonance of polystyrenesulphonic acid and found that throughout the range of polymer concentrations employed the concentration of the hydronium ions corresponds to a full ionization of the sulphonium groups. This finding is supported by the exciting study of low angle x-ray scattering of DNA-sodium solutions carried out by Bram and Beeman<sup>29</sup>. It was found that the molecules can be represented as two concentric cylinders—that of the macromolecule and a 20 Å thick outer ionic shell—which could not be resolved since no site binding could be attributed to the counterions.

There exist, however, numerous cases of divalent ion interaction with a polyion in which the electrostatic attraction leads ultimately to specific association. Thus, in calculating  $\phi_p$  values for alginates it was found that  $\phi_p$  for  $\text{Cu}^{2+}$  is 0.03 and  $\phi_p$  for Ca is only 0.01, i.e. five to ten times lower than the value expected from a consideration of electrostatic factors only. Lyons and Kotin<sup>30</sup> indicated that  $\text{Mg}^{2+}$  may undergo some site binding to DNA, and several months ago Sander and Ts'o<sup>31</sup> could evaluate the complex binding of  $\text{Mg}^{2+}$  to a variety of nucleic acids and synthetic polynucleotides.

In the case of stronger binding, precipitation may occur at a critical ratio of divalent ions to polymeric charges. Thus phase transitions were observed with the addition of  $\text{Ca}^{2+}$ ,  $\text{Sr}^{2+}$  or  $\text{Ba}^{2+}$  to ionized solutions of polyacrylic or polymethacrylic acids.

In a detailed study of the precipitation behaviour, Michaeli<sup>32</sup> found that the critical concentration of the precipitating ion ( $n_{\text{Ca}^{2+}}^{\text{crit}}$ ) is simply related to the concentration of the polymeric charged groups  $n_p v$ , namely

$$n_{\text{Ca}^{2+}}^{\text{crit}} \simeq 0.8 n_p v$$

The point should be stressed that it is not the overall concentration of the counterion which leads to phase separation, but when  $n^{\text{crit}}/n_p v \simeq 0.8$ , whatever  $n_p$  may be and for any degree of ionization, precipitation occurs.

A similar behaviour was observed also by Zwick in charged polyelectrolyte gels<sup>33</sup>. When a slightly crosslinked gel of a polymethacrylic acid is fully or partially neutralized by sodium hydroxide, it swells to a large extent and the ratio of the swollen volume  $V$  to the initial volume  $V_0$  may reach values of several hundreds. If the sodium counterions are subsequently exchanged by alkali-earth ions, there is first a gradual shrinking of the volume up to say

50 per cent of the fully swollen value. When, however, about 80 to 87 per cent of the sodium is exchanged by  $\text{Ca}^{2+}$  or  $\text{Ba}^{2+}$ , a dramatic shrinking to almost dry volume takes place. The process is reversible and closely resembles the precipitation behaviour in solution.

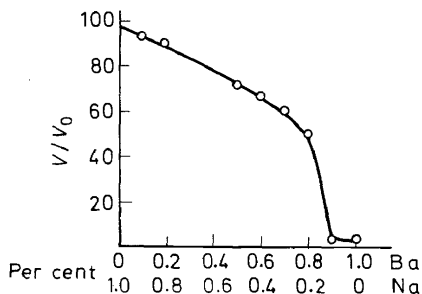
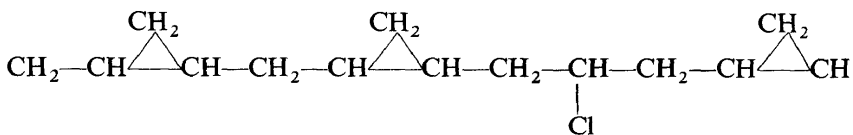


Figure 9. The swelling of a fully ionized polymethacrylic acid gel as a function of the ratio of barium to sodium counterions in the gel. (Ref. 33)

No rigorous theoretical explanation exists for this interesting phenomenon. The fact, however, that precipitation or gel contraction always occurs when about 80 per cent of the polymer ionized groups are occupied by a divalent ion, and this independent of the nature of the polyion, of its degree of ionization or of the specificity of the interaction, seems to indicate that the process is governed by a statistical rule. Since the phenomenon takes place only with ions of very low  $\phi_p$  it may be assumed that their binding is strong enough that the bond may be kinetically approximated by an irreversible, quasi-covalent bond, the lifetime of which is long enough to stabilize certain links between polyelectrolyte molecules. Indeed, for covalent interaction with uncharged polymers such a statistical behaviour was found many years ago by Marvel *et al.*<sup>34</sup>

When treating dioxane solutions of polyvinylchloride with metallic zinc to obtain polymeric cyclopropane it was found that no matter how much Zn was added, only 84 to 86 per cent of the chlorine was removed from the PVC molecules. Flory<sup>35</sup> showed statistically that the random interaction of the Zn with two neighbouring chlorine atoms always leaves some lone atoms, such as



the percentage of which is  $1/e^2$ , or 13.5 per cent, so that the maximum extent of crystallization is  $1 - 1/e^2$ , or 86.5 per cent.

Applying Flory's theory to the strongly bound  $\text{Ca}^{2+}$  or  $\text{Ba}^{2+}$ , we would assume that the ions first combine with neighbouring pairs of  $\text{COO}^-$  ions and remain rather fixed in position. This combination leaves about 13.5 per cent of lone carboxyls, which can be satisfied only if  $\text{Ca}^{2+}$  or  $\text{Ba}^{2+}$  ions

## POLYELECTROLYTES

will bind a carboxyl from one polymer molecule with another. But this type of binding leads to a crosslinking with concomitant precipitation from solution or the contraction of a gel.

No matter which interpretation is adopted, the importance of the interaction of divalent ions with biopolyelectrolytes cannot be overestimated. Thus the studies on the binding of  $Mg^{2+}$  to nucleic acids are directly stimulated by the interest in the forces holding the ribosomes and the integrity of nucleic acids in their native state. An extensive literature exists on  $Ca^{2+}$  and its role in physiology, which cannot be covered here. It is worth mentioning only that the crosslinking of polyelectrolyte molecules may play a role in the attachment of cell to cell through junctions between polyuronic acids in cellular surfaces.

Thus the classical work of Roux and Herbst, followed by the studies of Holtfreter, Bonner and other pioneers of embryology, demonstrated that the removal of  $Ca^{2+}$  by Versene causes a dissociation of the intracellular cement and the separation of young or embryonic tissue into single cells; the addition of  $Ca^{2+}$  to geometrically separated intact cells allows reformation of the original tissue. Our recent observations<sup>36</sup> show that the amount of  $Ca^{2+}$  required for maintenance of the tissue has a critical value which is probably close to  $\sim 80$  per cent of the negative surface groups. Here we have, therefore, an indication that the animals and plants learned how to form tissue when they discovered the crosslinking ability of  $Ca^{2+}$  and made use of the right statistics to establish strong intercellular combination.

5.4 In concluding this section it is worth considering another experimental verification of polyelectrolyte theory. Dolar and his co-workers<sup>37</sup> have

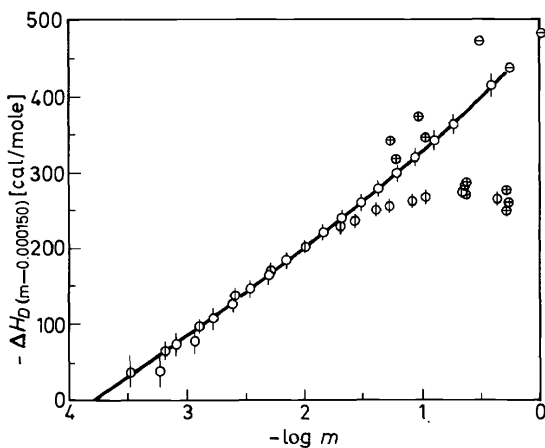


Figure 10. Intermediate molal enthalpies of dilution of polystyrenesulphonates in water at 25°C. Experimental values for the acid (○—this work, ⊖—F. Vaslow) and its sodium salt (◻—this work, ⊕—F. Vaslow). Calculated values (—). (Ref. 37)

recently measured over a wide range of concentrations the heat of dilution of polystyrenesulphonic acid and its sodium salt. Since knowledge of the potential of polyelectrolyte molecules permits evaluation of the inner and

free energies of polyelectrolyte solutions, it is possible to derive in a straightforward manner the enthalpy of dilution. Thus, the inner electrostatic energy ( $U$ ) is given by the well known expression  $U = (D/8) \int_V (\text{grad } \psi)^2 dV$ , from which the free energy is obtained by a charging process due to Verwey and Overbeek<sup>38</sup>. Applying the equation of Gibbs-Helmholtz, the dilution enthalpy  $\Delta H_D$  was evaluated and compared with the experimental data. The excellent agreement between the predicted and measured results for the free acid and for the sodium polystyrenesulphonate is shown in *Figure 10*.

## 6. POLYELECTROLYTE-SALT MIXTURES

6.1 Although the case of salt-free polyelectrolyte solutions is of theoretical interest since it can be rigorously solved from fundamental equations, it is of limited importance to the physiologist, the biochemist or the applied chemist. Polyelectrolytes are generally surrounded by low molecular salts and their behaviour is a function of their interaction with the surrounding charged particles. On *a priori* grounds, the influence of low molecular ions on the highly charged polyelectrolyte should be very complex; but due to the existence of a rather fixed screening atmosphere consisting of the 'condensed' counterions, the general features of polyelectrolyte-salt mixtures turn out to be remarkably simple.

Let us start by considering the osmotic pressure of a mixture of a polyelectrolyte and a mono-monovalent low molecular salt: it was found<sup>39</sup> that, to a good approximation, the total osmotic pressure ( $\pi$ ) is equal to the sum of the osmotic pressures of the salt-free polyelectrolyte ( $\pi_p$ ) and the polyelectrolyte-free salt solution ( $\pi_s$ ), i.e.

$$\pi = \pi_p + \pi_s \quad (73)$$

or inserting the value of  $\pi_p = (n_p + n_p v \phi_p) kT$  and writing for the salt solution  $\pi_s = 2n_s \phi_s kT$ , where  $\phi_s$  is the osmotic coefficient of the polyelectrolyte-free salt solution.

$$\pi = (n_p + n_p v \phi_p + 2n_s \phi_s) kT \quad (74)$$

As pointed out before,  $n_p$  may generally be neglected. Moreover, if we define an osmotic coefficient  $\phi$  for the mixture by the evident relation  $\pi \equiv \phi(n_p + n_p v + 2n_s) kT$ , we may write

$$\phi \approx (n_p v \phi_p + 2n_s \phi_s) / (n_p v + 2n_s) \quad (75)^*$$

---

\* In a recent publication<sup>19</sup>, Manning derived our equation 75 from considerations of counterion concentration. Assuming that for dilute salt solutions  $\phi_s \approx 1$  and denoting  $n_p v / n_s \equiv x$ , he writes equation 75 in the form

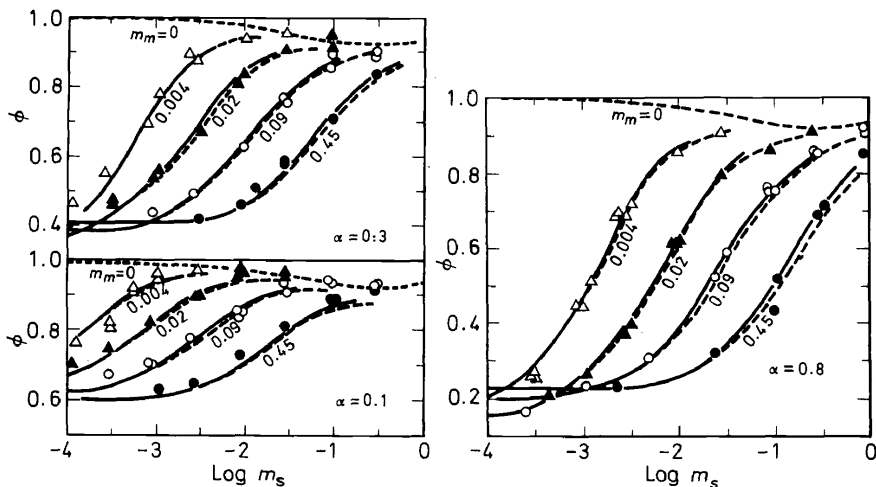
$$\phi = (x\phi_p + 2)/(x + 2) \quad (75')$$

If we further assume that the polyelectrolyte concentration is very low so that equations 67 and 68 may be used for  $\phi_p$ , we obtain: at  $\lambda < 1$ ,

$$\phi = \frac{x(1 - \lambda/2) + 2}{x + 2} = 1 - \frac{\lambda x}{2(x + 2)} \quad \text{and for } \lambda > 1, \phi = \frac{x/2\lambda + 2}{x + 2}.$$

POLYELECTROLYTES

The excellent agreement of equation 75 with a large number of experimental results can be seen in *Figure 11*, taken from the study of Alexandrowicz on mixtures of NaBr and polyacrylic acid at different degrees of ionization and different concentrations<sup>3</sup>.



