Asian Journal of Chemistry Vol. 18, No. 1 (2006), 86–96

Study of Membrane Surface Potential with Pericardial Membrane

K.P. SINGH*, SANJU, SANJESH† and RAMESH K. PRAJAPATI‡
Department of Chemistry, Janta Vedic College, Baraut-250 611, India
E-mail: drkpsingh68@yahoo.co.in

Membrane surface potential was calculated using Poisson-Boltzman equation. It was observed that this equation is applicable to pericardial membrane and so this membrane falls in the category of low charged surface density. At higher concentration, the values of membrane surface potential decreased due to the greater deformation caused by increased value of transport of ions. Negative values of membrane surface potential showed that pericardial membrane is a positively charged structure. Moreover, this study also reveals that as soon as the negative value of –log C increases, the value of membrane surface potential also increases which is a symbol of decrease of ion transport across pericardial membrane when the concentration falls down. Increase in the value of membrane surface potential with decrease in concentration advocates the high sensitivity and self-surviving behaviour of heart function as this tissue is extensively used in constructing bioprosthetic heart valves.

Key Words: Membrane surface potential, Pericardial membrane, Trans-membrane.

INTRODUCTION

Structural changes associated with the membrane surface give rise to an electrostatic membrane surface potential, ψ_0. This potential participates in the processes of membrane interaction, recognition or solute binding. In addition to this, an electrostatic trans-membrane potential, $\Delta\psi_m$, exists across many membranes and across all such semipermeable membranes like barriers which divide two aqueous compartments filled with different, differently concentrated, or asymmetric electrolytes[1–3]. Such trans-membrane potential is found in almost all living cells. Intracellular sodium concentration (9 mmol L^{-1}), for example, is typically much lower than the extracellular space (120 mmol L^{-1}) whilst the concentration of K^+ is higher (in the muscle cell interior for example 140 mmol L^{-1}) than outside the cell (2.5 mmol L^{-1}) owing to the activity of the Na^+K^+-ATPase pumps. Chloride concentration is similar to sodium concentration.

---

†Department of Botany, Janta Vedic College, Baraut-250 611, India.
‡Department of Chemistry, D.J. College, Baraut-250 611, India.

The assumption that the ion distribution close to the membrane surface is governed solely by the coulombic surface-ion interactions in combination with the previously mentioned approximation results in Gouy-Chapman electrostatic theory[4, 5]. An essential implication of this model is that the ion concentration near the membrane surface differs from the bulk over a zone of finite thickness, the so-called ionic diffuse double layer. The width of this layer is approximately proportional to a characteristic, concentration dependent (Debye) screening length, λ, which under physiologial conditions (ion strength $I = 0.154$, temperature $T = 37°C$) is of the order of 1 nm. Moreover, the net (excess) ion change within the double layer is deduced to be identical in size, but opposite in sign, to the total charge on the membrane, because of the overall neutrality of the system.

Theories of membrane surface potential

Gouy-Chapman model: The Gouy-Chapman (G.C.) approximation[4, 5] is obtained directly from eqn (i):

$$d^2\psi_{00000}(x)/dx^2 \equiv -dE(x)/dx = -\rho_{el}(x)/\varepsilon\varepsilon_0$$

$$\equiv -(\rho_{el,\,m}(x) + \rho_{el,\,i}(x))/\varepsilon\varepsilon_0 \qquad (i)$$

under the proviso that all structural membrane charges are confined to an infinitely narrow plane and that the ion distribution is governed solely by coulombic forces.

Poisson-Boltzmann approximation: Traditionally, but not inevitably, the most popular electrostatic surface model, the Gouy-Chapman diffuse double layer theory, is derived from Poisson eqn. (1) by neglecting the spatial distribution of the membrane associated charge. This is tantamount to writing: $\rho_{el,\,m}(x) \equiv \sigma_{el}\delta$ (interface) for the surface and $\rho_{el}(x) \equiv \rho_{el,\,i}(x)$ everywhere else. For a planar surface one then has

$$d^2\psi_0(x)/dx^2 = -\rho_{el,\,i}(x)/\varepsilon\varepsilon_0 \equiv -(\rho_{el,\,i}/\varepsilon\varepsilon_0)/\psi(x) \qquad (1)$$

With the boundary condition: $d\psi(\text{interface})/dx = -\sigma_{el}/\varepsilon\varepsilon_m$. Parameters ε, ε_m and ε_0 are the dielectric constants of the aqueous and membrane subphase and the permittivity of free space, respectively.

