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MEMBRANE POTENTIALS ACROSS THE BOVINE LENS CAPSULE *IN VITRO*

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SUMMARY

Membrane potentials of the lens capsule were studied *in vitro* for electrolytes of various types: KCl, NaCl, LiCl, $MgCl_2$, $CaCl_2$, $LaCl_3$, Na_2SO_4, K_2SO_4, $MgSO_4$ and HCl.

The lens capsule had positively fixed charges. The parameter $l_-^*/(l_+^* + l_-^*)$ for an electrolyte in the membrane phase was equal to the parameter $l_-/(l_+ + l_-)$ for the electrolyte in bulk solution. Theoretical results at c_2/c_1 = constant coincided with experimental data in $c_2 > \Theta$, but experimental values of the membrane potential deviated systematically below theoretical curves as c_2 decreased in $c_2 < \Theta$, where l_+^* and l_-^* are the ionic mobilities in the membrane phase of positive and negative ions, respectively, l_+ and l_- are the ionic mobilities in bulk solutions of positive and negative ions, respectively, c_2 is the higher concentration of bulk solutions (moles/l), and Θ is the effective charge density of the membrane (moles/l).

The interaction parameter between dissociated ions of the membrane and ions of the bulk solution is discussed.

INTRODUCTION

Our previous paper was concerned with the derivation of an equation for the concentration dependence of permeability coefficients for an electrolyte component, and with its applicability to experimental data obtained on the bovine lens capsule *in vitro*[1].

In the present paper, we describe theoretical and experimental studies on the membrane potential arising between two solutions of an electrolyte that are separated by a lens capsule in *vitro*. Many experimental studies on the membrane potential have been restricted to uni-univalent electrolyte systems for simplicity, although multivalent electrolyte systems are important in biological systems. Here we will examine uni-univalent and multivalent electrolyte systems.

EQUATIONS FOR THE MEMBRANE POTENTIAL

The system is isothermal and consists of a charged membrane that separates two electrolyte solutions of the same kind but of different concentrations c_2 and c_1 (moles/l) ($c_2 > c_1$) at the same hydrostatic pressure.

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From the thermodynamics of irreversible processes, we derived the expression for the diffusion potential of uni-univalent and multivalent electrolyte systems in the previous paper[1] and we obtained the revised Nernst–Planck equation for the membrane system, *i.e.*, flux = (effective mobility) × (mobile ion concentration) × (force). Here, the mobile ion concentration is assumed to be equal to the analytical ion concentration. Then we obtain for the membrane potential the same expression that has been obtained by TEORELL[2] and MEYER AND SIEVERS[3]. But the meaning of charge density of the membrane Θ, which appears in the electric neutrality condition, is different from the original meaning of TEORELL[2] and MEYER AND SIEVERS[3]. The Θ in the present paper is the product of the real charge density of the membrane X and the interaction parameter Φ that is characteristic of a given pair of membrane and electrolyte ($\Theta = \Phi X$). Φ was discovered by TOYOSHIMA *et al.*[4,19,20], KOBATAKE *et al.*[5], YUASA *et al.*[6] and TOYOSHIMA AND NOZAHI[7]. Although our treatment, based on the assumptions that the mobile ion concentration is equal to the analytical ion concentration and $\Theta = \Phi X$, is different from that of refs. 4–7, 19, 20, the results are essentially equivalent. For example, Eqn. 20 in ref. 7 is also derived from Eqns. 2 and 3 in this paper. Here. Φ is assumed to be independent of bulk concentration. (YUASA *et al.*[6] and TOYOSHIMA AND NOZAKI[7] recently argued the dependence of Φ on concentration.)

In the following, we summarize equations that are necessary for the calculation of the membrane potential.

For the diffusion potential in the membrane $(\Delta\Psi)_{\text{in membrane}}$ in the steady state

$$(\Delta\psi)_{\text{in membrane}} = -(RT/F)\frac{(l_+{}^* - l_-{}^*)}{(l_+{}^*Z_+ - l_-{}^*Z_-)} \ln \frac{\bar{c}_+{}^{\text{II}}l_+{}^*Z_+{}^2 + \bar{c}_-{}^{\text{II}}l_-{}^*Z_-{}^2}{\bar{c}_+{}^{\text{I}}l_+{}^*Z_+{}^2 + \bar{c}_-{}^{\text{I}}l_-{}^*Z_-{}^2} \qquad (1)$$