*Figure 11.* Osmotic coefficient  $\phi$  of polyacrylic acid–NaBr solutions, as a function of the salt concentration ( $\log m_s$ ) at several constant monomolal concentrations of the polymer ( $m_m$ ) (after Alexandrowicz, ref. 39). Experimental results: (○)  $m_m = 0.45$ ; (●)  $m_m = 0.09$ ; (△)  $m_m = 0.02$ ; (▲)  $m_m = 0.004$ . (—) theoretical, as calculated from the hybrid potential of equation 43; (---) the empirical additivity rule 75\*. (..) represents the osmotic coefficient of pure sodium bromide solutions.

6.2 A further conclusion from the additivity of the colligative properties of polyelectrolyte–salt solutions concerns the magnitude of the activity coefficient of the counterion. It will be recalled that according to equation 34, the osmotic pressure for the solution is

$$\pi = (n_+ f_+ + n_- f_-) kT \tag{76}$$

where  $n_+$  is the concentration of the counterions,  $n_+ = n_p v + n_s$ ;  $n_-$  is the concentration of the co-ions  $n_- \simeq n_s$ , and where  $f_+$  and  $f_-$  are the corresponding activity coefficients. Since the electrostatic repulsion restricts the co-ions to regions of low potential, it is plausible to assume that the coefficient  $f_- \simeq 1$ , and this assumption is amply supported by the measurements of Kagawa and Katsura<sup>40</sup>, and of Nagasawa *et al.*<sup>14</sup>. Inserting the value  $\pi$  from equation 74 into 76, and taking  $f_- = 1$ , we find that

$$f_+ = (n_p v \phi_p + n_s) / (n_p v + n_s) \tag{77}$$

and that the activity of the counterions [ $a_+ = f_+(n_p v + n_s)$ ] is equal to

$$a_+ = n_p v \phi_p + n_s \tag{78}$$

As in this approximation

$$a_- = n_s \quad (79)$$

the activity of the salt in the presence of charged polyelectrolyte is

$$a_s = a_+ a_- = n_s(n_p \nu \phi_p + n_s) \quad (80)$$

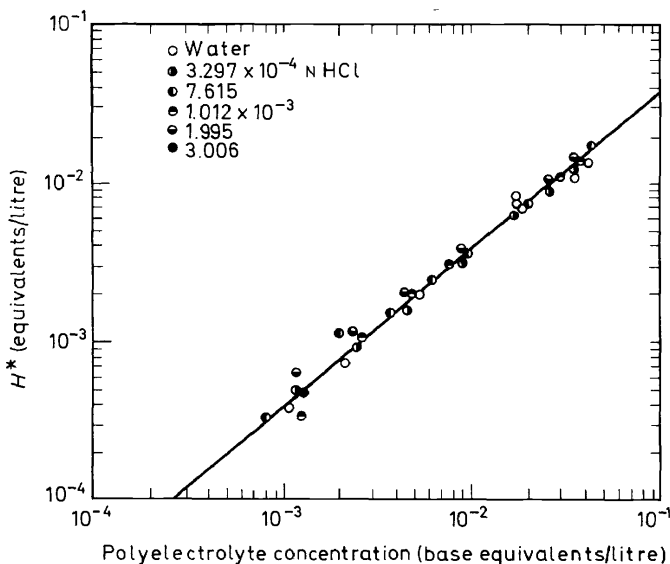
Equation 78 is corroborated experimentally by the measurements of Mock and Marshall<sup>41</sup> on the pH of mixtures of a fully ionized polysulphonic acid and a low molecular mineral acid. In this case, the counterion is the hydrogen ion and the concentration of the fully ionized mineral acid is equivalent to that of a low molecular salt. Equation 78 may therefore be rewritten as

$$a_H = c_m \phi_p + c_{\text{acid}} \quad (81)$$

where  $c_m$  is the monomolar concentration of the polysulphonate. Taking the negative logarithm of equation 81, we obtain

$$\text{pH} = -\log(c_m \phi_p + c_{\text{acid}}) \quad (82)$$

which represents very closely the experimental data shown in *Figure 12*.



*Figure 12.*  $c_m \phi_p$  (denoted as  $H^*$  in the figure) versus the polymer concentration  $m_m$  (after Mock and Marshall<sup>41</sup>)

The single  $\phi_p$  value used in the plot is  $\phi_p = 0.4$ , which is practically independent of  $c_m$  or  $c_{\text{acid}}$ .

Extensive tests of the validity of equation 73 were carried out by Lyons and Kotin<sup>42</sup>. They write the equation in the form  $a_+^{\text{obs}} = a_+^v + a_+^s$ , where  $a_+^{\text{obs}}$  is the measured activity of the positive counterions to a strong polyacid,



$a_s^0$  is the activity of the counterions in a pure polyacid solution and  $a_+^s$ , the activity of the cation in a polyelectrolyte-free salt solution. The polyelectrolytes studied were sodium salts of polystyrene sulphonic acid, polyvinyl sulphonic acid, polyacrylic acid, polyphosphoric acid and DNA. In all cases the deviation from additivity did not exceed a few per cent. Indeed, the additivity rule holds so well that the deviation from it, as observed with Mg salts of DNA, could be used by Lyons and Kotin as an indicator of site binding.

6.3 Another test of equation 80 is provided by the study of the Donnan distribution of salt. At equilibrium, the activity of a salt across a membrane is the same in the polyelectrolyte-salt mixture as in the polyelectrolyte-free salt solution. Let the activity of the dilute salt solution in the polyelectrolyte-free medium be

$$a_s' = (n_s')^2 \tag{83}$$

where the activity coefficient is taken as unity. Equating the activity of the salt in the outer solution (equation 83) with that in a polyelectrolyte compartment (equation 80), we find that

$$(n_s')^2 = n_s(n_p\nu\phi_p + n_s) \tag{84}$$

or

$$n_s = -\frac{n_p\nu\phi_p}{2} + \left[ \left( \frac{n_p\nu\phi_p}{2} \right)^2 + (n_s')^2 \right]^{\frac{1}{2}} \tag{85}$$

In the case of excess salt, when  $n_p\nu\phi_p/n_s' < 1$ , equation 85 can be reduced to

$$n_s = n_s' - \frac{n_p\nu\phi_p}{2} \left( 1 - \frac{n_p\nu\phi_p}{4n_s'} \right) \tag{86}$$

The Donnan distribution of salt between a polymer solution and an external salt solution is generally characterized by the parameter  $\Gamma$ , defined as salt deficiency in the polyelectrolyte medium per unit polymeric charge, i.e.

$$\Gamma = (n_s' - n_s)/n_p\nu \tag{87}$$

Inserting equation 86 into 87, we find that

$$\Gamma = \frac{\phi_p}{2} \left( 1 - \frac{n_p\nu\phi_p}{4n_s'} \right) \simeq \frac{\phi_p}{2} \tag{88}$$

or for very dilute polyelectrolyte solutions

$$\Gamma = \frac{1}{2}(1 - \lambda/2) \text{ for } \lambda < 1 \text{ and } \Gamma = 1/4\lambda \text{ for } \lambda > 1.$$

The agreement of equation 88 with experimental data determined by different authors is shown in *Table 1* from Manning's paper<sup>19</sup>.

A rather detailed study of Donnan distribution between gel and solution was carried out by Marinsky and his co-workers<sup>46</sup>. The polyelectrolyte gels investigated were ion exchange systems of different degrees of crosslinking, interacting with different salt solutions. Upon introducing pertinent

Table 1. Comparison of theoretical and experimental values of  $\Gamma$ 

System	Reference	$\lambda$	$\Gamma_{\text{expt}}^a$	$\Gamma_{\text{theoret}}^b$
NaPVS; NaCl	14	1.85	0.10	0.14
KPP; KBr	43	2.85	0.08	0.09
KDNA; KBr	44	4.20	0.09	0.06
NaPA; NaBr ( $\alpha = 0.8$ )	45	2.28	0.11	0.11
NaPA; NaBr ( $\alpha = 0.3$ )	45	0.86	0.24	0.29
NaPA; NaBr ( $\alpha = 0.1$ )	45	0.29	0.38	0.43

<sup>a</sup> In each case, these values correspond to the lowest salt concentration used in the measurements.

<sup>b</sup> Calculated from equation 88.

corrections for the high density of the gels, Marinsky found a close agreement between the experimental data and the values predicted by equation 84.

6.4 Finally, we would like to consider the Donnan osmotic pressure or the oncotic<sup>†</sup> pressure, in physiological terms. It is the difference between the osmotic pressure of the polyelectrolyte-salt mixture and an external salt solution with which it maintains a membrane equilibrium. The Donnan osmotic pressure is clearly the hydrostatic pressure head which develops in an osmometer filled with a polyelectrolyte solution and equilibrated with an outer salt solution.

By definition, the ideal Donnan osmotic pressure equals

$$\pi = (n_p + n_p \nu \phi_p + 2n_s - 2n'_s) kT \quad (89)$$

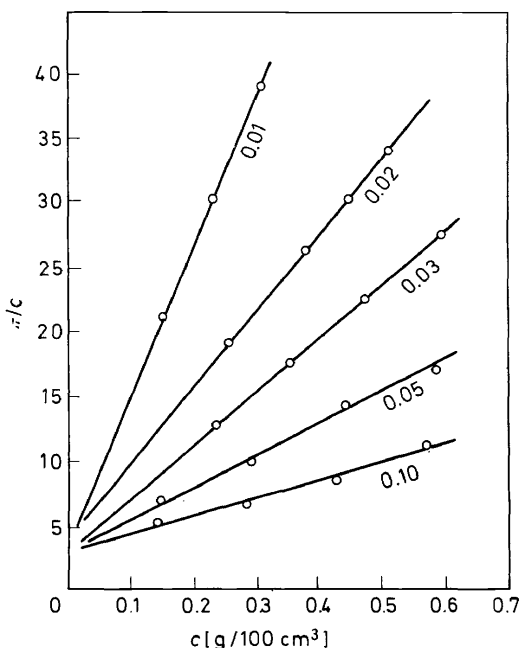


Figure 13. The reduced osmotic pressure  $\pi/c$  versus polymer concentration  $c$  in NaCMC-NaCl solutions, for several constant salt concentrations (after Inagaki *et al.*<sup>47</sup>)

In the case of excess salt, the use of equation 86 is allowed, and the expression becomes

$$\pi = \left( n_p + \frac{(n_p v \phi_p)^2}{4n'_s} \right) kT = n_m kT \left( \frac{1}{P} + \frac{n_m}{4n'_s} (\alpha \phi_p)^2 \right)$$

or, in a more convenient form,

$$\frac{\pi}{n_m kT} = \frac{1}{P} + \frac{n_m}{4n'_s} i^2 \quad (90)$$

Equation 90 permits the evaluation of the degree of polymerization, by plotting  $\pi/n_m kT$  versus  $n_m$ , or  $\pi/c$  versus  $c$ , as has been done by Inagaki<sup>47</sup> for various salt concentrations.

