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On the Activities of Sodium and Potassium Ion in the Solution Containing Bovine Albumine

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Activity of sodium or potassium ion is an important factor in the theory dealing with cellular trans-membrane potential. It was assumed, in the previous report (1), that this activity in the 0.15M (molar) solution containing bovine albumine was the same as in the simple solution. The assumption, however, induced negligible error in the case where the liquid junction potential between 3M KCl solution and 0.15M NaCl or KCl solution was studied. Of course, there would be many cases where a probable decrease of these activities in the solution containing protein can not be neglected.

In the present experiment, the activity in the solution containing albumine was directly measured and its decrease was seen in sodium ion but not in potassium.

METHOD

The cation-selective glass electrode (Horiba Inst. Inc.) was employed for measuring the activity of Na^+ (sodium ion). The potassium-selective electrode has not been worked out in Japan. Refering to the literature, this electrode seemed to be poor in its selectivity and stability. Therefore, the liquid junction potential between the 2M potassium citrate solution and the KCl solution was measured, and the activity of K^+ (potassium ion) was estimated. The former solution filled the conventional glass-capillary electrode, the tip of which was not less than 2μ in diameter avoiding the generation of tip-potential (2). When the 3M KCl solution, instead of K-citrate, filled the capillary, the potential was not stable. The cause of this unstability was not clear.

The concentration of the KCl solution, containing or discontaining the bovine serum albumine (Armour & Co.), ranged from 0.001 to 1.0 molar. The solution is denoted below as the test solution. The potential was measured, at the room temperature (20~24°C), with the potentiometer having the balanced circuit of single tube (Toshiba UX-54B). The relationship between the input voltage (0~150mV) and the deflection of galvanometer was linear. The detail of potentiometrie was previously reported (3).

RESULTS

The ordinate of Fig. 1 is the measured potential and the abscissa, the activity of Na^+ in logarithmic scale. The positive or negative sign on the former means the electrical polarity of the test solution with reference to the glass-electrode.

The relationship between the potential and the logarithm of activity calculated according to Robinson and Stokes (4) was linear when the test solution was the simple NaCl, and the straight line was drawn by the method of least mean square. The potential was measured with each NaCl solution, in which the bovine albumine was dissolved by 7 %, and the results were shown with open circles in the figure.

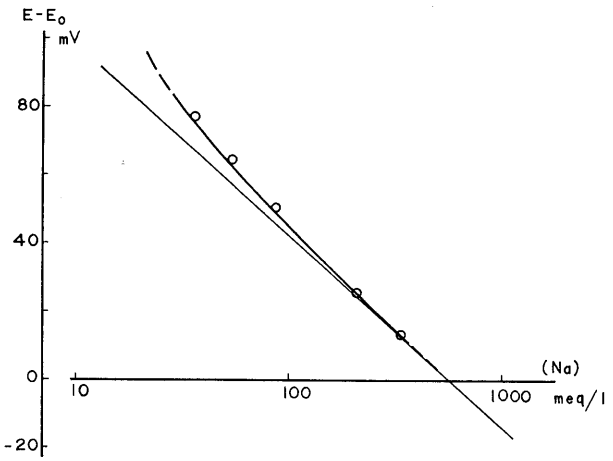

Fig. 1. Electrical potential of the solution with reference to Na-selective glass electrode is noted on the ordinate and the activity of Na, on the abscissa. Related to the straight line or the curve, see in the text.

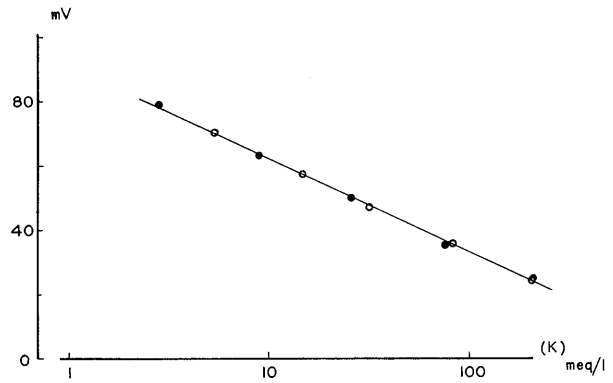

Fig. 2. Relationship between the liquid junction potential (ordinate) of K-citrate solution in the glass capillary and the activity of K (abscissa) in the KCl solution, containing (open circles) or discontaining (filled ones) albumine.

The employed albumine, however, contained not negligible amount of sodium, and the concentration was not the same as that in the simple solution. It was corrected refering the potential measured with lmM NaCl solution containing the albumine. Sometimes, the concentration determined by flame-photometrie was the reference. The curve in the figure was theoretically drawn with the assumption that an activity of Na^+ decreased by 10meq/1 in each solution containing albumine.