R is the gas constant, T is the absolute temperature, F is the Faraday constant, and Z_+ and Z_- are the valencies of positive and negative ions, respectively. $\bar{c}_+{}^{\text{I}}$ and $\bar{c}_-{}^{\text{I}}$ are the analytical concentrations of positive and negative ions, respectively, in the membrane at the phase boundary I. $\bar{c}_+{}^{\text{II}}$ and $\bar{c}_-{}^{\text{II}}$ are the analytical concentrations of positive and negative ions, respectively, in the membrane at the phase boundary II. The phase boundaries I and II are the boundaries between the membrane and Solution I (its concentration is c_1) and Solution II, respectively. $l_+{}^*$ and $l_-{}^*$ are the effective ionic mobilities of positive and negative ions, respectively, in the membrane, and these are assumed to be independent of concentration. We will check the extent of the difference between the experimental value of $l_-{}^*/(l_+{}^* + l_-{}^*)$ and the value of $l_-/(l_+ + l_-)$ in the literature[8] without using the assumption of $l_-{}^*/(l_+{}^* + l_-{}^*) = l_-/(l_+ + l_-)$, where l_+ and l_- are the ionic mobilities of positive and negative ions, respectively, in the bulk solution. In the derivation of Eqn. 1, we used the requirement of electric neutrality in any element of the membrane,

$$Z_+\bar{c}_+ + Z_-\bar{c}_- + \Theta = 0 \qquad (2)$$

Using the Donnan equilibrium,

$$(c_+{}^0/\bar{c}_+)^{Z-} = (c_-{}^0/\bar{c}_-)^{Z+} \qquad (3)$$

we obtain for the sum of two phase boundary potentials

$$(\Delta\psi)_{\text{phase boundary}} = -(RT/FZ_+)\,(\ln c_+{}^{0\text{II}}/c_+{}^{0\text{I}} - \ln \bar{c}_+{}^{\text{II}}/\bar{c}_+{}^{\text{I}}) \qquad (4)$$

$c_+^{0\mathrm{I}}$ and $c_+^{0\mathrm{II}}$ are the concentrations of positive ions in solutions I and II, respectively. $\bar{c}_+^{\mathrm{I}}$, $\bar{c}_+^{\mathrm{II}}$, $\bar{c}_-^{\mathrm{I}}$ and $\bar{c}_-^{\mathrm{II}}$ in Eqns. 1 and 4 can be calculated from Eqns. 2 and 3. The total membrane potential is the sum of the phase boundary potential and the diffusion potential,

$$\Delta\psi = (\Delta\psi)_{\text{phase boundary}} + (\Delta\psi)_{\text{in membrane}} \tag{5}$$

For electrolytes of $Z_+ \neq |Z_-|$ (for the most multivalent electrolytes), we have to calculate $\bar{c}_+$ and $\bar{c}_-$ by solving cubic, biquadratic and higher order equations derived from Eqns. 2 and 3 (ref. 1). But for $Z_+ = |Z_-|$ (for uni-univalent electrolytes and bi-bivalent electrolytes), we can get the simplier expression from Eqns. 1–5 because $\bar{c}_+$ is given in the solution of the quadratic equation from Eqns. 2 and 3. For $Z_+ = |Z_-| = Z$,

$$(FZ/RT)\Delta\psi = -\ln\gamma + \ln\left[\frac{-\Theta^* + \sqrt{\Theta^{*2} + 4c_2^2}}{-\Theta^* + \sqrt{-\Theta^{*2} + 4(c_2/\gamma)^2}}\right]$$
$$- (1 - 2A)\ln\left[\frac{(2A-1)\Theta^* + \sqrt{\Theta^{*2} + 4c_2^2}}{(2A-1)\Theta^* + \sqrt{\Theta^{*2} + 4(c_2/\gamma)^2}}\right] \tag{6}$$

where

$$c_2/c_1 = \gamma,\ \Theta^* = \Theta/Z \text{ and } A = l_-^*/(l_+^* + l_-^*)$$

Limiting form in the region of $c_2 \gg \Theta$ for electrolytes of $Z_+ = |Z_-| = Z$