As was shown by Inagaki and Hiram and by others<sup>47</sup> extrapolation to zero ionic strength is not a sure procedure. The slope of the straight lines provides, however, an independent method for determining the value of  $i$ ; it was found to be close to the values found by other methods.

## 7. TRANSPORT PHENOMENA IN POLYELECTROLYTE SOLUTIONS

7.1 Equilibrium studies based on the colligative properties of polyelectrolyte systems led us to the comprehension of some general characteristics, such as those expressed by  $\phi_p$  and related parameters. They do not lead, however, to a deeper insight into the specific properties of individual polyelectrolytes, do not teach us about the shape of the poly-molecule and its dynamics, about its structural variability or its participation in biological organization. Such insight can be obtained in principle from study of the transport processes, which not only can yield richer information, but are generally more precise and easier to study than equilibrium processes.

From the very start, viscometric studies<sup>48</sup> and investigations on the diffusional behaviour of charged polymers<sup>49</sup> indicated that in many polyelectrolytes, the molecules are dynamic and undergo a conformational change upon ionization. Some of the studies on the conformational changes in biopolymers<sup>50</sup> and their expression in irreversible transport phenomena were reviewed recently<sup>61</sup> and will not be considered here. In the following paragraphs we shall consider some of the data concerning synthetic polyelectrolytes, based primarily on a non-equilibrium thermodynamic analysis of the data.

It is well known that the conductance of electrolyte solutions can be measured experimentally most simply and precisely. The equivalent conductance of polyacrylic and polymethacrylic acid solutions as a function of the degree of ionization,  $\alpha$ , and of concentration, was studied by Kern, by Wall and by Eisenberg<sup>51</sup>. It was found that upon increasing  $\alpha$  the equivalent conductance  $\Lambda$  does not remain constant, as was expected, but decreases appreciably. This finding is illustrated in *Figure 14* taken from Eisenberg's paper. To be sure,  $\Lambda$  depends also on the nature of the counterion, but when comparing the conductance of two polyelectrolyte salts,  $\Lambda_{c_1}$  and

$A_{c2}$ , Eisenberg found that the ratio of this difference to the difference of the counterion conductances, i.e.

$$\gamma = (A_{c1} - A_{c2}) / (A_{c1}^0 - A_{c2}^0) \quad (91)$$

is a characteristic parameter independent of the nature of the counterion, and determined by the degree of ionization.

7.2 A first interpretation of the findings presented in *Figure 14* can be made on the basis of the simple association theory of Wall<sup>51</sup>. This theory, based on

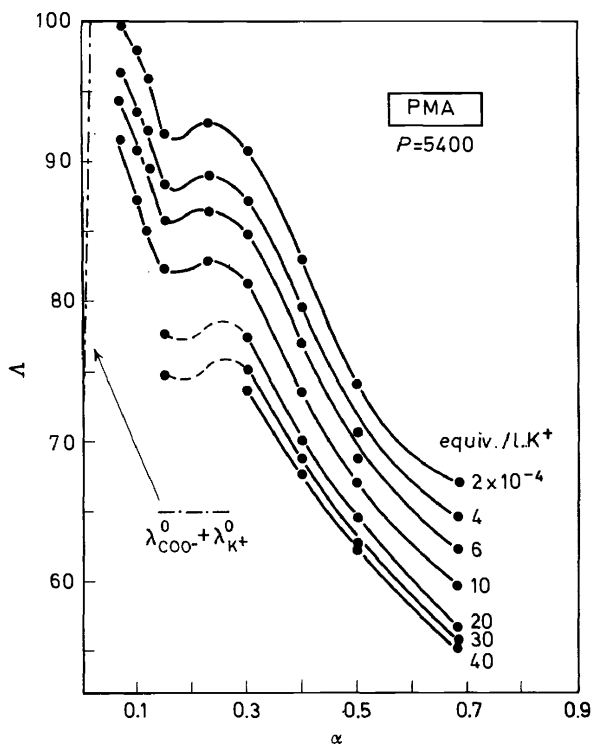


Figure 14. Dependence of conductance on degree of ionization, at 25°C (from Eisenberg<sup>51</sup>)

Wall's classical experiments on diffusion, self-diffusion and conductance in polyelectrolytes, assumes that a major part of the counterions is associated with the polyion—the fraction of the free ions being  $f$ , and that of the associated ions  $1 - f$ . The evaluation of  $f$  is now carried out theoretically in the following way.

The flow of electrical current  $I$  through the polyelectrolyte is carried by the flow of the counterions  $J_c$  and that of the polymeric ions,  $J_p$ . Since a mole of counterions carries one Faraday ( $F$ ), while a mole of negative polyelectrolyte carries  $-vF$  Faradays, the current may be written

$$I = (J_c - vJ_p) F \quad (92)$$

POLYELECTROLYTES

Now, the flux of any species is the product of the concentration ( $c$ ) and the velocity ( $\vec{v}$ ), so that

$$J_c = c_c \vec{v}_c \quad \text{and} \quad J_p = c_p \vec{v}_p \quad (93)$$

In a salt-free solution, the concentration of the counterions is  $\nu c_p$ , but according to Wall, only the fraction  $f$  of these ions contributes to the conductance, so that  $c_c = \nu c_p f$ . The velocity  $\vec{v}_c$  is, as usual, proportional to the electrical field  $E$  and to the mobility of free ions  $u_c^0$ , or,  $\vec{v}_c = u_c^0 E$ . In the case of the polymeric ions, the velocity is influenced by the fact that the total charge is screened by the associated counterions. Thus the effective charge number is not  $\nu$  but  $\nu f$ , and hence the polymer velocity is given by its mobility  $u_p$  multiplied by the effective field  $-\nu f E$ , or  $\vec{v}_p = u_p(-\nu f) E$ . Inserting these expression into equations 93 and 92, we find that

$$I = f \nu c_p (u_p + u_c^0) E \quad (94)$$

and hence the equivalent conductance is given by

$$\Lambda = I / \nu c_p E = f (u_p + u_c^0) F = f (\Lambda_p + \Lambda_c^0) \quad (95)$$

where  $\Lambda_p = u_p F$  and  $\Lambda_c^0 = u_c^0 F$ . Since in equation 95,  $u_c^0$  is well known and  $u_p$  can be readily determined electrophoretically, measurement of  $\Lambda$  provides an easy way for the evaluation of the association factor,  $f$ .

Indeed, equation 95 provides also a simple interpretation of Eisenberg's  $\gamma$ . Since the mobility of the polyion is practically independent of the nature of the counterion, and since the electrostatic association should also be insensitive to the specific properties of the small ions, we may write  $\Lambda_{c_1} = f(\Lambda_p + \Lambda_{c_1}^0)$ ,  $\Lambda_{c_2} = f(\Lambda_p + \Lambda_{c_2}^0)$  and hence

$$\gamma = (\Lambda_{c_1} - \Lambda_{c_2}) / (\Lambda_{c_1}^0 - \Lambda_{c_2}^0) = f \quad (96)$$

The equality of  $\gamma$  and  $f$  is a reasonable approximation, although the differences, as shown in Table 2, are beyond the limits of the experimental error. On the other hand, the values of  $f$  determined conductometrically and evaluated by Wall *et al.*<sup>51</sup> also from self diffusion, are practically identical.

Table 2

$\alpha$	$f$ diffusion	$f$ transference	$\gamma$	$\phi_p$
0.096	0.92	0.90	0.70	0.55
0.24	0.79	0.73	0.62	0.40
0.41	0.62	0.58	0.60	0.28
0.615	0.51	0.45	0.56	0.21
0.815	0.39	0.38	0.50	0.16
0.98	0.38	0.38	0.40	0.14

Fractions  $f$  from transference and diffusion experiments (after Huizenga *et al.*<sup>51</sup>) for Na polyacrylate at  $m_m = 0.015$ , and fraction  $\gamma$  calculated by equation 91 from increments of ionic conductances (after Eisenberg<sup>51</sup>) for Na polymethacrylate and polyacrylate.

7.3 There is, however, a large and baffling difference between the value of  $f$  and that of  $\phi_p$ . *A priori*, one would tend to believe that the number of ions contributing to the osmotic pressure ( $\nu \phi_p$ ) should equal the number of 'free'

ions, as determined by transport methods ( $\nu f$ ). We find, however, that at higher degrees of ionization of the acrylates the percentage of counterions determining the colligative properties is about fourteen, while the corresponding percentage of free counterions, as evaluated by conductance measurements, is 38 to 40.

A rather naive way to resolve this contradiction would be to apply Onsager's idea that up to  $\lambda = 1$ , all the counterions are moving freely in solution, although some accumulation around the polyion is taking place in the form of a diffuse atmosphere. Increasing the degree of ionization, so as to make  $\lambda > 1$ , leads to a 'condensation' of the fraction

$$(\lambda - 1)/\lambda = 1 - 1/\lambda \tag{97}$$

while the fraction  $1/\lambda$  of the counterions remain in free solution\*.

If we assume that the transport phenomena take into account *all* the non-condensed ions, then in highly dilute solutions  $f \simeq 1/\lambda$ . But as shown above (equation 68), in the range of 'condensation',  $\phi_p \simeq 1/2\lambda$ , so that

$$f \simeq 2\phi_p \tag{98}$$

This approximation means that the colligative properties reflect the partial immobilization of the counterions both in the condensation or Bjerrum layer and in a diffuse atmosphere; on the other hand, transport properties depend on all the counterions except those 'fixed' in the condensation zone. The relation 98 is expected to hold for  $\phi_{p,s}$  lower than or equal to one half, while for higher  $\phi_{p,s}$  and  $\lambda$ s smaller than unity,  $f \simeq 1$ .

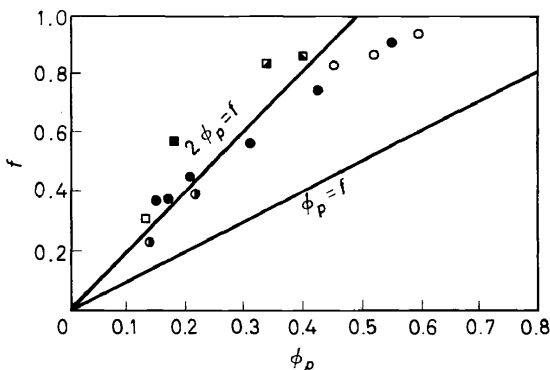


Figure 15. The interrelation between  $f$  and  $\phi_p$  (following Alexandrowicz and Danie<sup>49</sup>). (●) Na polyacrylate at neutralization degrees 0.096, 0.24, 0.41, 0.615, 0.815 and 0.98 [ $f$  from transport of current,  $\phi_p$  osmotic]; (○) Na albuminate with 10, 20 and 27 charges [ $f$  from transport of current];  $\phi_p$  osmotic; (⊙) Na and TMA polyphosphate [ $f$  from transport of current,  $\phi_p$  osmotic]; (■) polylysine hydrochloride [ $f$  from sedimentation]; (□) Na polymethacrylate [ $f$  from sedimentation;  $\phi_p$  osmotic], (◐) Na carboxymethylcellulose degree of substitution *ca.* 0.7 [ $f$  from sedimentation;  $\phi_p$  osmotic]; (◑) Na polyglutamate [ $f$  from sedimentation and  $\phi_p$  estimated from the Donnan exclusion factor  $\Gamma$  at low ionic strengths]

\* NOTE: Denoting the charge number at the point of condensation by  $\nu_c$ ,  $\lambda_c = 1 = \nu_c \epsilon^2 / DhkT$ . Thus  $(\lambda - 1)/\lambda = (\nu - \nu_c)/\nu$  which is evidently the fraction of the counterions which have undergone condensation.

