The Behaviour of Weak Electrolytes in Moving Boundary Systems

I. The Moving Boundary Equation

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After the introduction of the moving boundary method as a tool for the measurement of ionic mobilities by Lodge[1] and Whetham[2-7], a thorough theoretical treatment of the method was given by Kohlrausch[8] and Weber[9]. In these publications we find the differential equation governing the motion of ions in an electric field. Moreover, a complete solution of the differential equation was given for the case of three ion species, which has been of the greatest importance in all applications of the method. Another result of great consequence was Kohlrausch's finding that the function:

$$\sum \frac{c_i}{u_i} \tag{1}$$

named by Kohlrausch *die beharrliche Funktion*, and nowadays generally called the Kohlrausch function or the regulating function (MacInnes and Longsworth[10]), is independent of time if the ionic mobilities u_i can be regarded as constants ($c_i =$ the equivalent concentrations, taken with signs). Simplified theoretical treatments of the moving boundary method have later been presented by MacInnes and Longsworth (*l. c.*) and by Hartley and Moilliet[11]. MacInnes and his collaborators have collected a great number of transference numbers by the moving boundary method, which data are the most accurate hitherto available.

Picton and Linder[12], Whitney and Blake[13], and Hardy[14] were the first who applied the method to colloids. During thirty years, the method was gradually developed, but great difficulties were encountered in localizing the

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diffuse boundaries, and another obstacle was the fact that moving colloid boundaries always showed different rates of migration on the rising and descending sides. Tiselius [15] obtained accurate and reproducible mobility data for proteins by using low protein and fairly high salt concentrations. The assymmetry between the two limbs of the U-tube was depressed in this way, and the processes going on in the two limbs were nearly mirror image processes.

Henry and Brittain [16] attacked the problem in another way. They applied the Kohlrausch and Weber differential equation to the colloidal ions and came to the conclusion that the descending boundary, although more diffuse, gave the correct mobility, which was also in agreement with that obtained by the transference method.

When Tiselius [17] developed the moving boundary method into a quantitative analytical method for colloids seven years later, an extension of the theory to multicomponent systems became desirable. A first attempt in this direction was made by Svensson [18] in 1943. An equation giving the concentration change for an arbitrary ion across a moving boundary was given, and by summing this equation over all ions, an expression for the conductivity change at the boundary was derived. By applying the equation to the colloidal ions in the mixture, the conclusion was drawn that every ion changes its concentration at every boundary. The importance of this in the interpretation of electrophoresis patterns was discussed.

The moving boundary equation was given a more general form by Longsworth [19] and Dole [20] two years later. Dole's ingeneous mathematical treatment of the whole moving boundary system revealed not only the number of boundaries but also a complete solution of the complicated system of equations. The solution was based upon the assumption of constant relative mobilities, as was also Svensson's treatment. The experimental test given simultaneously by Longsworth showed that the theory yielded a good quantitative agreement with experiments under the conditions defined in the theory. Longsworth and Dole clearly understood and expressed the fact that the assumption of constant relative mobilities restricted the validity of the theory to strong electrolytes.

However, a bulk of experimental evidence has also been collected giving strong support to the theoretical predictions even for colloidal systems. Svensson [18] showed that the apparent composition of pig serum varied considerably with the salt concentration in the way that could be expected, and Perlmann and Kaufman [21] found the same independently for human serum. Continued experimental research in this field has been reported by Koenig and Hogness [22], Koenig, Perrings, and Hogness [23], and by Armstrong, Budka, and

Morrison [24]. These works give a qualitative confirmation of the theories, but a quantitative verification for proteins is still lacking.

Thus we have now a complete understanding of moving boundary systems of strong electrolytes, but we miss the corresponding knowledge of the behaviour of weak electrolytes. An extended treatment of moving boundary systems is therefore desirable already for the interpretation of simple systems with only a small number of weak electrolytes. Moreover, Tiselius' electrophoresis technique involves multi-component systems where weak electrolytes cannot be avoided: the buffer contains at least one weak electrolyte, and the colloids under investigation also belong to this class.