From thermodynamics we obtain another equation for the membrane potential, using the transference number t_+;

$$\Delta\psi = -(RT/FZ)\,(2t_+ - 1)\ln\gamma \tag{7}$$

It is known experimentally that at a fixed $c_2/c_1 = \gamma$ the inverse of an apparent transference number t_+ for the co-ion species in a positively charged membrane is proportional to the inverse of concentration c_2 (or c_1) in the region of $c_2 \gg \Theta$ (ref. 9). Expanding Eqn. 6 with Eqn. 7 for $1/t_+$ in power of $1/c_2$ gives

$$1/t_+ = \frac{1}{1-A} + \frac{(\gamma-1)A}{(1-A)\ln\gamma}\left(\frac{\Theta^*}{c_2}\right) + \mathrm{O}\left[\left(\frac{\Theta^*}{c_2}\right)^2\right] \tag{8}$$

For the negatively charged membrane ($\Theta < 0$), we get

$$1/t_- = \frac{1}{A} - \frac{(\gamma-1)(1-A)}{A\ln\gamma}\left(\frac{\Theta^*}{c_2}\right) + \mathrm{O}\left[\left(\frac{\Theta^*}{c_2}\right)^2\right] \tag{9}$$

in the derivation of which we have used

$$\Delta\psi = -(RT/FZ)\,(1 - 2t_-)\ln\gamma \tag{10}$$

In Eqns. 8 and 9, $\mathrm{O}[(\Theta/c_2)^2]$ means the second order term and is assumed to be negligibly small compared with the first and the second terms on the right hand of these equations in the region of $c_2 \gg \Theta^*$. From Eqn. 8, the intercept for a plot of $1/t_+$ against $1/c_2$ at a fixed γ is equal to $1/(1-A)$. Once A is known, Θ can be evaluated from the initial slope for $1/t_+$ *vs.* $1/c_2$.

MATERIALS AND METHODS

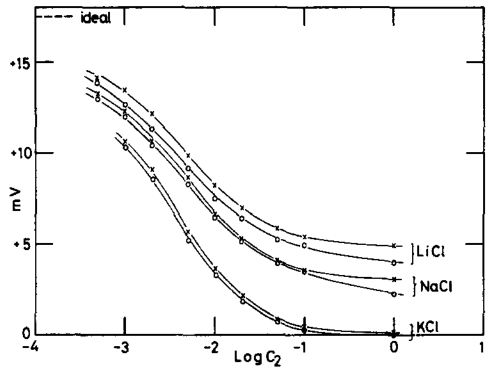
The electromotive force was measured by a vibrating reed electrometer (Toshiba Co., Tokyo, Japan, UFE-26301 type). A positive membrane potential indicates that the electric potential in Solution II was positively higher than the electrical potential in Solution I. The caps of the salt bridges were carefully ground so that test solutions might not be contaminated by leakage of KCl. Measured electromotive forces were not corrected for liquid junction potentials between the salt bridge and the electrolyte solution. Ag–AgCl electrodes of the electrolytic type, prepared according to the method reported by BROWN[10], had been found to be appropriate by preliminary experiments. For Ag–AgCl electrodes, the measured potential is the sum of the membrane potential and the electrode potential. In calculating the electrode potential, we need the single ion activity coefficient. For uni-uni electrolytes, single ion activity coefficients are replaced by the thermodynamic mean ion activity coefficients following the GUGGENHEIM[11] assumption. For bi-uni electrolytes and tri-uni electrolytes, the activity coefficient for the critical ion, Cl^-, is calculated by assuming that the activity coefficient of a given ion is a function only of the ionic strength of the solution. Then the activity coefficient γ_{Cl^-} in a solution of a bi-uni electrolyte or tri-uni electrolyte is the same as γ_{Cl^-} in a KCl solution having the same ionic strength, assuming that $\gamma_{Cl^-} = \gamma_{K^+}$ in a solution of KCl alone[12].

The lens capsule was removed by the method described in the previous paper[1]. The methods of cleaning and conditioning of the lens capsule and of setting it in the measuring cell were the same as before[1]. The stability of the well-cleaned lens capsules was also discussed previously[1]. The salts used were analytical grade reagents and were subjected to no purification except drying. The water was distilled and deionized with a mixed bed ion exchange column. CO_2 dissolved in water was not removed. The pH of aqueous solutions in equilibrium with laboratory air was about 5.8.

RESULTS AND DISCUSSION

Fig. 1 shows data of the membrane potential as a function of log c_2 for lens capsule 10 with LiCl, NaCl and KCl at $c_2/c_1 = 2$. (The number placed after the lens capsule indicates the individual membrane.) In Fig. 1, marks ○ show experimental data measured by calomel electrodes and marks × show experimental data measured by Ag–AgCl electrodes. The difference between two values of the membrane potential measured by two kinds of electrode at the same concentrations of electrolyte is small for systems of KCl and NaCl and is relatively large for the system of LiCl. We consider that experimental results obtained with Ag–AgCl electrodes are more accurate than those obtained with calomel electrodes because the membrane potential measured by Ag–AgCl electrodes include no ambiguous liquid junction potential.