## POLYELECTROLYTES

The validity of equation 98 can be tested on a larger number of data assembled by Alexandrowicz and Daniel<sup>49</sup>; as seen in *Figure 15* this rough prediction holds rather well.

7.4 The rough agreement of equation 98 with the experimental values of  $f$  should not lead to the hasty conclusion that all the counterions in the Bjerrum condensation layer are immobilized and do not contribute to conductivity. Such a conclusion would contradict the finding that no site binding could be discerned with monovalent alkali counterions nor is it in agreement with the results of the studies on the dielectric dispersion of polyelectrolyte solutions.

Since the 'fifties, it is known from the work of Heckmann, of Schindewolf and of Jacobson<sup>52</sup>, that orientation of polyelectrolyte molecules in a field of flow leads to a pronounced anisotropy of conductance. This effect is clearly related to the observation of Eigen and Schwarz<sup>53</sup> that the conductance of stretched polyions becomes anisotropic in an alternating electrical field. These phenomena were interpreted theoretically by Schwarz<sup>54</sup>, who attributed the polarizability to that of the atmosphere, in concordance with the conclusion of O'Konski on the conductivity of the counterions along the surface of the polyion<sup>55</sup>. From their measurements on polyphosphates, Eigen and Schwarz deduced that the polarizability of these compounds in dilute solutions may reach the enormous value of  $10^{-12}$  cm<sup>3</sup>, about  $10^{10}$  times larger than the regular value for low molecular compounds.

More recent studies of Mandel<sup>56</sup> in fields of higher frequency (5 to 8 KHz) led to an evaluation of dielectric behaviour in terms of a model for a polarizable rodlike molecule. During the last few years, Sachs<sup>57</sup> measured in this laboratory the dielectric dispersion in the region of very high frequencies between 0.5 and 100 MHz. In this region, orientation effects due to rotary diffusion of the polarized macromolecules vanish and it is possible to study the pure response of the atmosphere. In corroboration, in the range of megahertz frequencies, the dielectric dispersion is independent of the degree of polymerization and when  $v\phi_p$  becomes constant the dispersion becomes independent also of the degree of ionization. The new absorption peak, discovered in these measurements, shows a spectrum of relaxation times which could be theoretically explained by assuming that the counterions are not all fixed, and although restricted in movement perpendicular to the axis of the polyion, can move relatively freely on equipotential surfaces parallel to the surface of the polymeric cylinder. These conclusions, derived from studies of polyacrylic acid, polystyrene sulphonates and polyvinylsulphonates with counterions comprising H<sup>+</sup>, Li<sup>+</sup>, Na<sup>+</sup>, K<sup>+</sup>, Cs<sup>+</sup> and a couple of alkyl ammonium ions, support strongly the view that *all* the counterions have to be considered in constructing a consistent model for the conductance of polyelectrolytes.

To sum up, it is plausible to assume that although part of the counterions are sufficiently strongly attached to the polyions as to 'condense' and lose part of their mobility perpendicular to the 'surface' of the macromolecule, they retain their capacity to move parallel to the axis of the polyelectrolyte molecules, and thus contribute both to their strong polarization in an electric field and to the transport phenomena.

If all the counterions may move with relative freedom in an electric field,

it can be expected that the two basic effects of low molecular electrolytes, the electrophoretic and relaxation effects, will also be exhibited by polyelectrolyte solutions. As is well known, the relaxation effect is an expression of the fact that the effective field ( $E - \Delta E$ ) acting on an ion is a resultant of the external field  $E$  and a local retarding field  $\Delta E$ , which is created by all other ions in the vicinity. The electrophoretic effect is due to the hydrodynamic drag caused by the relative movement of the solvent molecules in the vicinity of the ions under consideration. Moller *et al.*<sup>58</sup> expressed both effects in the following simple equation

$$u_c = (1 - \Delta E/E)u_c^0 - \Delta u_{\text{electroph.}} \quad (99)$$

where  $u_c$  is the mobility of the counterion and  $u_c^0$  is its free mobility;  $\Delta u_{\text{electroph.}}$  is the contribution of the electrophoretic effect, which depends only on the structure of the atmosphere and is independent of the nature of the counterion. Thus from equation 99 we may derive the conclusion that the difference in the mobilities of two counterions of the same polyelectrolyte, and at the same degree of polyion ionization, can be expressed by the equation

$$\frac{u_{c_1} - u_{c_2}}{u_{c_1}^0 - u_{c_2}^0} = \frac{A_{c_1} - A_{c_2}}{A_{c_1}^0 - A_{c_2}^0} = \gamma = 1 - \frac{\Delta E}{E} \quad (100)$$

Thus equation 100 provides a new interpretation for the factor  $\gamma$  and relates it to the relaxation effect.

For the theoretical evaluation of  $\gamma$  we shall now take the opposite view to that inherent in equation 98—i.e. we shall assume that only the fraction  $\phi_p$  of the counterions is free to move in all directions, while all  $1 - \phi_p$  are within an atmospheric shell which confines their movement to equipotential surfaces parallel to the axis of the polyion. Moreover, the mobility assigned to the counterions will be  $u_c^0$  of the free counterions. Consider now a

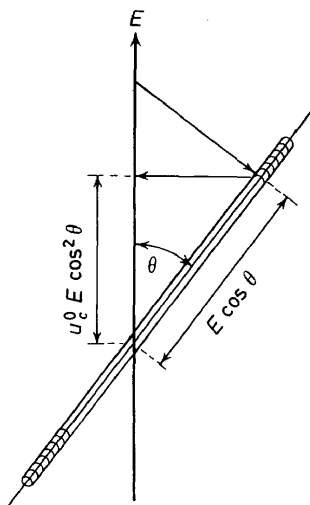


Figure 16



POLYELECTROLYTES

counterion in the atmosphere of a polyion which makes an angle  $\theta$  with the direction of the field  $E$ . Evidently, the field component in the atmosphere is  $E \cos\theta$  which causes the counterions to move along the polyion with a velocity  $\vec{v}_{\parallel} = u_c^0 E \cos\theta$ . The contribution of this velocity in the direction of the field itself is, however,  $\vec{v} = \vec{v}_{\parallel} \cos\theta = u_c^0 E \cos^2\theta$ . The average contribution of all counterions in the atmosphere to the electric flow is obtained by integrating over all values of  $\theta$ , i.e.

$$u_c^0(1 - \phi_p)EF \int_0^{\pi} \cos^2\theta \frac{\sin\theta d\theta}{2} = \frac{1}{3}u_c^0(1 - \phi_p)EF \quad (101)$$

On the other hand, the contribution of the 'free ions' per ion is clearly

$$u_c^0 \phi_p EF \quad (102)$$

so that the total counterion flow, unhampered by the electrophoretic effect, is

$$\frac{1}{3}u_c^0(1 - \phi_p)EF + u_c^0 \phi_p EF = \frac{1}{3}(1 + 2\phi_p)u_c^0 EF \quad (103)$$

To evaluate the electrophoretic effect we shall again use a rough estimate and assume that all the 'atmospheric' counterions are in the field of flow of the polymeric ions, which moves with a velocity  $u_p E$  and which contributes to the flow per counterion

$$-(1 - \phi_p)u_p EF \quad (104)$$

Combining equations 103 and 104 and comparing with equation 99, we find

$$u_c = \frac{1}{3}(1 + 2\phi_p)u_c^0 - (1 - \phi_p)u_p = \gamma u_c^0 - \Delta u_{\text{electroph.}} \quad (105)$$

from which one obtains

$$\gamma = 1 - \Delta E/E = \frac{1}{3}(1 + 2\phi_p) \quad \text{or} \quad \phi_p = \frac{1}{2}(3\gamma - 1) \quad (106)$$

Despite the crude approximations, in numerous cases the experimental validity of equation 106 was found to hold remarkably well. Thus Nelson and Ander<sup>59</sup> studied the electrical conductivities of salts of arabic acid and found that at high dilutions  $\gamma = 0.42$ , from which  $\phi_p = 0.13$  in good agreement with the experimental value  $\phi_p = 0.15$ . For polymethacrylates we have used the  $\gamma$  values of Eisenberg<sup>51</sup> and compared them through equation 106 with the  $\phi_p$ s of Alexandrowicz. The overall agreement is tolerable, as shown in Table 3.

Table 3

$\alpha =$	0.1	0.24	0.41	0.615	0.815	0.98
$\phi_p^{\text{calc}} = \frac{1}{2}(3\gamma - 1)$	0.55	0.43	0.40	0.34	0.25	0.10
$\phi_p^{\text{meas}}$	0.55	0.40	0.28	0.21	0.16	0.14

Since the model discussed in this paragraph does not imply association it will be clear that the equivalent conductance will not imply  $f$  explicitly but will be written in the conventional manner

$$\Lambda = (u_p + u_c)F \quad (107)$$

which upon inserting  $u_c$  from equation 105 is equal to

$$A = (\gamma u_c^0 + \phi_p u_p) F = \gamma A_c^0 + \phi_p A_p \quad (108)$$

Comparing equations 108 and 95 we find a new expression for  $f$

$$f = (\phi_p A_p + \gamma A_c^0) / (A_p + A_c^0) \quad (109)$$

The usefulness of equation 109 was tested by Dolar *et al.*<sup>60</sup> on the hydrogen form and sodium salts of polystyrene sulphonic acid at different concentrations. Their data are represented in *Table 4*.

*Table 4*

$C_m \times 10^3$ mole/kg water	$\gamma$	$\phi_p$	$f_{calc}$	$f_{expt}$
92.17	0.5415	0.235	0.515	0.509
42.18	0.4865	0.20	0.45	0.458
25.99	0.4615	0.17	0.42	0.435
5.156	0.3910	0.16	0.36	0.379

We are now in the baffling situation that both equations 98 and 109 describe fairly well part of the experimental data. To be sure a more stringent test, based on more extensive measurements, would be needed to choose between the alternatives. But also from a theoretical point of view it is advantageous to follow a more rigorous approach of a phenomenological nature, which would provide a suitable framework for the selection of the best model. Such a framework is provided by the thermodynamics of non-equilibrium processes in polyelectrolyte solutions.