In the general case, with an arbitrary number of components of weak or strong electrolytes, the complete solution of the mathematical problem, which would allow us to predict the number and velocities of the boundaries and the composition of each inter-boundary layer from the known compositions of the two end solutions, or *vice versa*, is not available. If it were given, it would probably be too complicated to be of any practical significance. There is, however, another way of attacking the problem, namely to apply the moving boundary equation to each constituent and to each boundary in moving boundary systems that are actually observed experimentally.

THE CHARACTERISTICS OF WEAK ELECTROLYTES

By definition, a weak electrolyte is an electrolyte which is incompletely dissociated. One could also say that the ions of weak electrolytes react chemically with each other to form complex ions or uncharged molecules. Strong electrolytes are completely dissociated, and the interaction between positive and negative ions is restricted to that described by the Debye-Hückel theory. There is no reason to assume that there is a sharp boundary between the two groups, but there is strong experimental evidence that many salts, acids, and bases are pratically completely dissociated in dilute solutions in polar solvents.

A very important group within the weak electrolytes are the weak acids, bases, and ampholytes. In these substances, we have complexes where one of the water ions is a partner. According to Brønsted's views, however, the hydroxyl ion is dispensed with as a complex partner; Brønsted's protolytes are proton-donors or proton-acceptors.

Ions of weak electrolytes are thus characterized of being complex-formers. It should be noted in this connection that the partner in a complex formation need not be another ion, it may also be a non-electrolyte. The hydrated ions, the metal-ammonia complexes, the ammonium ion, and the iodide-iodine complex are examples of this type of particles.

The term »ion» means nowadays a free ion. To denote the sum of free ions and those bound in different complexes, MacInnes [25] uses the term »ion constituent», and Hartley and Moilliet [11] have proposed the term »radical». An ion constituent or a radical of a weak electrolyte is thus capable of existence in a number of different forms which we will subsequently call *subspecies*. The difference between an ion and a subspecies is then that a subspecies may be uncharged. In a solution, the relative concentrations of the different subspecies depend upon the concentrations of those ions or molecules with which they enter into complex formation. For protolytes, pH is the chief factor. The acetic acid radical exists partly as undissociated acid, HAc, and partly as the acetate ion, Ac⁻ (the hydration being neglected); it has two subspecies. The oxalic acid radical has three subspecies, one uncharged, one monovalent, and one bivalent. The citric acid radical has six subspecies because there are two isomeric forms of the monovalent, and two of the bivalent citric acid anions. With an increased number of ionizable groups in the molecule, the number of subspecies increases very rapidly. All subspecies are in equilibrium with each other, the equilibria being quantitatively described by a number of mass-action equations equal to one less than the number of subspecies.

From the point of view of moving boundary systems, it is not essential to distinguish between strong and weak electrolytes. The only matter of importance is to decide whether the mobilities can be regarded as constant or not. It is true that complex formation may cause a mobility to vary in a much more radical way than other influences (ionic interaction, viscosity, temperature, etc.), but nevertheless it is well possible to design moving boundary systems containing typical weak electrolytes where the assumption of constant mobilities can be used more successfully than in other systems with only strong electrolytes.

THE MOVING BOUNDARY EQUATION

This equation is the corner-stone in the theory for the migration of strong electrolytes, and it must be so even for weak electrolytes. It is now available in a number of different forms. As a matter of fact, it was given already by Weber [9] for a three-ion system in the case where no ion disappeared at the boundary. Svensson [18], being primarily interested in colloidal systems with small changes across the boundaries, gave the equation in terms of increments in concentration, mobility, and conductivity. The forms now most generally used are those given by Longsworth [19] and Dole [20]:

$$V^{\alpha\beta} \ (C_j^\alpha - C_j^\beta) = T_j^\alpha - T_j^\beta \tag{2}$$

$$\left(\frac{r_j}{\sigma^\alpha} - V^{\alpha\beta}\right) C_j^\alpha = \left(\frac{r_j}{\sigma^\beta} - V^{\alpha\beta}\right) C_j^\beta \tag{3}$$

$$(r_j - V^{\alpha\beta}\sigma^\alpha)\ T_j^\alpha = (r_j - V^{\alpha\beta}\ \sigma^\beta)\ T_j^\beta \tag{4}$$

Here V is the velocity of the moving boundary in cm³ per faraday, C is the concentration of the component in faradays per cm³, T are transference numbers, r are relative mobilities assumed to be constant, σ is the »relative conductivity» $= \Sigma\ C_j r_j$, the superscripts α and β refer to the two phases on either side of the boundary, and the subscript j refers to the component.