In figure 2, a relationship between the measured potential (ordinate) and the activity of K^+ (abscissa) is illustrated. The filled circles represent the results obtained with the simple KCl solutions and the open ones, with that containing the albumine by 7 %. As the albumine-preparation also contained potassium, the activity was similarly corrected. The straight line in the figure was drawn with the eye and had no mathematical meaning. Experimental data obtained with the solution containing the albumine by 5 % were nearly the same as that by 7 %, and were not illustrated in the figure.

DISCUSSION

1. Activity of sodium ion

The potential (E) of cation-selective glass electrode with reference to the solution, which contains two monovalent cations (A and B), is determined by the following equation(Eisenman et al., 5 and 6).

$$E = E_0 + \frac{RT}{F} ln \left\{ (A)^{\frac{1}{n}} + k^{\frac{1}{n}} (B)^{\frac{1}{n}} \right\}^n$$

In this equation, (A) and (B) mean the activities. E, k and n are the constants determined by the physico-chemical character of the glass. F, R and T have the usual meanings.

In the present experiment, A is sodium ion and B, potassium ion. When the simple NaCl solution is used, the equation reduces to;

$$E = E_0 + \frac{RT}{F} ln\ (Na^+)$$

Therefore, the slope of straight line in the figure 1 is, as illustrated, 57 mV (18°C) for the ten-fold change of activity. The decrease of Na^+-activity in the solution containing bovine albumine can be seen in the figure. It is, however, not proportional to the existing activity but substractive. In other words, about 10 meq/1 of Na^+ seems to be inactive in each solution containing albumine by 7 %.

It was observed, as stated above, that the concentration of Na^+ was increased when the albumine (preparation) was dissolved in the simple NaCl solution. Therefore, the decrease of its activity would not be caused by the binding of Na^+ to the molecule of that protein. The Coulomb's force between the Na^+ and the protein molecule having the negative charge would not be the cause, as the activity of

K^+ did not decrease in the similar condition (see below). The plausible cause cannot be found out in the present knowledge of authors.

2. Activity of potassium ion

The liquid junction potential can be measured with the conventional micro-electrode, the tip of which is not less than 2 micra (7). In the present study, the scheme of the electro-motive force is,

$$\underset{}{\text{Hg}\cdot\text{HgCl}} \;\Big|_{P_1}\; \underset{}{3\text{M}\cdot\text{KCl}} \;\Big|_{P_2}\; \underset{}{\text{test solution}} \;\Big|_{P_3}\; \underset{}{2\text{M}\cdot\text{K-Citrate}} \;\Big|_{P_4}\; \underset{}{3\text{M}\cdot\text{KCl}} \;\Big|_{P_5}\; \underset{}{\text{HgCl}\cdot\text{Hg}}$$

Each vertical line, in this scheme, means the boundary where the electro-motive force is generated and marked by a letter, P_1 or P_2 etc. If the two calomel-electrodes have the same physico-chemical characters, P_1 and P_5 cancel out mutually. The potential marked by P_2 is small and can be neglected (8). Therefore, the total potential is approximately the algebral sum of P_3 and P_4. In the experiment when the test solution is the simple KCl solution, P_3 and P_4 can be calculated according to the Henderson's equation (2). It is, however, a hard work to calculate the ionic activities and mobilities of 2M potassium citrate solution in the glass-capillary. Practically, on the other hand, the activity of K^+ in the KCl solution containing the albumine can be estimated by comparing the two potentials, the one of which is obtained by using the simple KCl solution as the test solution and the other, by using the solution of the same KCl concentration containing albumine; because the ionic activities and mobilities in the glass-capillary are the same in the two cases, and P_4 in the above scheme is constant. The practice is illustrated in the fig. 2 and shows that the difference of the two potentials is negligible.

It is likely, therefore, that the activity of K^+ is the same in the KCl solution containing albumine by 7 % as in the simple solution. The probable decrease of the activity in the former solution may be masked by the change of mobility of the K^+ and/or of Cl^- (chloride ion). It can, however, be estimated by the theoretical calculation, which will be presented elsewhere, that the change of mobility has a minor effect on the potential.

SUMMARY

1. An activity of sodium ion was measured with the cation-selective glass electrode. In the NaCl solution containing bovine serum albumine by 7 %, about 10 meq/l of the ion seemed to be inactive. The ionic concentration of the solution was ranged between 0.01 and 0.3 molar.

2. A liquid junction potential of the 2 molar potassium citrate in the glass capillary with reference to the simple KCl solution was compared to that with the solution of the same ionic concentration containing albumine by 5 or 7 %. The estimated activities of the potassium ion were nearly the same in both solutions, containing

or discontaining albumine.

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