## 8. NON-EQUILIBRIUM THERMODYNAMIC TREATMENT OF TRANSPORT IN POLYELECTROLYTE SOLUTIONS<sup>61,3</sup>

8.1 The phenomenological description of non-equilibrium thermodynamics requires a clear specification of the flows and their conjugate forces. Since we are concerned here with salt-free solutions, the only flows are those of the polyions  $J_p$  and of the counterions  $J_c$ . The driving forces are the negative values of the corresponding gradients of the electrochemical potentials  $\nabla(-\tilde{\mu}_p)$  and  $\nabla(-\tilde{\mu}_c)$ . Since the chemical potential of the polyelectrolytic salt is related to the electrochemical potential of the component ions by  $\mu_p = \tilde{\mu}_p + v\tilde{\mu}_c$ , we may write

$$\nabla\mu_p = \nabla\tilde{\mu}_p + v\nabla\tilde{\mu}_c \quad (110)$$

It is generally useful to consider the potentials per unit charge, so that for a polyacid, with monovalent cationic counterions,

$$\nabla\mu_p/v = \nabla\tilde{\mu}_p/v + \nabla\tilde{\mu}_c = \nabla\tilde{\mu}_- + \nabla\tilde{\mu}_+ \quad (111)$$

If we follow the same procedure and consider single charge flows, rather than molecular flows, evidently  $J_+ \equiv J_c$  while  $J_- \equiv vJ_p$ . The pheno-

menological equations of the thermodynamic description may then be written in a conventional manner :

$$\begin{aligned} J_- &= L_{11}\nabla(-\tilde{\mu}_-) + L_{12}\nabla(-\tilde{\mu}_+) \\ J_+ &= L_{21}\nabla(-\tilde{\mu}_-) + L_{22}\nabla(-\tilde{\mu}_+) \end{aligned} \quad (112)$$

where the coefficients  $L_{ij}$  fulfil the Onsager condition

$$L_{12} = L_{21} \quad (113)$$

Now these general equations can readily be adopted in important special cases, which allow the evaluation of the phenomenological coefficients. Indeed, since the number of independent coefficients is three, three independent transport methods are required for the evaluation of the coefficients.

The first method of choice is electrical conductance which is generally measured under conditions of equal concentration throughout the measuring cell. Since an electrochemical potential can be formally written at constant temperature and pressure as  $\tilde{\mu}_i = \mu_i^c + z_i F \psi$ , where  $\mu_i^c$  is the concentration dependent part and  $\psi$  the electrical potential, the constancy of the concentration ( $c$ ) means that  $\text{grad } \mu_i^c = 0$ , and hence

$$(\nabla \tilde{\mu}_i) = z_i F \nabla \psi \quad (114)$$

As is well known from electrostatics,  $\nabla \psi = -E$ , where  $E$  is the intensity of the electrostatic field; hence in a conductance experiment

$$\nabla(\tilde{\mu}_-) = -F \cdot E \text{ and } \nabla(-\tilde{\mu}_+) = F \cdot E \quad (115)$$

Inserting equation 115 into equation 112 and noting the definition of  $J_+$  and  $J_-$ , we obtain:

$$\begin{aligned} -vJ_p &= -J_- = (L_{11} - L_{12})FE \\ J_c &= J_+ = (L_{22} - L_{12})FE \end{aligned} \quad (116)$$

Equations 116 may be directly used for the evaluation of electrical flow in terms of the phenomenological coefficients. With the aid of equation 92 we find that

$$1 = (J_c - vJ_p)F = (L_{11} - 2L_{12} + L_{22})F^2E \quad (117)$$

and hence the specific conductance  $\kappa$  is given by

$$1/E = \kappa = (L_{11} - 2L_{12} + L_{22})F^2 \quad (118)$$

Equation 118 is, however, expressed in a non-conventional way and cannot be readily compared with textbook equations of the Kohlrausch type. It is therefore preferable to introduce regular mobilities and express the  $L_{ij}$ s in a more tangible form. For this purpose we turn to equation 93 and represent the velocities  $v_c$  and  $\bar{v}_p$  in terms of their corresponding mobilities. Since we do not want to follow the association theory—which transgresses the phenomenological thermodynamic analysis—it is necessary to write in a general way that  $\bar{v}_c = u_c E$ , or  $J_c = v c_p u_c E$  and  $\bar{v}_p = -u_p E$ , or

$$J_p = c_p u_p (-E) \quad (119)$$

Inserting equation 119 into equation 116 we find that the mobilities  $u_p$  and  $u_c$  may be expressed in terms of phenomenological mobilities:

$$\begin{aligned} u_p &= (L_{11} - L_{12})F/vc_p = u_{11} - u_{12} \\ u_c &= (L_{22} - L_{12})F/vc_p = u_{22} - u_{12} \end{aligned} \quad (120)$$

where

$$u_{ij} = L_{ij}F/vc_p$$

Thus equation 118 can be rewritten

$$\kappa = vc_p(u_{11} - 2u_{12} + u_{22})F \quad (121)$$

from which we derive the equivalent conductance  $A$

$$A = \kappa/vc_p = (u_{11} - 2u_{12} + u_{22})F \quad (122)$$

Equation 122 is the first equation for the evaluation of the three  $u_{ij}$ s. The second equation is derived immediately, since the transference numbers of the ions ( $t_i$ ) represent the fraction of the current carried by either the positive counterion or the negative polyion, i.e.

$$t_{+} = \frac{J_+F}{I} = \frac{u_{22} - u_{12}}{u_{11} - 2u_{12} + u_{22}} \quad \text{and} \quad t_- = \frac{-J_-F}{I} = \frac{u_{11} - u_{12}}{u_{11} - 2u_{12} + u_{22}} \quad (123)$$

For the third equation we shall make use of the diffusion of the polyelectrolyte.

In a regular diffusion experiment no electric current is flowing so that

$$I = 0, \quad \text{or} \quad J_+ = J_- \quad (124)$$

Here, however, the concentration is variable  $\therefore \nabla\mu_i \neq 0$  and hence the general equations 112 have to be used, although the  $L_{ij}$ s will now be written in terms of the  $u_{ij}$ s. It will be noted that equation 124 is equivalent to

$$u_{11}\nabla(-\tilde{\mu}_-) + u_{12}\nabla(-\tilde{\mu}_+) = u_{12}\nabla(-\tilde{\mu}_-) + u_{22}\nabla(-\tilde{\mu}_+)$$

or

$$\frac{u_{11} - u_{12}}{u_{11} - 2u_{12} + u_{22}} \nabla(-\tilde{\mu}_-) = \frac{u_{22} - u_{12}}{u_{11} - 2u_{12} + u_{22}} \nabla(-\tilde{\mu}_+)$$

which by equation 115 gives

$$t_- \nabla(-\tilde{\mu}_-) = t_+ \nabla(-\tilde{\mu}_+) \quad (125)$$

Adding to both sides of equation 125,  $t_- \nabla(-\tilde{\mu}_+)$ , and considering equation 111, and the requirement  $t_+ + t_- = 1$ , we obtain

$$t_- \nabla(-\mu_p)/v = \nabla(-\tilde{\mu}_+) \quad \text{and} \quad t_+ \nabla(-\mu_p)/v = \nabla(-\tilde{\mu}_-) \quad (126)$$

The diffusion flow is that of the polymeric ion  $J_p$ , which by equations 112 and 120 is given by

$$J_p = (c_p/F) [u_{11}\nabla(-\tilde{\mu}_-) + u_{12}\nabla(-\tilde{\mu}_+)]$$

or, inserting the gradients of the electrochemical potentials from equation 126.

$$J_p = \{(u_{11}t_+ + u_{12}t_-)/vF\}c_p \nabla(-\mu_p) \quad (127)$$

The definitions of  $\Lambda$ ,  $t_+$  and  $t_-$  give directly

$$(u_{11}t_+ + u_{12}t_-)/F = (u_{11}u_{22} - u_{12}^2)/\Lambda$$

Further, the thermodynamic equation of Gibbs and Duhem relates  $c_p \nabla(-\mu_p)$  to the solvent parameters by  $c_p \nabla(-\mu_p) + c_w \nabla(-\mu_w) = 0$  where  $c_w$  is the water concentration in the polyelectrolyte solution and  $\mu_w$  its chemical potential. Taking into account still another known relation  $\nabla(-\mu_w) = -\bar{V}_w \nabla(-\pi)$  and since  $c_w \bar{V}_w \approx 1$ , we find that  $c_p \nabla(-\mu_p) = \nabla(-\pi)$ . Finally since the osmotic pressure varies only through the change in polyelectrolyte concentration,  $\nabla(-\pi) = (d\pi/dc) \nabla(-c_p)$ , with all these transformations, equation 127 assumes a more familiar form

$$J_p = [(u_{11}u_{22} - u_{12}^2)/\Lambda v] (d\pi/dc_p) \nabla(-c_p) = D \nabla(-c_p) \quad (128)$$

where the diffusion coefficient is defined as

$$D = [(u_{11}u_{22} - u_{12}^2)/\Lambda v] (d\pi/dc_p) \quad (129)$$

Since in the salt-free case  $\pi \approx v c_p \phi_p RT$  and in dilute solutions  $\phi_p$  depends only slightly on concentration, it may be assumed that  $d\pi/dc_p = v \phi_p RT$  and hence the polyion diffusion coefficient is found to obey the following elegant relation

$$\Lambda D = (u_{11}u_{22} - u_{12}^2) \phi_p RT \quad (130)$$

which is the third equation for the  $u_{ij}$ s. It will be noted *en passant* that equation 130 can be reduced to the Nernst equation in the ideal uncoupled case when  $u_{12} = 0$  and  $\phi_p = 1$ .

From equations 122, 123 and 130 we find that :

$$u_{11} = \Lambda t_-^2/F + DF/\phi_p RT; \quad u_{22} = \Lambda t_+^2/F + DF/\phi_p RT$$

and

$$u_{12} = \Lambda t_+ t_- /F + DF/\phi_p RT \quad (131)$$

The  $u_{ij}$ s are the required thermodynamic parameters for our further analysis. If there should be enough experimental data for the evaluation of the  $u_{ij}$ s it would be possible to obtain a further insight into the nature of the transport processes according to the following reasoning.

It was shown above that the phenomenological equations 112 may be rewritten in a more convenient form which allows the explicit evaluation of the polyion velocity  $\vec{v}_p$

$$vJ_p = v c_p \vec{v}_p = (v c_p /F) [u_{11} \nabla(-\tilde{\mu}_-) + u_{12} \nabla(-\tilde{\mu}_+)]$$

or

$$\vec{v}_p = (1/F) [u_{11}\nabla(-\tilde{\mu}_-) + u_{12}\nabla(-\tilde{\mu}_+)] \quad (132)$$

Let us now carry out a *mental* experiment in which the direct force acting on the counterions  $\nabla(-\tilde{\mu}_+)$  is made to vanish, and the polyions are driven by their conjugate force only,  $u_{11} = F[\vec{v}_p/\nabla(-\tilde{\mu}_-)]$  at  $\nabla(-\tilde{\mu}_+) = 0$ . The fact that such an experiment cannot be carried out in practice is of no importance, since its significance is interpretative only; it enables us to recognize the significance of the straight mobility  $u_{11}$ . Since in this experiment no force acts on the counterions we may reason that  $u_{11}$  represents a mobility of the polyion unperturbed by the atmosphere. Moreover, we expect that  $u_{11}$  will increase linearly with the degree of ionization since the friction per charge goes down with  $\alpha$ , and hence

$$u_{11} = \alpha u_m^0 \quad (133)$$

Here  $u_m^0$  is the mobility of a monomeric segment, which for the cylindrical model moves together with the 'bound' ions. The validity of equation 133 was tested for polyacrylates and it was found that  $u_m^0 = 3.2 \times 10^{-3}(\text{cm/sec})/(\text{volt/cm})$  and is approximately constant. The most interesting aspect, from the present point of view, is the evaluation of the 'free ions' in the polyelectrolyte solution. Continuing our mental experiment in which  $\nabla(-\tilde{\mu}_+) = 0$  we may enquire, what is the velocity of the counterions under the influence of  $\nabla(-\tilde{\mu}_-)$  alone? It is readily seen that in this case  $\vec{v}_c = u_{12}\nabla(-\tilde{\mu}_-)$ , or that

$$(\vec{v}_c/\vec{v}_p) = u_{12}/u_{11} \text{ at } \nabla(-\tilde{\mu}_+) = 0 \quad (134)$$

Equation 134 represents the drag effect of the polyion on the small surrounding ions. It should be clear that  $\vec{v}_c$  is the average velocity of all the counterions and if we assume that the free ions do not participate in the polyion movement while all bound counterions—whatever be the mode of their attraction to the polyelectrolyte molecule—move at a velocity  $\vec{v}_p$ , then  $\vec{v}_c/\vec{v}_p$  measures the fraction of bound ions and

$$1 - \vec{v}_c/\vec{v}_p = 1 - u_{12}/u_{11} = f_1 \quad (135)$$

represents the fraction of the 'free' counterions.