In order to find the parameter of $Z_+ = |Z_-|$ electrolyte systems, we plotted $1/t_+$ against $1/c_2$ at $\gamma = 2$ in the region of relatively higher concentrations, *i.e.*, $c_2 \geqslant 0.005$ M in Fig. 2. (Except for $MgSO_4$, membrane potentials were measured by Ag–AgCl electrodes for Fig. 2.) The values of $l_-^*/(l_+^* + l_-^*)$ and Θ were determined from this graph using Eqn. 8. Table I shows the values of Θ and compares the values of $l_-^*/(l_+^* + l_-^*)$ with the values of $l_-/(l_+ + l_-)$. From Table I it is evident that the effective charge density of the membrane, Θ, differed from one membrane to another



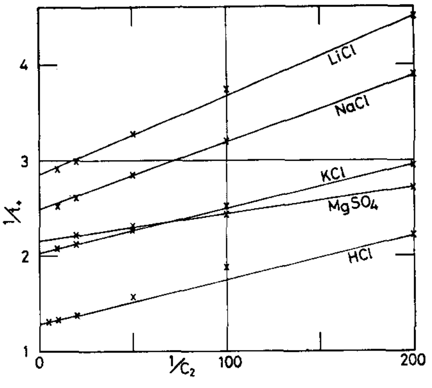


Fig. 1. Experimental data of membrane potentials as a function of log c_2 for systems of lens capsule 10 (LiCl, NaCl and KCl) at $c_2/c_1 = 2$. ○ show data measured by calomel electrodes and × show data measured by Ag–AgCl electrodes.

Fig. 2. Plots of $1/t_+$ *vs.* $1/c_2$ for systems of lens capsule 10 (LiCl, NaCl and KCl) (Ag–AgCl electrodes)) and for systems of lens capsule 16 ($MgSO_4$ (calomel electrodes)) and lens capsule 29 (HCl (Ag–AgCl electrodes)).

TABLE I

VALUES OF PARAMETERS FOR SYSTEMS OF LENS CAPSULES AND ELECTROLYTES

Hg means calomel electrodes and Ag means Ag–AgCl electrodes.

Chemical	*Membrane No.*	*Types of electrode*	$\Theta \times 10^3$ *M*	$l_-^*/(l_+^* + l_-^*)$	$l_-/(l_+ + l_-)$*
NaCl	10	Hg	3.0_5	0.60	0.60
NaCl	10	Ag	3.2_3	0.60	0.60
NaCl	15	Hg	3.7_6	0.59	0.60
NaCl	16	Hg	4.7_7	0.59	0.60
NaCl	17	Hg	2.6_5	0.59	0.60
NaCl	18	Hg	3.5_2	0.59	0.60
KCl	10	Hg	2.9_1	0.50	0.51
KCl	10	Ag	3.1_6	0.51	0.51
KCl	16	Hg	3.5_4	0.50	0.51
KCl	18	Hg	3.5_4	0.50	0.51
KCl	19	Hg	4.1_6	0.50	0.51
LiCl	10	Hg	2.7_1	0.64	0.68
LiCl	10	Ag	3.1_6	0.65	0.68
$MgSO_4$	16	Hg	3.2_3	0.54	0.60
HCl	29	Ag	1.1_4	0.22	0.18
$CaCl_2$	12	Hg	6.0_4	0.72	0.72
$CaCl_2$	12	Ag	6.0_4	0.74	0.72
$MgCl_2$	12	Hg	6.1_7	0.73	0.74
$MgCl_2$	12	Ag	6.0_4	0.76	0.74
$LaCl_3$	12	Hg	7.9_5	0.82	0.70
$LaCl_3$	12	Ag	6.7_7	0.84	0.70
Na_2SO_4	16	Hg	6.6_1	0.37	0.44
K_2SO_4	16	Hg	2.7_6	0.33	0.35

* Taken from ref. 8.