Svensson [25] gave the equation in the following forms in 1946:

$$\frac{c_{i1}\ (u_{i1} - U')}{\varkappa_1} = \frac{c_{i2}\ (u_{i2} - U'')}{\varkappa_2} \tag{5}$$

$$(c_{i1} - c_{i2})\ (u_{i1} - U') = \left(\frac{\varkappa_1}{\varkappa_2} - \frac{u_{i1}}{u_{i2}}\right) c_{i2}\ u_{i2} \tag{6}$$

$$(c_{i2} - c_{i1})\ (u_{i2} - U'') = \left(\frac{\varkappa_2}{\varkappa_1} - \frac{u_{i2}}{u_{i1}}\right) c_{i1}\ u_{i1} \tag{7}$$

In these equations, c is the concentration of the constituent in equivalents per unit volume, and u its mobility. U' and U'' are the velocities of the boundaries divided by the field strengths prevailing above and below it, respectively; they have the dimension of a mobility and may be called the mobilities of the boundaries. $\varkappa$ is the conductivity. The first subscript, i, denotes the number of the constituent, and the second, 1 or 2, refers to the two solutions above and below the boundary. Between the absolute velocity, V, in cm. per sec., and the quantities U' and U'' we have the relations:

$$V = \frac{U'I}{\varkappa_1} = \frac{U''I}{\varkappa_2} \tag{8}$$

where I is the current density.

We will restrict this discussion of the moving boundary equation to the forms (5) — (7) above and to (3), which is very nearly identical. The equations (2) and (4), containing the transference numbers, will be discussed in a forthcoming paper in connection with a discussion on the concept of transference number.

The validity of the moving boundary equation in the forms (2)—(4) has been demonstrated by Longsworth [19] for strong electrolytes. If we want

to extend its applicability to arbitrary systems, however, we have to give three definitions. First, it is necessary to define the constituents for which the equation can be used. Second, we must define the concentration scale or scales that can be used. Third, we must give an extended definition of the concept of mobility.

THE CONSTITUENTS TO WHICH THE MOVING BOUNDARY EQUATION MAY BE APPLIED

In systems of strong electrolytes, the equation may be used for every ion species. All but one of the ionic concentrations can be chosen at will, the concentration of the last ion being given by the electroneutrality condition. In systems of weak and non-electrolytes, the situation is not so simple. First, to describe the behaviour of the whole system, it is necessary to consider not only ions, but also non-electrolytes that may be electrically transferred due to complex formation with ions. We must also consider the fact that a radical of a weak electrolyte exists in several forms, subspecies, and decide whether we shall use the equation for the radicals or for the subspecies.

What concerns non-electrolytes, the most natural view-point is to regard them as parts of a mixed solvent and not to include them into the general treatment of the moving boundary system or to treat the non-electrolytes together as one component. However, if the non-electrolytes enter into complex formation with ions selectively, a relative motion between the different non-electrolytes will take place, and we cannot any longer describe the phenomena from the point of view of a mixed solvent. In the general case, therefore, we must regard each non-electrolyte as an independent constituent and apply the moving boundary equation to it in the same way as to other constituents. For some non-electrolytes, this is immediately evident: ammonia is formally a non-electrolyte, but it has a strong electrical transference due to complex formation with protons and other cations. For the same reason, nitrogen monoxide can be expected to be electrically transported in the presence of ferrous ions, and iodine in the presence of iodides. It is quite evident that certain mobility values may be ascribed to these non-electrolytes and, consequently, that they will have concentration increments at moving boundaries. The affinity between the ions and the solvent in the solvated ions is far less specific, but the effect upon the electrical migration of the solvent is quite analogous. Thus even the solvent will possess a certain mobility. However, this mobility is equal to zero by definition as long as we choose the solvent as the frame of reference.