Another key assumption of Gouy-Chapman approximation[4, 5] is that the charge distribution near the membrane is governed solely by the electrostatic, coulombic interactions according to the Boltzmann law:

$$\rho_{el}(x) = N_A e \Sigma_i Z_i c_i \exp\,[Z_i e \psi(x)/kT]$$

$$\equiv F \Sigma_i Z_i C_i \exp\,[Z_i F \psi(x)/RT] \qquad (2)$$

where e is the elementary electronic charge, Z_i is the ion valency, k is the Boltzmann constant, N_A is the Avogadro number, $F = {}_eN_A$ and $R = kN_A$ are Faraday and gas constant, respectively; T is the absolute temperature.

In electrochemistry and in all electrostatic theories it is customary to introduce the Debye-Huckel screening length by the general definition[6]

$$\lambda = (\varepsilon\varepsilon_0 kT/10^3\, N_A e^2 \Sigma Z_i^2 c_i)^{1/2} \qquad (3)$$

where the summation goes over all ionic species and the factor 10^3 appears because the bulk ion concentration is normally given in moles per litre. For a symmetrical electrolyte the Debye length λ can be conveniently written as

$$\lambda_{Z+Z\equiv-Z} = (\varepsilon\varepsilon_0 kT/20N_A c)^{1/2}/10Ze$$

which after insertion of numerical values for $T = 298$ K (25°C) yields

$$\lambda = 0.304Z^{-1}c^{-1/2} \text{ nm}$$

Combination of eqns. (1) and (2) affords Poisson-Boltzmann equation for a planar charged surface. Specifically for the case of symmetrical ionic solution, this is for an A^{z+}, B^{z-} electrolyte; one obtains

$$d^2\psi(x)/dx^2 = (2 \cdot 10^3\ N_A Zec/\varepsilon\varepsilon_0) \sinh [Ze\ \psi(x)/kT] \tag{4}$$

Similar equation can be written for the membrane electric field.

In terms of Debye length, the Poisson-Boltzmann equation for a symmetrical electrolyte reads

$$\lambda^2 d^2\psi(x)/dx^2 = kT/Ze) \sinh [Ze\ \psi(x)/kT] \tag{5}$$

In principle, either the electrostatic surface potential, ψ_0, or the surface density of the structural membrane charge, $\sigma_{el} = e/A_c$, can be kept constant, say, by some metabolic process. In the majority of the biologically relevant cases the latter possibility is the more probable one. Therefore, the usual boundary conditions with which the solution of eqn. (4) should be sought is σ_{el} = constant.

For little charged surfaces of the membranes, the relation between the electrostatic membrane potential and the surface charge density from the Poisson-Boltzmann equation is found (by expanding eqn. (ii) in terms of surface potential) to be:

$$\psi_0 = \{(2kT/Ze) \sinh^{-1} (Ze\sigma_{el}\lambda/2\varepsilon\varepsilon_0 kT)(\sigma_{el}\lambda/\varepsilon\varepsilon_0,\ \psi_0 < 2kT/Ze = 25 \text{ mV})\} \tag{ii}$$

$$\psi_0 = \sigma_{el}\lambda/\varepsilon\varepsilon_0 \ll 2RT/ZF = 25 \text{ mV}$$

$$\equiv -69(AcZc^{1/2})^{-1} \text{ mV}, \quad T = 25° \text{ C} \tag{6}$$

For highly charged membranes the corresponding result is found from eqn. (ii)

$$\psi_0 \equiv (2kT/Ze) \text{ in } [(Ze/kT)(\sigma_{el}/\varepsilon\varepsilon_0)] \gg 2RT/ZF$$

$$\equiv -51.4Z^{-1} \text{ in } (0.36\ A_c c^{1/2} \text{ mV}, T = 25°\text{C}) \tag{7}$$

In the numerical result the area per charge, A_c, is in nm^2 and the concentration in mol L^{-1}.

In this study, pericardium falls under the category of low charged membrane, hence equation (6) holds good. Results summarized in the figures are calculated using this equation (6).

EXPERIMENTAL

The pericardial membrane of buffalo (*Bof. bubalis*) aged between 18–24 months was taken out and immediately submerged into ice-cold Ringer's solution of pH 7.4 ± 10.2 for the preservation of membrane tissues. The following composition in g/L for Ringer's solution was used: NaCl 8.00, KCl 0.20, $CaCl_2$ 0.20, $NaHCO_3$ 1.00, NaH_2PO_4 0.05, $MgCl_2$ 0.10 and glucose 1.00.

The pericardium was washed with deionised water several times to remove Ringer's solution. The membrane was then cut into disc type and installed between the two flanges of Pyrex glass cell. The values of electrical potential difference arising across the membrane were used as such as recorded in our previous paper[7] using the Osaw vernier potentiometer (CAT. No. 37001) by maintaining a ten-fold difference in electrolyte's concentration, such that $C_2/C_1 = 10$.