although signs of Θ were always positive; the values of $l_-^*/(l_+^* + l_-^*)$ were the same for the same electrolyte despite the broad distribution of Θ. For the same electrolyte, values of $l_-^*/(l_+^* + l_-^*)$ in membranes are nearly in accord with values of $l_-/(l_+ + l_-)$

in bulk solutions. Therefore the assumption $l_+^*/l_-^* = l_+/l_-$ which had been used in our previous paper[1] is shown to be correct when the same kind of electrolyte exists in both Solutions I and II. A rough estimate of the order of Φ is $\Phi_{\mathrm{NaCl}} > \Phi_{\mathrm{KCl}} = \Phi_{\mathrm{LiCl}}$ for the lens capsule 10 if the real charge density X in various electrolytes is unchanged for the same membrane. $\Phi_{\mathrm{NaCl}} > \Phi_{\mathrm{KCl}}$ was also found from our diffusion experiment (see Table I in ref. 1 for Membrane 7) and in the collodion membrane (see Table I on p. 2823 in ref. 19). Unfortunately, we cannot compare the value of Θ in Table I of this paper with the value of Θ in Table I of the previous paper[1] because the membranes are not the same. However, the magnitude of Θ obtained here from experiments of the membrane potential is nearly the same as that obtained from the diffusion experiments[1]. When experiments of the diffusion and the membrane potential are made on the same membrane, we get roughly equal Θ from both experiments. By way of example, for NaCl at $\gamma = 2$, Θ from the diffusion experiment was $2.3 \cdot 10^{-3}$ and Θ from the membrane potential was $2.5 \cdot 10^{-3}$.

Solid lines in Fig. 3 show theoretical membrane potentials for KCl, NaCl and LiCl as a function of log (c_2/Θ) calculated from Eqn. 6 alone at $\gamma = 2$, in which $l_-^*/(l_+^* + l_-^*)$ has the values given in Table I, *i.e.*, 0.60 for NaCl, 0.51 for KCl and 0.65 for LiCl. Experimental values of membrane potentials for lens capsule 10 with NaCl, KCl and LiCl measured by Ag–AgCl electrodes indicated × are also plotted in Fig. 3, using parameter $\Theta = 3.23 \cdot 10^{-3}$ for NaCl, $\Theta = 3.16 \cdot 10^{-3}$ for KCl and $\Theta = 3.16 \cdot 10^{-3}$ for LiCl. These Θ values were obtained from the $1/t_+$ *vs.* $1/c_2$

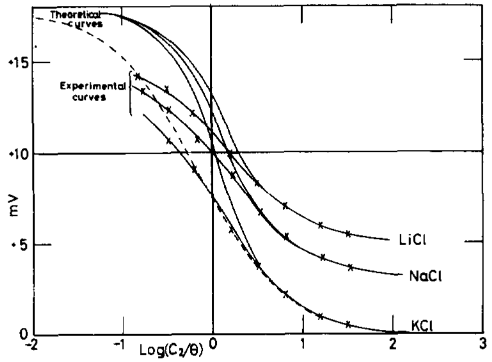


Fig. 3. Solid lines show theoretical curves of membrane potentials for systems of NaCl, KCl and LiCl at $c_2/c_1 = 2$. The parameters $l_-^*/(l_+^* + l_-^*)$ used for calculations of membrane potentials were 0.60 for NaCl, 0.51 for KCl and 0.65 for LiCl. The dotted line shows the theoretical curve of the membrane potential of KCl at $c_2/c_1 = 2$, taken from Fig. 2 in the paper by KOBATAKE *et al.*[15]. Lines ×—× show experimental curves for the same systems in Fig. 1 measured by Ag–AgCl electrodes, and the experimental data (×) are plotted using $\Theta = 3.16 \cdot 10^{-3}$ for KCl, $\Theta = 3.23 \cdot 10^{-3}$ for NaCl and $\Theta = 3.16 \cdot 10^{-3}$ for LiCl.

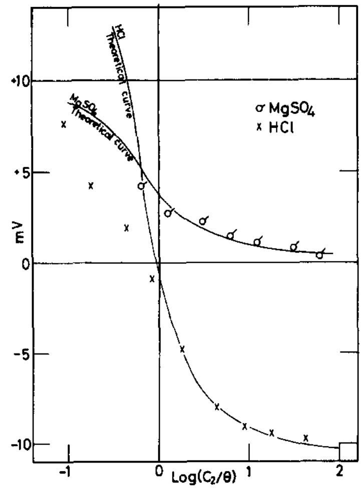


Fig. 4. Solid lines represent theoretical curves of membrane potentials for systems of HCl with $l_-^*/(l_+^* + l_-^*) = 0.22$ and $MgSO_4$ with $l_-^*/(l_+^* + l_-^*) = 0.54$. The experimental data of HCl (×) measured by Ag–AgCl electrodes and $MgSO_4$ (○) measured by calomel electrodes are plotted using $\Theta = 1.14 \cdot 10^{-3}$ for HCl and $\Theta = 3.23 \cdot 10^{-3}$ for $MgSO_4$.

relationship. It is clear that theoretical curves fit well with experimental data in the range of $c_2 > \Theta$ but deviate from experimental curves when $c_2 < \Theta$.