The electrical behaviour of ion species existing as two or several subspecies has been considered by a number of authors. When introducing the mobilities of the ions, Kohlrausch stated: »Gemeint ist hier natürlich die empirisch bestimmte mittlere Beweglichkeit aller ortsanwesenden Ionen eines Elementes, nicht diejenige der dissociierten Ionen, die man im allgemeinen nicht kennt.» At that time, all electrolytes were believed to be incompletely dissociated. Miller [27] pointed out that the transference numbers determined by means of the Hittorf method are identical with those from moving boundary experiments whether the ions of the solution be complex or simple. Tiselius [15] wrote that a substance that in a certain medium contains the fractions a_1, a_2, $a_3 \ldots$ of the forms 1, 2, 3, $\ldots$ (all in equilibrium) with the mobilities u_1, u_2, u_3, $\ldots$ will migrate as a uniform substance with a mobility u given by:

$$u = a_1 u_1 + a_2 u_2 + a_3 u_3 + \ldots \qquad (9)$$

provided that the time of existence of each subspecies is small in comparison with the time of migration (momentaneous equilibrium reactions).

Already from this historical survey, it is evident that the application of the moving boundary equation to subspecies is not justified. The equation is not valid for ions that can be formed or disappear in chemical reactions when the ion traverses the boundary. Let us assume that we are considering a moving boundary across which there is a considerable pH change. When acetate ions traverse this boundary from the alkaline side, a certain fraction of them will disappear due to the chemical reaction:

$$H^+ + Ac^- = HAc$$
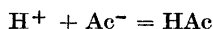
Thus the moving boundary equation cannot be applied to the acetate ion if acetic acid is present on one or on both sides of the boundary. For the same reason, it cannot be applied to the undissociated acetic acid in the presence of acetate ions.

The incorrectness of using the moving boundary equation for subspecies is also evident from the following discussion. Let us assume that we know the composition of the solution above a certain moving boundary, and that we know the conductivity in both solutions. With the aid of the moving boundary equation, we can then compute the composition of the solution below the boundary if the assumption of constant relative mobilities is valid. Let us assume that we apply the equation to HAc, to Ac^-, to $H_2PO_4^-$, and to HPO_4^{--}. From the values obtained for the concentrations of HAc and Ac^- below the boundary, we would be able to compute the pH of this solution. We could also

compute pH from the ratio of the concentrations obtained for the two phosphate ions. These two pH values cannot be proven to be identical, thus there must be something wrong. We can obtain a third value of pH by computing the concentration of free hydrogen ions below the boundary with the aid of the moving boundary equation, and a fourth value by doing the same for the hydroxyl ions. All these pH values are wrong and not identical, and the reason is that the moving boundary equation is not valid for subspecies.*

There are two ways to escape from this difficulty. One is to modify the moving boundary equation and to take into account the chemical reactions with the aid of the mass-action law. The other way is to give up the treatment of subspecies altogether and to apply the equation to the radicals, which include the specific chemical groupings wherever they are found, in free or complex form (cf. Hartley and Moilliet [11]). There is no doubt that the latter procedure is the more rational one. The treatment of individual subspecies is on the whole not possible if there are more than two or three, and even in these simple cases, the computation of their concentrations involves non-thermodynamical assumptions.

A protein is a protolyte with a large number of ionizable groups and an innumerable number of subspecies. We would never consider the possibility of using the moving boundary equation for all these subspecies because it is impossible; we know nothing about them. But since we know to a high degree of accuracy both the concentrations and mobilities of HAc, Ac^-, $H_2PO_4^-$, and HPO_4^{--} in buffer mixtures, we are inclined to apply the equation to each of them. This is not logical, and unless we take into account chemical reactions between them and modify the moving boundary equation accordingly, it is also incorrect.