RESULTS AND DISCUSSION

The values of membrane surface potential (ψ_0) were calculated by using the Poissoen-Boltzmann equation. Eqns. 6 and 7 were used for the calculation of ψ_0 for the pericardial membrane. The pericardium is having low charge surface density. These values calculated using eqn. (6) at 25 ± 0.1°C hold good and reveal that this membrane falls under the category of low surface charge density. Calculations were done using the values of surface charge density (A_c) calculated by the method adopted by Arif. *et al.*[8]

It is obvious from Tables 1–4 that the value of membrane surface potential increases with the decrease in the concentration of electrolytes. This trend is in good accordance with the fact that as the ions moved into the membrane interior it created a local deformation. More the transport of ions, more the deformation in the membrane surface texture. Therefore at higher concentration the transport of ion increases. As a result, there is more deformation in the membrane surface texture. Therefore, membrane surface potential is decreased.

It is clear from Tables 1-4 that the values of membrane surface potential are higher when calculated considering the membrane as a little charged surface than those calculated by considering it as a highly charged membrane.

Comparison of the results of eqns. (6) and (7) and interpretation of established electrostatic theories of biomembranes tends towards the positive charge of pericardial membrane. This is also obvious from the result that pericardial membrane comes into the range of highly charged biomembranes. Figs. 1–4 give good account of the relative permeability of different metal ions in accordance to their ionic radii.

Different values of membrane surface potential were plotted against negative logarithm of concentration. Figs. 1–4 show that at lower values of –log C the membrane surface potential increases comparatively solely which increases sharply at higher values of –log C. This trend justifies the Gouy-Chapman model of electric double layer formed at the interface of pericardial membrane and

TABLE-1

VALUES OF MEMBRANE SURFACE POTENTIAL WITH DIFFERENT CONCENTRATIONS OF ELECTROLYTES WITH PERICARDIAL MEMBRANE ($\gamma = C_2/C_2 = 10$)

S. No.	Concentration (c) of electrolytes in mol/L	Name of electrolyte	Values of charge density used in calculation calculated by Kobatake's method $\theta_d \times 10^2$	Values of membrane surface potential (in millivolt) calculated by eqn. (6)
1.	0.5	NaCl	20.16	–4.842
		NaF	08.84	–11.041
		KCl	04.04	–24.159
		KF	13.71	–7.119
		NH_4Cl	—	—
2.	0.2	NaCl	20.16	–7.656
		NaF	08.84	–17.463
		KCl	04.04	–38.227
		KF	13.71	–11.259
		NH_4Cl	—	—
3.	0.1	NaCl	20.16	–10.823
		NaF	08.84	–24.709
		KCl	04.04	–54.048
		KF	13.71	–15.927
		NH_4Cl	—	—
4.	0.05	NaCl	20.16	–15.348
		NaF	08.84	–35.000
		KCl	04.04	–76.590
		KF	13.71	–22.571
		NH_4Cl	—	—
5.	0.02	NaCl	20.16	–24.278
		NaF	08.84	–55.377
		KCl	04.04	–121.233
		KF	13.71	–35.493
		NH_4Cl	—	—
6.	0.01	NaCl	20.16	–34.226
		NaF	08.84	–78.054
		KCl	04.04	–170.792
		KF	13.71	–50.328
		NH_4Cl	—	—

aqueous phase. Dissimilar behaviour of NaCl is due to the smaller size of sodium ion. The gross of ions in double layers is weekly associated with the membrane surface only negatively. A few ions bind to the membrane components and these ions then may affect the membrane and structure significantly. This binding of ions is more for the ions having smaller size. Binding affinity of polyvalent ions is more than of univalent ions. We have used only the univalent ions, therefore, their binding affinity is in good accordance with their ionic size. This trend is

TABLE-2

VALUES OF MEMBRANE SURFACE POTENTIAL WITH DIFFERENT CONCENTRATIONS OF ELECTROLYTES WITH PERICARDIAL MEMBRANE ($\gamma = C_2/C_2 = 10$)