Table 5. The fraction of 'free counterions'  $f_1$  and  $f_2$  from thermodynamic analysis of polyacrylate behaviour

$\alpha$	$f_1$	$\phi_p$	$f_2$	$\frac{1}{3}(1 + 4\phi_p)$
0.3	0.44	0.34	0.83	0.81
0.4	0.33	0.28	0.78	0.71
0.5	0.26	0.24	0.71	0.67
0.6	0.24	0.21	0.64	0.61
0.7	0.22	0.19	0.59	0.58
0.8	0.21	0.16	0.54	0.54

Table 5 presents the values of  $f_1$  and there is little doubt that they are sufficiently close to  $\phi_p$  as to permit the assumption that  $f_1$  is a measure of the 'truly' free ions. To be sure, the assumption that all counterions in the polyion atmosphere move at the same velocity  $\vec{v}_p$  is very crude, and a more detailed theory is required for the velocity distribution. Presumably, only the condensed counterions have a velocity  $\vec{v}_p$  while the velocity of the atmospheric ions drops gradually from  $\vec{v}_p$  to zero. The agreement of  $f_1$  with  $\phi_p$  is, however, close enough to justify the simplified assumption.

In analysing the phenomenological equations it is possible to make another mental experiment. This time, we allow both  $\nabla(-\tilde{\mu}_-)$  and  $\nabla(-\tilde{\mu}_+)$  to operate, but the forces are so chosen as to make the flow of the polymer zero. This means formally that :

$$\begin{aligned} 0 &= u_{11}\nabla(-\tilde{\mu}_-) + u_{12}\nabla(-\tilde{\mu}_+) \\ \vec{v}_c &= u_{12}(-\tilde{\mu}_-) + u_{22}\nabla(-\tilde{\mu}_+) \end{aligned} \tag{136}$$

From which we obtain

$$\left[ \frac{\vec{v}_c}{\nabla(-\tilde{\mu}_+)} \right]_{J_p=0} = u_{22} - \frac{u_{12}^2}{u_{11}} \tag{137}$$

Here, again we surmise that the average velocity  $\vec{v}_c$  is contributed only by the free ions, and since it is only the counterions which participate in the transport (because the polyions were immobilized!) we may attribute the whole velocity to a fraction  $f_2$  of the free ions with a mobility  $u_c^0$ —where  $u_c^0$  is the mobility of counterions in an infinitely dilute solution. We thus obtain

$$f_2 u_c^0 = u_{12} - u_{12}^2/u_{11} \quad \text{or} \quad f_2 = (u_{11}u_{12} - u_{12}^2)/u_{11}u_c^0 \tag{138}$$

Now, Table 5 shows that  $f_2$  is quite different from  $\phi_p$  indicating that in this mental experiment a much larger fraction of free ions participates in this transport phenomenon. Indeed,  $f_2$  is even larger than  $f = 2\phi_p$ —indicating that it is not only all the counterions outside the condensation layer, but also those in the condensation layer which participate.

It is rather difficult to see what determines the mobility of the counterions at  $\vec{v}_p = 0$ . We may assume, however, in an *ad hoc* manner, that all counterions outside the Bjerrum layer contribute fully and condensed counterions participate by polarization, and then the interpretation of  $f_2$  is as follows. The  $2\phi_p$  'relatively free' ions move 'without hindrance' while the  $1 - 2\phi_p$  ions in the condensation layer contribute also but their movement is confined to the equipotential surfaces parallel to the polyion axis. If the polyelectrolyte molecules or their larger segments are oriented at random, the contribution of the condensation layer to mobility is only a third of the total mobility, so that

$$f_2 \simeq 2\phi_p + \frac{1}{3}(1 - 2\phi_p) = \frac{1}{3}(1 + 4\phi_p) \tag{139}$$

Evidently, this holds only as long as  $\lambda > 1$ ; when  $\lambda < 1$  and  $\phi_p > \frac{1}{2}$ ,  $f_2 \rightarrow 1$ . The behaviour in the transport region is summarized in Table 5.

Despite the satisfactory agreement of  $f_2^{\text{meas}}$  with that calculated from

equation 139, the theoretical formulations are shaky; much more experimental and theoretical work will have to be done before the correct model for the thermodynamic coefficients can be calculated satisfactorily.

## 9. POLYELECTROLYTE-POLYELECTROLYTE INTERACTION

9.1 A suitable topic for discussion at this late point are the mutual interactions between polyelectrolytes to form complexes of great biological importance and some practical interest. Already during the first stages of polyelectrolyte research, Fuoss and Sadek<sup>62</sup> observed that the polyacids and polybases may coprecipitate at a critical concentration ratio. This finding is in accord with the classical studies of Bungenberg de Jong on complex coacervation of oppositely charged biocolloids<sup>63</sup>, studies which exerted a powerful influence on the biophysical outlook of the previous generation. Our own studies<sup>64</sup> on synthetic polyelectrolytes corroborated the data of Fuoss and Sadek and supplemented the additional fact the

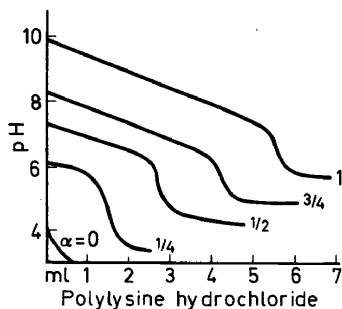


Figure 17. Potentiometric titration of 10 ml polymethacrylic acid ( $10^{-3}$  molar) of different degrees of ionization ( $\alpha = 0, 0.25, 0.5, 0.75,$  and  $1.0$ ) by  $2 \times 10^{-3}$  molar solutions of salt-free polylysine hydrochloride (ref. 64)

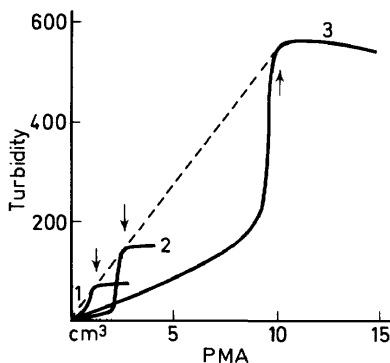


Figure 18. Turbidimetric titration of 20 ml of three polylysine solutions: (1)  $1 \mu\text{g/ml}$ ; (2)  $2 \mu\text{g/ml}$ , and (3)  $8 \mu\text{g/ml}$  by a  $10\text{M}^{-3}$  solution of polymethacrylic acid. The initial pH in all three cases was 6.8 (ref. 64)



precipitation occurs when the total number of the positive groups provided by the polybase ( $=\beta c_b$ , where  $c_b$  is the monomolar concentration of the polybase and  $\beta$  its degree of ionization) equals the number of negative groups fixed on the polyacid ( $=\alpha c_a$ ). This is readily seen in *Figure 17*, where the pH of polymethacrylic acids of different degrees of ionization is given as a function of the amount of added polybases. The polybase was a polylysine which is fully ionized in the experimental pH range. It is seen that the very addition of polylysine lowers the pH through the screening effect of the positive polymer. At the electrochemical equivalence point, there is, however, a marked drop in pH with concomitant precipitation. The turbidimetric change accompanying precipitation is so sensitive that it was developed by Terayama<sup>65</sup> into an analytical method for polyelectrolyte titrations.

A more detailed theory of the phase equilibria involved in polyelectrolyte interactions was developed by Overbeek and Voorn<sup>66</sup>, and applied to biological systems by A. Veis<sup>67</sup>.

Recently, the polyelectrolyte complexes began to find their way into membrane technology as hydrogels for selective ultrafiltration. The interactions of some strong polyacids and polybases when mixed in an ion-equivalent ratio produce strong and durable membranes of high selectivity. Thus the mixture of the sulphonates of polystyrene or polyethylene with polyvinyl-benzyltrimethyl ammonium chloride give stable and strong films with a remarkably high water permeability—about ten times higher than that of cellophane of equal thickness—and pronounced selectivity for the transport of small molecules. Such films are promising for desalination by reverse osmosis and are already in use for selective filtration of organic molecules<sup>68</sup>.

Ultrafine films of polyacid–polybase complexes may be prepared by an ingenious technique developed by Shashoua<sup>69</sup>. If a drop of polybase is spread on the surface of a polyacid solution, a bimolecular film is formed which can be transferred to a mechanical support and used as a membrane of molecular thickness. Such duplex films have the rectifier properties of electrical bilayers and when ‘excited’ by an electrical current of suitable intensity develop a characteristic ‘firing’ pattern which strikingly resembles the firing of nerve membranes. A theory for this firing was worked out by Katchalsky and Spangler<sup>70</sup> and was tested experimentally by Shashoua.

9.2 Among the polybase–polyacid complexes, specially interesting are the interaction products of DNA with basic polyamino acids. The study of these complexes has a bearing on our interpretation of the forces involved in the organization of chromosomal structures based on the interaction of nucleic acids with protamines or histones.

Since this paper is not a review article, I shall consider here only one piece of research, namely that carried out by Felsenfeld<sup>71</sup> on the interaction of DNA with polylysine in 0.9 to 1 N NaCl solutions. An aggregate phase is readily formed which can be separated by centrifugation. The ratio of DNA to polylysine is one to one, and it is strongly indicated that the polybase lies in the groove of the double helix in such a manner that the charged ammonium groups are attached to the phosphates of the nucleic acid. The stereospecificity of the reaction is seen in the fact that poly-D-lysine or poly-L-lysine give more precipitation with DNA than poly-D,L-lysine. If the

DNA is of heterogeneous composition, the fraction richest in A-T pairs gives the highest degree of complexation.

The most remarkable finding is that the light scattering data seem to indicate that the complexes have a relatively sharp particle size distribution, their diameter is close to 1700 Å, independent of the size of the DNA chains. It seems that the particles of the complex have a ring form and the diameter evaluated from turbidity is that of a circular structure. Here, complex formation leads to a structural feature which may be related to that underlying the organization of circular chromosomes in bacteria, or other structural elements of the cell.

## 10. CONCLUDING REMARKS

10.1 Within the limited framework of a lecture, even if transcribed into a lengthy paper, there is no possibility to cover all interesting aspects of polyelectrolyte chemistry. Thus, I have been forced to omit some important equilibria phenomena such as the potentiometric titration of weak polyacids and polybases; or the discussion of polyelectrolyte viscosity from the viewpoint of irreversible phenomena. Despite the fundamental information provided by viscosity on the shape of the polyions, its dependence on ionic strength and degree of ionization, it was obligatory to forego its discussion because of lack of time. Similarly we have to postpone to a more comprehensive review the interesting studies on the reactivity of polyelectrolyte molecules and on the influence of polyelectrolytes on reaction kinetics. Here we would like to pay attention only to one additional phenomenon related to metastable configurations of polyelectrolytes and its bearing on molecular memory.