As already pointed out, no sharp distinction can be made between strong and weak electrolytes, and the situation is the same between weak and non-electrolytes. Even the alkali metal cations, being characterized by a very small tendency to complex formation, are well known to be strongly solvated in polar solvents, although the degree of solvation is very difficult to measure. Moreover, alkali salts of high-molecular poly-acids have been found to be poorly dissociated, probably due to non-specific electrostatic forces. This suggests that we must try to find a mode of treatment that is not restricted to any group of substances but applicable to strong as well as to weak and non-electrolytes. In other words, as far as the question is how to define a constituent, we have to neglect the difference between strong and weak electrolytes.

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* The failure of the moving boundary equation for ions of weak acids in the presence of the acid itself has recently been discovered by Alberty and Nichol [33] in independent research.

The only modification that is necessary to introduce in the definition of an electrical constituent for strong electrolytes to make it adaptable for arbitrary substances is thus to replace the word »ion» by »radical», and to include non-electrolytes; thus: *the moving boundary equation may be applied to every radical, charged or uncharged, that cannot be dissociated into simpler radicals or transformed into any other radical under the conditions to which the investigation is restricted.*

All these constituents are not mutually independent as to their concentrations. Since some of them are positive or negative radicals, we have here, as in the case of strong electrolytes, the restriction that the sum of all equivalent concentrations, taken with signs, must add up to zero. Furthermore, since we have now taken the solvent or its radicals as electrical constituents, we have also another restriction, namely that the sum of all partial volumes (that fraction of the total volume that is occupied by the constituent) must be equal to unity.

The requirement that the radical must be non-dissociable is obvious since its migration cannot be described if it can dissociate into two parts with different mobilities. On the other hand, the property of being stable or dissociable depends upon the conditions to which the investigation is restricted. For instance, in a system containing ammonium phosphate, the radicals of this salt can be defined as NH_4^+ and HPO_4^{--} if the investigation is restricted to pH:es below 7, for in this pH range these subspecies can be regarded as stable. However, if the investigation is extended to the entire pH range, we have to define the radicals as NH_3 and PO_4^{---}.

The situation is similar for water. In many cases we can regard the water as non-dissociable and thus define H_2O as a constituent. However, in a solution of tri-sodium phosphate, it is obviously erroneous to say that we have the constituents Na^+, PO_4^{---}, and H_2O, since we also have a quite appreciable concentration of hydroxyl ions; in this case the water cannot be treated as nondissociable. Instead of the water, we have two smaller radicals: the hydrogen radical and the hydroxyl radical.

It should be noted that, according to the above definition, the hydrogen radical is a constituent as soon as a protolyte is present, not only in acid, but also in neutral and alkaline solutions, and that all ions containing the hydrogen radical must be considered as subspecies, not only of the protolyte, but also of hydrogen. The concentration of the hydrogen radical stands in no relation to pH. pH is only related to one subspecies of the hydrogen radical, namely to the free hydrogen ions.

In water solutions containing no other weak electrolytes than protolytes, two constituents have an exceptional position: the hydrogen radical as being

the only positive complex-partner, and the water as being the solvent (or the hydroxyl radical in the case that the water must be treated as dissociable). It is therefore convenient, in these systems, to calculate these two concentrations, if they are of interest, from the two restrictive conditions and to apply the moving boundary equation to all the other constituents. Thus it is never necessary to use the moving boundary equation for the hydrogen and hydroxyl radicals. In the absence of protolytes and in mixed solvents, the exceptional positions of the hydrogen radical and the water disappears.

THE CONCENTRATION

In all earlier contributions to the theory of the moving boundary method, the concentrations of the constituents have been expressed in equivalents per unit volume, in later works even with the signs of the charges. This is extremely convenient for many reasons. The valences of the components need not be introduced, the same equations apply to both cations and anions, and the electroneutrality condition takes the simple form: $\Sigma c_i = 0$.

Since the equations (3), (5), (6), and (7) contain the concentrations as factors on both sides, the concentration scale seems to be arbitrary. However, it is evident for dimensional reasons that the concentration must be given *on a volume basis*. The equation cannot be derived and does not hold if the concentrations are expressed in mols per 1000 grams of solvent, in grams per 1000 grams of solvent, or in percent. *The concentration unit must be mols, equivalents, or grams per unit volume.*

The equations (2) and (4), containing the transference numbers, require that the concentrations be expressed in equivalents per unit volume. Thus these equations cannot be used in cases where the concepts of equivalent concentration and transference number are not well defined. It is inapplicable to non-electrolytes, to amino acids, to proteins and to most other colloids. The question how to define equivalent concentrations and transference numbers for these substances will be dealt with in a separate communication.