Let us consider the origin of this deviation. The disagreement found here in the biological membrane has been found in ion exchange membranes (for example, HILLS *et al.*[13]). First, in this paper the activity coefficient of ions in bulk solutions is assumed to be unity although it is known to be dependent on bulk concentrations. In fact in the region of $c_2 \gg \Theta$ this factor becomes important, and has been discussed for artificial ion exchange membranes[14]. But in the region of low concentration, the activity coefficient of ions in bulk solutions tends to unity as the concentration decreases; hence the deviation of the membrane potential observed here cannot be explained by the small change of activity coefficient of ions in bulk solutions because the deviation becomes greater as the concentration decreases. Second, this deviation might be due to the over-simplification of the phase boundary potential by using the ideal Donnan potential as given by Eqn. 4, in which $\bar{c}_+$ is the concentration averaged over a cross-section of a pore in the membrane. Actually, the distribution of the ions in a pore is not homogeneous throughout the cross-section, but is determined by the Poisson–Boltzmann equation taking into consideration the fact that the electric charges of the membrane are located on the periphery of the cross-section. One of the authors had already worked out this problem for uni-univalent electrolytes, and concluded that the ratio $(c_+^0 c_-^0/\bar{c}_+\bar{c}_-)$ decreases with decrease of the concentration c_2, although the value of the ratio in the range of high c_2 value is equal to 1, which is the value for the ideal Donnan equilibrium[15]. The numerical result for KCl according to this theory[15] is drawn in Fig. 4 with a dotted line, showing that the agreement between the theoretical and experimental results is seen in a wider range of concentrations. It is, however, difficult to extend this method of the theoretical treatment[15] to electrolytes of multivalent ions because of the complicated expression containing the first and the second kinds of elliptic function. On the other hand, the theoretical equations given in the present paper are much simpler and still give fairly good agreement with experimental results at least in the range of $c_2 > \Theta$. The simple theoretical equations have the advantage that they can be applied to electrolytes of multivalent ions as is to be discussed here, and even to the bi-ionic potential which will be discussed in a separate paper in preparation.

In Fig. 4, we show theoretical curves of membrane potentials for lens capsule 16 with $MgSO_4$, and for lens capsule 29 with HCl, as a function of log (c_2/Θ) from Eqn. 6 alone using the parameter $l_-^*/(l_+^* + l_-^*) = 0.54$ for $MgSO_4$ and $= 0.20$ for HCl given in Table I. Experimental values of membrane potentials are also plotted in Fig. 4 using Θ obtained from the $1/t_+$ *vs.* $1/c_2$ relationship. ($\Theta = 3.23 \cdot 10^{-3}$ for $MgSO_4$, $\Theta = 1.14 \cdot 10^{-3}$ for HCl.) The ranges of measured bulk concentration c_2 are 0.002–0.2 M for $MgSO_4$ (ó) and 0.001–0.5 M for HCl (×). Experimental results fit the theoretical curves in the full range of measured concentration for $MgSO_4$ and in the range of $c_2 > 0.01$ M for HCl, but deviate from the theoretical curve for HCl in the range of $c_2 < 0.01$ M. If we measure the membrane potential of $MgSO_4$ in lower concentrations than 0.002 M, we should find a similar deviation. The deviation of experimental result from the theoretical result occurs in the range of $c_2/\Theta < 1$ as in systems of NaCl, KCl and LiCl. In the HCl system, the pH of the bulk solution changes according to bulk concentration and hence the potential is a function of pH. The state of dissociation of fixed charges of the lens capsule might change, and therefore the real charge density X might change from concentration to concentration. But, in the range of $c_2 > 0.01$ M, experimental results coincide with the theoretical result, and we cannot find anomalous behavior owing to the change of dissociated state of fixed ions. In the range of $c_2 < 0.01$ M, we could not estimate the extent of the deviation owing to the change of the dissociated state of ions.