10.2 It will be observed that throughout the previous paragraphs we assumed that the ionization of the polyions exerts only a secondary effect on the conformation of the polyelectrolyte molecules. The macromolecules were regarded as partially stretched cylindrical filaments the lengths of which could be further stretched by increased ionization—phase transitions were, however, excluded from our consideration. There is of course no doubt that many biopolymers undergo chemical melting and the study of their phase transitions is of major importance for the characterization of natural polyelectrolytes. This fundamental aspect will, however, be left to the discussion of biopolymer behaviour while these concluding paragraphs will be devoted only to certain metastable conformational transitions, discovered for the first time in the potentiometric titration of ribosomal RNA. The work of Peacocke and Cox<sup>72</sup> showed that the titration of RNA gives a different branch when titrated from neutral to acid pH, than that obtained in the back titration from the acid pH to the neutral starting point. This titration behaviour is time-independent and it is possible to move around the hysteresis cycle any number of times with reproducible results. A more convenient model is that of certain polynucleotide complexes, in particular the triple stranded complex of poly A with two poly U molecules. The hysteresis behaviour of these complexes is shown in potentiometric and spectrophotometric acid-base titrations<sup>73</sup>.

In the acid titration of the complex, from pH 7 to pH 3 it dissociates and

POLYELECTROLYTES

protonated double helical poly A·poly A and single stranded poly U are formed. Upon titrating backwards the triple stranded complex is reformed, but while its dissociation occurs abruptly and irreversibly at a pH of  $\sim 3.5$ , at  $20^\circ\text{C}$  and in  $0.1\text{ N}$  NaCl solution the reassociation takes place gradually

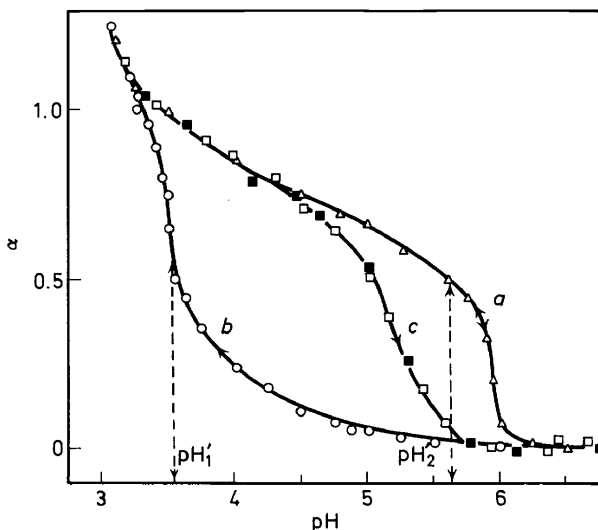


Figure 19. Potentiometric titration ( $\alpha$ , degree of protonation as a function of pH) at  $20^\circ\text{C}$ , (a) of an  $0.1\text{ M}$  NaCl solution of poly A and poly U, mixed in the molar ratio 1:2.  $\circ$ , acid titration (b),  $\square$ , subsequent base titration (c), and (b) of an  $0.1\text{ M}$  NaCl solution of poly A,  $\Delta$ . curve A: Concentrations of polymer (residue) at pH 7: (a)  $c = 3.24 \times 10^{-4}\text{ M}$  (A·2U) and (b)  $c = 3.2 \times 10^{-4}\text{ M}$  (A) [see ref. 73]

along an equilibrium path between pH 4 and pH 5.5. The appearance of a time-independent hysteresis loop is an indication for the existence of long-lived metastable configurations, which are stabilized by energetic barriers preventing transitions. This is shown in Figure 20, which describes the non-

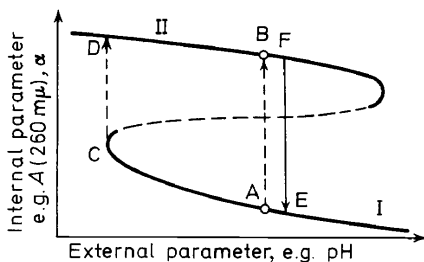


Figure 20. Hysteresis behaviour of a cooperative system domain, resulting from the existence of metastability. State I, poly A·2 poly U; state II, (poly A·poly A) $\text{H}^+$  and poly U; EF equilibrium transition; EC, range of metastability of state I; CD, non-equilibrium transition I  $\rightarrow$  II; AB, non-equilibrium transition induced by electric field pulses [see ref. 74]

equilibrium transition  $C \rightarrow D$  at about pH 3 and the equilibrium transition  $F \rightarrow E$  around pH 5<sup>74</sup>.

From a more general point of view, hysteresis is a memory device since the position of a state point on the hysteresis loop depends on the path and on its history. It is well known that the hysteresis of magnetization and demagnetization has been used for many years in computer memory devices. Magnetic hysteresis is, however, based on larger crystalline domains, while the phenomena described above are single molecule properties observable in dilute solutions of RNA or polynucleotide complexes. Here the domains are intramolecular patches of crystallinity, the phase transitions of which are controlled by the polyion electrostatic field, which determines also the barriers of metastability. In a small number of molecules a large amount of information could be recorded with a small expenditure of energy and it is intriguing to speculate whether such metastable states of bio-polyelectrolytes do not play a role in the accumulation of physical memory in living organisms. Recently, evidence was adduced that the incipient step of short-lived memory in living organisms is based on a conformational change of biopolymers<sup>75</sup> so that the study of macromolecular hysteresis may contribute to our understanding of memorizing in terms of metastable configurations in macromolecules<sup>76, 77</sup>.

To be sure, biological memory should be able to record the electrical pulses carried by the nerve membranes. These pulses are of the order of 100 mV, but since the thickness of the membrane is  $\sim 100 \text{ \AA}$  the electrical fields are about 100 kV/cm. A few months ago E. Neumann and I<sup>74, 77</sup> discovered that when pulses of 15 to 20 kV/cm are applied for 1 to 5  $\mu\text{sec}$  to protonated triple helical poly A $\cdot$ 2 poly U, it undergoes a transition to poly A $\cdot$ poly A and poly U at constant pH. These pulses remove partially the counterion atmosphere surrounding the polyelectrolyte complex which causes the complex to dissociate.

With these newer aspects of polyelectrolyte research we are leaving the secure realm of physicochemical considerations and allowing our charged polyions to enter the wide and difficult area of general biophysics.

## REFERENCES

- <sup>1</sup> *Colloid Science*, Vol. I (1952), Vol. II (1949), ed. H. R. Kruyt, Elsevier: Amsterdam.
- <sup>2</sup> S. A. Rice and M. Nagasawa, *Polyelectrolyte Solutions*. Academic Press: New York (1961); U. P. Strauss, 'Counter ion binding by macroions, in *Electrolytes*, p 215. Pergamon: London (1962); A. Katchalsky, Z. Alexandrowicz and O. Kedem, 'Polyelectrolyte solutions' in *Chemical Physics of Ionic Solutions*. B. E. Conway and R. G. Barradas, eds., p 295. Wiley: New York (1966); J. A. Marinsky, 'Interpretation of ion exchange phenomena' in *Ion Exchange*, Vol. I, p 353. Marcel Dekker: New York (1966); R. W. Armstrong and U. P. Strauss, 'Polyelectrolytes', in *Encyclopedia of Polymer Science and Technology*, Vol. X, p 781. Wiley: New York (1969).
- <sup>3</sup> The approach of this paragraph is based on the review of A. Katchalsky, Z. Alexandrowicz and O. Kedem, 'Polyelectrolyte solutions', in *Chemical Physics of Ionic Solutions*, B. E. Conway and R. G. Barradas, eds. p 295. Wiley: New York (1966).
- <sup>4</sup> W. Kern, *Z. Phys. Chem. A*, **181**, 249 (1938); **184**, 197 and 304 (1939);

POLYELECTROLYTES

- W. Kern, *Makromol. Chem.* **2**, 279 (1948);  
 C. S. Chadwick and J. M. Neal, *J. Polymer Sci.* **28**, 355 (1958);  
 Z. Alexandrowicz, *J. Polymer Sci.* **40**, 91 (1959);  
 Z. Alexandrowicz, *J. Polymer Sci.* **43**, 337 (1960);  
 Z. Alexandrowicz, *J. Polymer Sci.* **56**, 115 (1962);  
 O. D. Bonner and J. R. Overton, *J. Phys. Chem.* **67**, 1035 (1963);  
 N. Ise and T. Okubo, *J. Phys. Chem.* **71**, 1287 (1967).  
<sup>5</sup> cf. P. Flory, *Principles of Polymer Chemistry*, p 283. Cornell University Press: Ithaca, N.Y. (1953).  
<sup>6</sup> Z. Alexandrowicz, *J. Polymer Sci.* **40**, 91 (1959).  
<sup>7</sup> The cell model: an extensive discussion may be found in J. O. Hirschfelder, C. F. Curtiss and R. B. Bird, *The Molecular Theory of Gases and Liquids*. Wiley: New York (1954); applications to polymer solutions, cf. I. Prigogine, *The Molecular Theory of Solutions*, p. 331. North Holland: Amsterdam (1957).  
<sup>8</sup> L. Onsager, *Ann. N.Y. Acad. Sci.* **51**, 627 (1949).  
<sup>9</sup> J. J. Hermans and J. Th. G. Overbeek, *Rec. Trav. Chim. Pays-Bas*, **67**, 761 (1948);  
 F. T. Wall and J. Berkowitz, *J. Chem. Phys.* **26**, 114 (1957);  
 S. Lifson, *J. Chem. Phys.* **27**, 700 (1957);  
 N. Ise and M. Hosono, *J. Polymer Sci.* **39**, 389 (1959).  
<sup>10</sup> I. Langmuir, *J. Chem. Phys.* **6**, 873 (1938).  
<sup>11</sup> E. J. W. Verwey and J. Th. G. Overbeek, *Theory of Stability of Lyophobic Colloids*, p 92. Elsevier: Amsterdam (1948).  
<sup>12</sup> R. Marcus, *J. Chem. Phys.* **23**, 1057 (1955).  
<sup>13</sup> W. Kern, *Makromol. Chem.* **2**, 279 (1948).  
<sup>14</sup> M. Nagasawa and I. Kagawa, *J. Polymer Sci.* **25**, 61 (1957);  
 M. Nagasawa, M. Izumi and I. Kagawa, *J. Polymer Sci.* **37**, 375 (1959);  
 M. Nagasawa, A. Takahashi, M. Izumi and I. Kagawa, *J. Polymer Sci.* **38**, 213 (1959).  
<sup>15</sup> A. M. Liquori, F. Ascoli, C. Botrè, V. Crescenzi and A. Mele, *J. Polymer Sci.* **40**, 169 (1959).  
<sup>16</sup> N. Ise and T. Okubo, *J. Phys. Chem.* **69**, 4102 (1965); **70**, 2400 (1966); **71**, 1287 (1967); **72**, 1361, 1366, 1370 (1968); **73**, 4071 (1969).  
<sup>17</sup> D. Dolar and H. Leskovšek, *Makromol. Chem.* **118**, 60 (1968).  
<sup>18</sup> e.g. R. A. Robinson and R. N. Stokes, *Electrolyte Solutions*, 2nd ed., p 28. Butterworths: London (1959).  
<sup>19</sup> G. Manning, *J. Chem. Phys.* **51**, 924 (1969).  
<sup>20</sup> T. Alfrey, P. W. Berg and H. Morawetz, *J. Polymer Sci.* **7**, 543 (1951);  
 R. M. Fuoss, A. Katchalsky and S. Lifson, *Proc. Nat. Acad. Sci. Wash.* **37**, 579 (1951);  
 S. Lifson and A. Katchalsky, *J. Polymer Sci.* **13**, 43 (1953);  
 F. Oosawa, *J. Polymer Sci.* **23**, 421 (1957);  
 U. P. Strauss, *J. Am. Chem. Soc.* **80**, 6498 (1958).  
<sup>21</sup> H. Auer and Z. Alexandrowicz, *Biopolymers*, **8**, 1 (1969).  
<sup>22</sup> P. Chu and J. A. Marinsky, *J. Phys. Chem.* **71**, 4352 (1967).  
<sup>23</sup> I. Kagawa and H. P. Gregor, *J. Polymer Sci.* **23**, 477 (1957).  
<sup>24</sup> A. Katchalsky, R. Cooper, J. Upadhyay and A. Wasserman, *J. Chem. Soc.* 5198 (1961).  
<sup>25</sup> S. Oman and D. Dolar, *Z. Phys. Chem. (N.F.)* **56**, 1 (1967); **58**, 170 (1968).  
<sup>26</sup> D. Dolar and A. Peterlin, *J. Chem. Phys.* **50**, 3011 (1969).  
<sup>27</sup> D. Dolar and D. Kozak, *Proc. Leiden Symp. Vol. II*, p 363 (1970).  
<sup>28</sup> L. Kotin and M. Nagasawa, *J. Am. Chem. Soc.* **83**, 1026 (1961).  
<sup>29</sup> S. Bram and W. W. Beeman, *J. Molec. Biol.* **55**, 311 (1971).  
<sup>30</sup> J. W. Lyons and L. Kotin, *J. Am. Chem. Soc.* **87**, 1781 (1965).  
<sup>31</sup> Ch. Sander and P. O. P. Ts'o, *J. Molec. Biol.* **55**, 1 (1971).  
<sup>32</sup> I. Michaeli, *J. Polymer Sci.* **48**, 291 (1960).  
<sup>33</sup> A. Katchalsky and M. Zwick, *J. Polymer Sci.* **25**, 443 (1955).  
<sup>34</sup> C. S. Marvel, J. H. Sample and M. F. Roy, *J. Am. Chem. Soc.* **61**, 3241 (1939).  
<sup>35</sup> P. J. Flory, *J. Am. Chem. Soc.* **64**, 177 (1942);  
 F. T. Wall, *J. Am. Chem. Soc.* **64**, 269 (1942).  
<sup>36</sup> D. Mazia and A. Katchalsky, unpublished results.  
<sup>37</sup> J. Škerjanc, D. Dolar and D. Leskovšek, *Z. Phys. Chem. (N.F.)* **56**, 3 (1967).  
<sup>38</sup> E. J. W. Verwey and J. Th. G. Overbeek, *Theory of the Stability of Lyophobic Colloids*, p 56. Elsevier: Amsterdam (1948).  
<sup>39</sup> Z. Alexandrowicz, *J. Polymer Sci.* **43**, 337 (1960); **56**, 115 (1962); **56**, 97 (1962);