THE MOBILITY

If we look in a physico-chemical table-work (International Critical Tables; Landolt-Börnstein, Tabellen) for mobility data of the acetate and phosphate radicals, we find nothing but the data for the subspecies Ac^-, $H_2PO_4^-$ and HPO_4^{--}. For other weak electrolytes, the situation is similar, or there is no information at all. It is obvious that these data refer to a certain pH range (for Ac^-: above pH 8) or to a single pH value (for $H_2PO_4^-$: pH 4.5; for HPO_4^{--}:

pH 9), where these subspecies exist alone. Mobility data under conditions where both subspecies exist together are not available. Now, we have come to the conclusion that, under such circumstances, we cannot treat the subspecies as independent constituents, thus mobility data are required even in pH ranges where two or more subspecies exist simultaneously. An extended definition of a mobility is apparently necessary, a definition according to which it is possible to measure the required data experimentally. This definition must be given in terms of moving boundary or transference experiments. We are allowed to base the definition on both types of experiments since it is now well established that they give the same result if the plane of reference is the same (Miller [27]).

In terms of a moving boundary experiment, the definition is as follows. *The mobility in a certain medium of a constituent as defined above, comprising a radical in different subspecies, is the velocity relative to some plane of reference or other of a moving boundary at which the constituent with all its subspecies disappears, divided by the field strength next to the boundary in the solution containing the constituent in question* (cf. Longsworth [19], Dole [20], and Svensson [25 p. 13]).

Sometimes it is very difficult or impossible to arrange a moving boundary experiment satisfying the three necessary conditions: 1) sharp boundary; 2) gravitationally stable boundary; 3) the conductivity next to the boundary known. In such cases a transference experiment must be made instead, and the mobility derived from the following definition: *The mobility in a certain medium of a constituent in the sense given above is the mass transported relative to some plane of reference or other per unit time, unit cross section of the tube, unit field strength, and unit concentration of the constituent* (cf. Hartley and Moilliet [11]):

$$u = \frac{M}{c\,q\,t\,E} = \frac{M\,\varkappa}{c\,Q} \tag{10}$$

In the above equations, M is the transported mass of the radical, c its concentration, q the cross-sectional area of the tube, t the time, E the field strength, Q the amount of electricity passed, and $\varkappa$ the conductivity. The second expression is more useful than the first.

The classical frame of reference is the solvent, but this is not generally useful. Thus, in the case of mixed solvents, the components of which are in relative motion, it is evident that not the solvent as a whole, but well one component of it, can serve as the frame of reference. There are also other cases where it is not suitable to have the solvent as the plane of reference. Therefore, in the definitions given above, we have left the plane of reference

open to free choice, but evidently it is necessary to state to which plane of reference a given mobility refers.

Early workers in the field of moving boundaries proposed to use an electrically inert substance as the frame of reference instead of water. Transference numbers thus obtained were called »true transference numbers», and from the difference between them and the Hittorf transference numbers relative degrees of hydration between cations and anions could be estimated (Lewis [27], Buchböck [28], Washburn [29, 30]). A recent investigation by Longsworth [31] shows, however, that we have no means to demonstrate that there is any substance that is more electrically inert than water. The migration of water relative to the added non-electrolytes turned out to be different for different reference substances. Thus the concept of a true transference number involves an assumption, the validity of which is difficult to demonstrate.

It should be pointed out that the planes of reference used for the mobilities above and below a moving boundary must not be in relative motion towards each other if the moving boundary equation shall retain its absolute validity. This condition is never fulfilled by the solvent, nor by any other constituent in the mixture. It is satisfied by the apparatus and by any moving or stationary boundary in the system. The frame of reference was discussed at length by Dole [20], who gave a modified moving boundary equation with exact validity for the case that the solvent is chosen as the frame of reference in every interboundary layer.