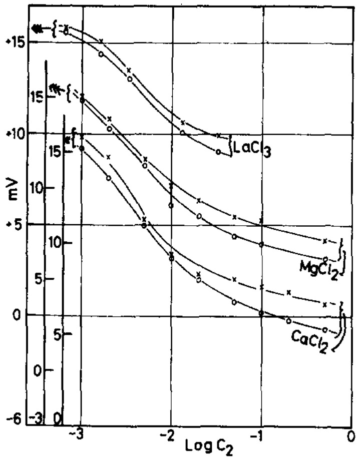


Fig. 5. Experimental data of membrane potentials for systems of lens capsule 12 ($CaCl_2$, $MgCl_2$ and $LaCl_3$) at $c_2/c_1 = 2$. ×, Ag–AgCl electrodes; ○, calomel electrodes.

Next we consider the case of $Z_+ \neq |Z_-|$. Fig. 5 shows membrane potentials for lens capsule 12 with $CaCl_2$, $MgCl_2$ and $LaCl_3$ at $\gamma = 2$ (○, calomel electrodes; ×, Ag–AgCl electrodes). The difference between membrane potentials by calomel electrodes and Ag–AgCl electrodes in these systems is larger than that in systems of NaCl and KCl. For $Z_+ \neq |Z_-|$, we cannot obtain the limiting form of Eqn. 5 which depends on concentration in the range of $c_2 \gg \Theta$. Therefore we cannot obtain the parameter $l_-^*/(l_+^* + l_-^*)$ and Θ from plots of $1/c_2$ *vs.* $1/t_+$. Theoretical curves derived from Eqns. 1–5 without the use of Eqn. 6, and experimental data of membrane potentials for these type of electrolyte ($Z_- = -1$) at $\gamma = 2$, tend to 17.8 mV in the range of $c_2 \gg \Theta$ as c_2 decreases and tend to some limiting values in the range of $c_2 \ll \Theta$ as c_2 increases. These latter limiting values depend only on the values of $l_-^*/(l_+^* + l_-^*)$. We can make the experimental results coincide with the theoretical curve by using the appropriate value of $l_-^*/(l_+^* + l_-^*)$. Here we determined the parameter $l_-^*/(l_+^* + l_-^*)$ by trial and error. First we calculated the membrane potential as a function of log (c_2/Θ) using an arbitrary value of $l_-^*/(l_+^* + l_-^*)$. (In the highest concentration, the phase boundary potential is nearly zero. Therefore, for the initial value of $l_-^*/(l_+^* + l_-^*)$, we easily approach the real value of $l^{-*}/(l^{+*} + l^{-*})$ by using the equation $\Delta\Psi = -(RT/F)\ [(l_+^*/l_-^* - 1)/(l_+^* Z^+/l_-^* - Z_-)\ \ln \gamma$, obtained from Eqn. 1 for the diffusion potential at the highest concentration.) The theoretical curve was then super-imposed on Fig. 5. If the coincidence of the theoretical curve and the experimental data in high bulk concentrations was not good, we repeated this procedure using another arbitrary value of $l_-^*/(l_+^* + l_-^*)$ till the theoretical curve came to fit the experimental data in high bulk concentrations. The experimental curve is a function of log c_2 and the theoretical curve is a function of log (c_2/Θ). So the value of Θ is obtained by the proper superimposition of these two curves. Fig. 6 shows theoretical curves obtained by the above method for these systems measured by Ag–AgCl electrodes. [$l_-^*/(l_+^* + l_-^*) = 0.74$ for $CaCl_2$, 0.76 for $MgCl_2$ and 0.84 for $LaCl_3$.] Experimental data obtained with Ag–AgCl electrodes are plotted in the same figure using the values $\Theta = 6.04 \cdot 10^{-3}$ for $CaCl_2$ (×), $\Theta = 6.04 \cdot 10^{-3}$ for $MgCl_2$ (○) and $\Theta = 6.67 \cdot 10^{-3}$ for $LaCl_3$ (▽). For $c_2 > \Theta$, experimental values of the membrane potential fit the theoretical curves. For $c_2 < \Theta$, experimental values of the membrane potential fall systematically below theoretical curves as c_2 decreases. A rough estimate of the order of Φ for lens capsule 12 is $\Phi_{LaCl_3} > \Phi_{CaCl_2} > \Phi_{MgCl_2}$ if X is unchanged from electrolyte to electrolyte.