- A. Katchalsky and Z. Alexandrowicz, *J. Polymer Sci.* **A1**, 2093 (1963);  
 Z. Alexandrowicz and A. Katchalsky, *J. Polymer Sci.* **A1**, 3231 (1963);  
 L. Kotin and M. Nagasawa, *J. Chem. Phys.* **36**, 873 (1962).  
<sup>40</sup> I. Kagawa and K. Katsuura, *J. Polymer Sci.* **9**, 405 (1952).  
<sup>41</sup> R. A. Mock and C. A. Marshall, *J. Polymer Sci.* **13**, 263 (1954).  
<sup>42</sup> J. W. Lyons and L. Kotin, *J. Am. Chem. Soc.* **87**, 1670 (1965).  
<sup>43</sup> U. P. Strauss and P. Ander, *J. Am. Chem. Soc.* **80**, 6494 (1958).  
<sup>44</sup> U. P. Strauss, C. Helfgott and H. Pink, *J. Phys. Chem.* **71**, 2550 (1967).  
<sup>45</sup> Z. Alexandrowicz and A. Katchalsky, *J. Polymer Sci.* **A1**, 3231 (1963).  
<sup>46</sup> J. Marinsky, *Ion Exchange*, Vol. I, p 353. Marcel Dekker: New York (1966).  
<sup>47</sup> H. Inagaki, S. Hotta and M. Hirami, *Makromol. Chem.* **23**, 1 (1956);  
 H. Inagaki and M. Hirami, *Z. Elektrochemie*, **63**, 419 (1959);  
 T. Orofino, *Rec. Trav. Chim. Pays-Bas*, **78**, 434 (1959); cf. also ref. 39.  
<sup>48</sup> A. Katchalsky, O. Künzle and W. Kuhn, *J. Polymer Sci.* **5**, 283 (1950);  
 H. Eisenberg, *J. Polymer Sci.* **23**, 579 (1957);  
 J. Hermans Jr and J. J. Hermans, *Kon. Ned. Akad. Wet. Proc. Ser. B*, **61**, 325 (1958);  
 Z. Alexandrowicz, *J. Chem. Phys.* **47**, 4377 (1967);  
 I. Noda, T. Tsuge and M. Nagasawa, *J. Phys. Chem.* **74**, 710 (1970).  
<sup>49</sup> J. R. Huizenga, P. F. Grieger and F. T. Wall, *J. Am. Chem. Soc.* **72**, 2636 and 4228 (1950);  
 F. T. Wall, P. F. Grieger, J. R. Huizenga and R. H. Doremus, *J. Chem. Phys.* **20**, 1206 (1952);  
 O. Kedem and A. Katchalsky, *J. Polymer Sci.* **15**, 321 (1955);  
 Z. Alexandrowicz and E. Daniel, *Biopolymers*, **1**, 447 (1963);  
 E. Daniel and Z. Alexandrowicz, *Biopolymers*, **1**, 473 (1963);  
 G. S. Manning, *J. Chem. Phys.* **47**, 2010 (1967);  
 S. Lifson and J. L. Jackson, *J. Chem. Phys.* **36**, 2410 (1962);  
 J. L. Jackson, S. Lifson and S. R. Coriell, *J. Chem. Phys.* **50**, 5045 (1969).  
<sup>50</sup> *Biological Polyelectrolytes*, A. Veis, ed., Marcel Dekker: New York (1970).  
<sup>51</sup> W. Kern, *Z. Phys. Chem. A*, **181**, 249 (1938);  
 J. R. Huizenga, P. F. Grieger and F. T. Wall, *J. Am. Chem. Soc.* **72**, 2636 and 4228 (1950);  
 H. Eisenberg, *J. Polymer Sci.* **30**, 47 (1958).  
<sup>52</sup> K. Heckmann, *Naturwissenschaften*, **40**, 478 (1953); *Kolloidzshr.* **136**, 67 (1954);  
 U. Schindewolf, *Naturwissenschaften*, **40**, 435 (1953); *Z. Phys. Chem. (N.F.)* **1**, 129 (1954);  
*Z. Elektrochemie*, **58**, 697 (1954);  
 B. Jacobson, *Rev. Sci. Instrum.* **24**, 949 (1953).  
<sup>53</sup> M. Eigen and G. Schwartz, *Z. Phys. Chem. (N.F.)* **4**, 380 (1955); *J. Colloid Sci.* **12**, 181 (1957).  
<sup>54</sup> G. Schwarz, *Z. Phys. Chem. (N.F.)* **19**, 286 (1959);  
 G. Schwarz, *Z. Phys.* **145**, 563 (1956).  
<sup>55</sup> C. T. O'Konski and A. J. Haltner, *J. Am. Chem. Soc.* **79**, 5634 (1957).  
<sup>56</sup> M. Mandel, *Molec. Phys.* **4**, 489 (1961);  
 M. Mandel and A. Jenard, *Trans. Faraday Soc.* **59**, 2158 (1963);  
 F. van der Touw and M. Mandel, *Proc. Leiden Symp.* Vol. I, p 375 (1970).  
<sup>57</sup> B. Sachs, 'Dielectric dispersion of polyelectrolyte solutions', *Ph.D. Thesis*, Weizmann  
 Institute of Science (1965);  
 B. Sachs, A. Raziell, H. Eisenberg and A. Katchalsky, *Trans. Faraday Soc.* **65**, 77 (1969).  
<sup>58</sup> W. J. H. M. Moller, G. A. J. van Os and J. Th. G. Overbeek, *Trans. Faraday Soc.* **57**, 312 and  
 325 (1961).  
<sup>59</sup> R. E. Nelson and P. Ander, *J. Phys. Chem.* **75**, 1691 (1971).  
<sup>60</sup> D. Dolar, J. Špan and A. Pretnar, *J. Polymer Sci. (Part C)*, **16**, 3557 (1968).  
<sup>61</sup> For a general treatment of electrolyte transport, see  
 A. Katchalsky and P. Curran, *Non Equilibrium Thermodynamics in Biophysics*, Harvard  
 University Press: Cambridge, Mass. (1965).  
<sup>62</sup> R. M. Fuoss and H. Sadek, *Science*, **110**, 552 (1949).  
<sup>63</sup> Review article of H. G. Bungenberg de Jong, 'Crystallization-Coacervation-Flocculation',  
 in *Colloid Science*, Vol. II, p 232 (1949).  
<sup>64</sup> A. Katchalsky and P. Spitnik, *Colloque Internat. Cen. Nat. Rech. Sci., Strassbourg*, p 103 (1954).  
<sup>65</sup> H. Terayama, *J. Polymer Sci.* **19**, 181 (1952).  
<sup>66</sup> J. Th. G. Overbeek and M. J. Voorn, *J. Cell. Comp. Biol.* **49** (suppl.) 1 and 7 (1957);  
 M. J. Voorn, *Rec. Trav. Chim. Pays-Bas*, **75**, 316, 405, 427 and 1021 (1956).  
<sup>67</sup> A. Veis, 'Phase equilibria in systems of interacting polyelectrolytes', in *Biological Poly-*  
*electrolytes*, ed. A. Veis, Chap. IV, p 211. Marcel Dekker: New York (1970).

## POLYELECTROLYTES

- <sup>68</sup> H. J. Bixler and A. S. Michels, 'Polyelectrolyte complexes', in *Encyclopedia of Polymer Science and Technology*, Vol. X, p 765. Wiley: New York (1969).
- <sup>69</sup> V. Shashoua, *Nature, London*, **215**, 846 (1967).
- <sup>70</sup> A. Katchalsky and R. Spangler, *Quart. Rev. Biophysics*, **1**, 127 (1968).
- <sup>71</sup> M. Leng and G. Felsenfeld, *Proc. Nat. Acad. Sci. Wash.* **56**, 1325 (1966);  
J. F. Shapiro, M. Leng and G. Felsenfeld, *Biochemistry*, **8**, 3219 (1969).
- <sup>72</sup> R. A. Cox, A. S. Jones, G. E. Marsh and A. R. Peacocke, *Biochim. Biophys. Acta*, **21**, 576 (1961);  
R. A. Cox, *Quart. Rev. Chem. Soc. London*, **22**, 499 (1968).
- <sup>73</sup> E. Neumann and A. Katchalsky, *Ber. Bunsenges. Phys. Chem.* **74**, 868 (1970).
- <sup>74</sup> E. Neumann and A. Katchalsky, *Proc. 1st Europ. Biophysics Congr.*, Baden, Austria (1971).
- <sup>75</sup> e.g. *Molecular Approaches to Learning and Memory*, ed. W. L. Byrne. Academic Press: New York (1970).
- <sup>76</sup> A. Katchalsky and A. Oplatka, *Neurosciences Res. Progr. Bull.* **4** (suppl.), p 71 (1966);  
A. Katchalsky, A. Oplatka and A. Litan, in *Molecular Architecture in Cell Physiology*, eds. T. Hayashi and A. G. Szent-Györgyi, p 3. Prentice-Hall: New York (1966).
- <sup>77</sup> A. Katchalsky and E. Neumann, in *Physical Principles of Neuronal and Organismic Behaviour*, University of Chicago Press: in press (1971).