Special difficulties are encountered in applying the definitions to the hydrogen and hydroxyl radicals in water solutions because these are the solvent radicals. As already pointed out, however, the moving boundary equation need not be applied to these radicals.

If we are concerned with chemical equilibria which are not instantaneously established, the above definitions will in general be inapplicable. It is evident that the observed mobility will be a function of time before the equilibrium is reached. After reaching equilibrium, the transference method may give consistent and reproducible mobilities, whereas the moving boundary method requires that the equilibrium is established momentaneously.

If the highest accuracy is not necessary, and if the number of subspecies is small, it is sometimes possible to omit the moving boundary or transference experiment and to calculate the desired mobility with the aid of equation (9). The use of this equation, however, necessarily involves some assumption that cannot be tested. For instance, in an acetate buffer with known amounts of sodium acetate and acetic acid, we can neglect the dissociation of the latter, so we know very accurately the concentrations of the two subspecies. Their mobilities, however, cannot be measured in the mixture where they coexist.

The mobility of the acetate ion must be measured in the absence of acetic acid (above pH 8), and *vice versa*, and to compute the mobility of the acetate radical in the mixture, one must assume that the mobility of one subspecies is independent of the presence of the other. This assumption is, however, justified to a high degree of accuracy in many cases.

DISCUSSION

Again considering the special case that the only weak electrolytes present are protolytes, the abundance of the different subspecies will essentially depend upon pH, and the mobilities of the constituents will be pH-functions. This explains why it is possible to avoid the treatment of a number of mass-action equations and the reformulation of the moving boundary equation by adding new terms to make allowance for chemical reactions. The law of mass-action is inherent in the function $u = f(\text{pH})$ in an operational manner.

In solutions of one single weak electrolyte, the necessity of treating the radicals instead of the subspecies is easily understood and has been so for a very long time. In mixtures of a weak acid and its salt, however, this point of view is not similarly self-evident. The dissociation of the acid being very much depressed in such a mixture, it lies close at hand to regard it as a non-electrolyte and to treat the salt as a strong electrolyte. This treatment is completely satisfactory in many cases, whereas in other cases it is quite untenable, which has been shown experimentally both in this laboratory and by Alberty and Nichol [33].

Moving boundary experiments with buffer mixtures designed according to the definition of a mobility given above have been carried out in this laboratory in great number during the last year. Thus the mobility of the acetate radical can be measured at arbitrary pH values by placing the acetate buffer in the bottom of the Tiselius U-tube and by using a propionate buffer as supernatant. Moving boundaries develop at which the acetate radical disappears from the bottom and the propionate radical from the top. These boundaries may be given any desired mobility between that of the free acetate ion and zero by varying the proportion between sodium acetate and free acetic acid (pH). This new type of moving boundary experiments showed the additional peculiarity that the moving boundary, although gravitationally stable and very sharp, had a negative refractive index increment.

Two special cases are of interest in showing the connection between viewpoints restricted to strong electrolytes and such of general validity. First, if the number of subspecies reduces to one, we have the case of strong electrolytes. This is very nearly the case for salts such as sodium acetate, ammonium

chloride, disodium phosphate, where the hydrolysis is very slight. Second, if pH is essentially constant in the system *after the boundaries have developed*, the relative mobilities of the radicals will also be constant. Chemical reactions will occur only to a small extent when subspecies traverse the boundaries or *vice versa*. Under such circumstances, the application of the moving boundary equation to subspecies will be approximately justified. This has been shown experimentally by Longsworth[34] who computed the small pH change across the moving boundary by applying the moving boundary equation to the acetate ions. In spite of the presence of undissociated acetic acid, the pH change thus computed showed a good agreement with experimental observations. The same result has been obtained in this laboratory.

The case mentioned above, with a constant pH throughout the moving boundary system, is of considerable importance in the field of electrophoresis. Electrophoretic analyses are nowadays carried out in well-buffered solutions, and thus we can conclude that the results may be interpreted with the aid of the theory for strong electrolytes to a very high accuracy. This was realized and pointed out by Longsworth[19].