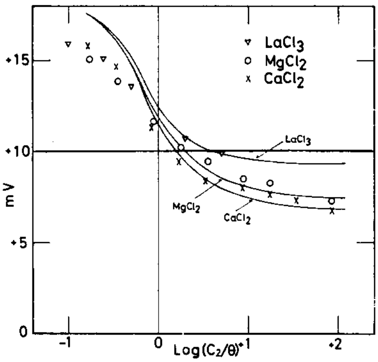


Fig. 6. Solid lines represent theoretical curves of membrane potentials for the same systems in Fig. 5 measured by Ag–AgCl electrodes with $l_-^*/(l_+^* + l_-^*) = 0.74$ for $CaCl_2$, 0.76 for $MgCl_2$ and 0.84 for $LaCl_3$. The experimental data measured by Ag–AgCl electrodes in Fig. 5 are also plotted using $\Theta = 6.04 \cdot 10^{-3}$ for $CaCl_2$ (×), $\Theta = 6.04 \cdot 10^{-3}$ for $MgCl_2$ (○) and $\Theta = 6.77 \cdot 10^{-3}$ for $LaCl_3$ (▽).

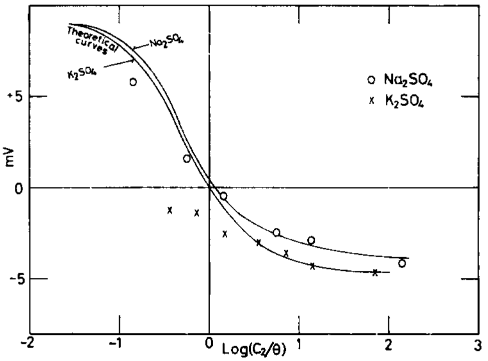


Fig. 7. Solid lines represent theoretical curves of membrane potentials for systems of lens capsule 16 (Na_2SO_4 with $l_-^*/(l_+^* + l_-^*) = 0.37$ and K_2SO_4 with $l_-^*/(l_+^* + l_-^*) = 0.33$) at $c_2/c_1 = 2$. Experimental data of membrane potentials for systems of lens capsule 16 (Na_2SO_4 and K_2SO_4) measured by calomel electrodes are plotted using $\Theta = 6.61 \cdot 10^{-3}$ for Na_2SO_4 (○) and $\Theta = 2.76 \cdot 10^{-3}$ for K_2SO_4 (×).

Fig. 7 shows theoretical curves of membrane potentials calculated from Eqns. 1–5 as a function of log (c_2/Θ) for lens capsule 16 with K_2SO_4 and Na_2SO_4. [$l_-^*/(l_+^* + l_-^*) = 0.33$ for K_2SO_4 and 0.37 for Na_2SO_4.] Membrane potentials for this type of electrolyte ($Z_- = -2$) tend to +8.9 mV [$\Delta\Psi = (RT/2F) \ln \gamma$] in low concentration. (If $\Theta < 0$, membrane potentials tend to −17.8 mV.) Experimental data for K_2SO_4 and Na_2SO_4 measured by calomel electrodes (indicated by × and ○, respectively) fit theoretical curves in the range of $c_2 > \Theta$. ($\Theta = 2.76 \cdot 10^{-3}$ for K_2SO_4 and $6.61 \cdot 10^{-3}$ for Na_2SO_4.) The ranges of measured bulk concentration c_2 are 0.0005–0.1 M for K_2SO_4 and 0.0005–0.5 M for Na_2SO_4. A rough estimate of the order of Φ for lens capsule 16 is $\Phi_{Na_2SO_4} > \Phi_{K_2SO_4}$.

Calculated curves of membrane potentials follow experimental data in the range of $c_2 > \Theta$ and deviate systematically above the latter as c_2 becomes smaller in the range of $c_2 < \Theta$ for every salt examined. On the other hand, our previous theory on membrane permeability[1], which has the same assumptions as made in this paper, yielded a good fit to the experimental data on the lens capsule. Because values of the membrane permeability coefficients, P_m, fall rapidly as c_2 becomes smaller, the theoretical results apparently approach the experimental results although they do not exactly coincide. On the other hand, values of the membrane potential become larger as c_2 becomes smaller and thus disagreement can be seen clearly in the case of the membrane potential.

It is said that the electrochemical properties (including the permeability properties) of this transparent well-cleaned membrane, which is known to be composed of a collagenous protein rich in hydroxyproline[16] and a carbohydrate (10 % of the capsular mass-mainly glucose)[17], are stable in electrolyte solutions for one or two weeks and are similar to synthetic ion exchange membranes that have a relatively small quantity of positively fixed charges. [For example, Θ_{NaCl} is —0.5 in ACI-C membrane (Asahi Chemical Industries of Japan)[18].]

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