SUMMARY

The fact that hitherto available theories for moving boundary systems are strictly valid only for strong electrolytes, as pointed out by Dole, made it evident that a generalization of the theory is required. This article is the first step in such a generalization in that it gives the moving boundary equation in a form of quite unrestricted validity. Formally, the equation is the same as earlier. The constituents to which it can be applied have been defined as non-dissociable radicals. It has been pointed out that not only equivalent concentrations, but also molar and other concentrations on a volume basis may be used, which is of importance for substances such as amino acids, proteins, other colloids, and non-electrolytes. A generalized definition of the mobility of a radical has been given in terms of moving boundary or transference experiments. A new type of moving boundary experiments according to this definition were briefly mentioned.

REFERENCES

1. Lodge, O. *Brit. Assoc. Advancement Sci. Rept.* (1886) 389.
2. Whetham, W. C. D. *Proc. Roy. Soc. London* **52** (1892—93) 283.
3. Whetham, W. C. D. *Phil. Trans. A* **184** (1893) 337.
4. Whetham, W. C. D. *Z. physik. Chem.* **11** (1893) 220.
5. Wetham, W. C. D. *Phil. Mag.* **38** (1894) 392.
6. Whetham, W. C. D. *Proc. Roy. Soc. London* **58** (1895) 182.
7. Whetham, W. C. D. *Phil. Trans.* **186** (1895) 507.
8. Kohlrausch, F. *Ann. Physik* **62** (1897) 209.
9. Weber, H. *Sitzungsber. Akad. Wiss. Berlin* (1897) 936.
10. MacInnes, D. A., and Longsworth, L. G. *Chem. Rev.* **11** (1932) 171.
11. Hartley, G. S., and Moilliet, J. L. *Proc. Roy. Soc. London A* **140** (1933) 141.
12. Picton, H., and Linder, S. E. *J. Chem. Soc. London* **71** (1897) 568.
13. Whitney, W. R., and Blake, J. C. *J. Am. Chem. Soc.* **26** (1904) 1339.
14. Hardy, W. B. *J. Physiol.* **33** (1905).
15. Tiselius, A. *Nova Acta Regiae Soc. Sci. Upsaliensis IV,* **7** (1930) no. 4.
16. Henry, D. C., and Brittain, J. *Trans. Faraday Soc.* **29** (1933) 798.
17. Tiselius, A. *Trans. Faraday Soc.* **33** (1937) 524.
18. Svensson, H. *Arkiv Kemi, Mineral. Geol. A* **17** (1943) no. 14.
19. Longsworth, L. G. *J. Am. Chem. Soc.* **67** (1945) 11u9.
20. Dole, V. P. *J. Am. Chem. Soc.* **67** (1945) 1119.
21. Perlmann, G. E., and Kaufman, D. *J. Am. Chem. Soc.* **67** (1945) 638.
22. Koenig, V. L., and Hogness, K. R. *Arch. Biochem.* **9** (1946) 119.
23. Koenig, V. L., Perrings, J. D., and Hogness, K. R. *Arch. Biochem.* **11** (1946) 345.
24. Armstrong, S. H., Budka, M. J. E., and Morrison, K. C. *J. Am. Chem. Soc.* **69** (1947) 416.
25. Svensson, H. *Arkiv Kemi, Mineral. Geol. A* **22** (1946) no. 10.
26. MacInnes, D. A. *The principles of electrochemistry* New York (1939).
27. Miller, W. L. *Z. physik. Chem.* **69** (1909) 436.
28. Lewis, G. N. *J. Am. Chem. Soc.* **32** (1910) 862.
29. Buchböck, G. *Z. Physik. Chem.* **55** (1906) 563.
30. Washburn, E. W. *J. Am. Chem. Soc.* **31** (1909) 322.
31. Washburn, E. W. *J. Am. Chem. Soc.* **37** (1915) 694.
32. Longsworth, L. G. *J. Am. Chem. Soc.* **69** (1947) 1288.
33. Alberty, R., and Nichol, J. C. *J. Am. Chem. Soc.* **70** (1948) 2297.
34. Longsworth, L. G. *J. Phys. Colloid Chem.* **51** (1947) 171.