S. No.	Concentration (c) of electrolytes in mol/L	Name of electrolyte	Values of charge density used in calculation calculated by Kobatake's method $\theta_c \times 10^2$	Values of membrane surface potential (in millivolt) calculated by eqn. (6)
1.	0.5	NaCl	10.70	–9.122
		NaF	04.93	–19.799
		KCl	03.94	–24.775
		KF	06.58	–14.810
		NH_4Cl	06.71	–14.547
2.	0.2	NaCl	10.70	–14.429
		NaF	04.93	–31.320
		KCl	03.94	–39.182
		KF	06.58	–23.461
		NH_4Cl	06.71	–23.007
3.	0.1	NaCl	10.70	–20.408
		NaF	04.93	–44.315
		KCl	03.94	–55.421
		KF	06.58	–31.189
		NH_4Cl	06.71	–32.547
4.	0.05	NaCl	10.70	–28.911
		NaF	04.93	–62.784
		KCl	03.94	–78.587
		KF	06.58	–47.324
		NH_4Cl	06.71	–46.122
5.	0.02	NaCl	10.70	–45.755
		NaF	04.93	–99.222
		KCl	03.94	–124.203
		KF	06.58	–74.433
		NH_4Cl	06.71	–72.930
6.	0.01	NaCl	10.70	–64.485
		NaF	04.93	–139.959
		KCl	03.94	–174.242
		KF	06.58	–104.863
		NH_4Cl	06.71	–102.831

apparent from Figs. 1–4. It is evident from the figures that as soon as the negative value of –log C increases, the values of membrane surface potential also increases which is a symbol of decrease of ion transport across the membrane when the concentration falls down. Since the pericardial tissue is a very sensitive tissue and is extensively used in constructing a bioprosthetic heart valve, this behaviour may help in the survival of heart functioning under loss of trace metal ions concentration.

TABLE-3

VALUES OF MEMBRANE SURFACE POTENTIAL WITH DIFFERENT CONCENTRATIONS OF ELECTROLYTES WITH PERICARDIAL MEMBRANE ($\gamma = C_2/C_2 = 10$)

S. No.	Concentration (c) of electrolytes in mol/L	Name of electrolyte	Values of charge density used in calculation calculated by Kobatake's perm selectivity method $P_a \cdot vs(C_1 + C_2)/2\ \theta_x \times 10^2$	Values of membrane surface potential (in millivolt) calculated by eqn. (6)
1.	0.5	NaCl	15.14	–6.446
		NaF	05.01	–19.480
		KCl	—	—
		KF	06.07	–16.080
		NH_4Cl	08.91	–10.954
2.	0.2	NaCl	15.14	–10.196
		NaF	05.01	–30.817
		KCl	—	—
		KF	06.07	–25.433
		NH_4Cl	08.91	–17.327
3.	0.1	NaCl	15.14	–14.423
		NaF	05.01	–43.588
		KCl	—	—
		KF	06.07	–35.974
		NH_4Cl	08.91	–24.511
4.	0.05	NaCl	15.14	–20.424
		NaF	05.01	–61.772
		KCl	—	—
		KF	06.07	–50.997
		NH_4Cl	08.91	–34.743
5.	0.02	NaCl	15.14	–32.333
		NaF	05.01	–97.733
		KCl	—	—
		KF	06.07	–80.619
		NH_4Cl	08.91	–54.922
6.	0.01	NaCl	15.14	–45.574
		NaF	05.01	–137.772
		KCl	—	—
		KF	06.07	–113.673
		NH_4Cl	08.91	–77.441

Variation in the values of membrane surface potential as a function of bulk salt concentration. In case the ion binds to the membrane and partly neutralizs the net surface charge, this neutralization of charge differs at different concentrations At higher concentration this neutralization is more, reducing the negative values of membrane surface potential. The slight deviation from the normal behaviour of the graphs is due to the fact that the inorganic monovalent cations

TABLE-4
VALUES OF MEMBRANE SURFACE POTENTIAL WITH DIFFERENT CONCENTRATIONS OF ELECTROLYTES WITH PERICARDIAL MEMBRANE ($\gamma = C_2/C_2 = 10$)

S. No.	Concentration (c) of electrolytes in mol/L	Name of electrolyte	Values of charge density used in calculation calculated by Tasake's method $\theta_x \times 10^2$	Values of membrane surface potential (in millivolt) calculated by eqn. (6)
1.	0.5	NaCl	04.11	–23.752
		NaF	01.29	–75.657
		KCl	04.76	–20.505
		KF	01.64	–61.883
		NH_4Cl	01.90	–51.377
2.	0.2	NaCl	04.11	–37.561
		NaF	01.29	–119.784
		KCl	04.76	–32.440
		KF	01.64	–94.133
		NH_4Cl	01.90	–81.213
3.	0.1	NaCl	04.11	–53.158
		NaF	01.29	–169.533
		KCl	04.76	–45.877
		KF	01.64	–133.204
		NH_4Cl	01.90	–114.954
4.	0.05	NaCl	04.11	–75.327
		NaF	01.29	–240.396
		KCl	04.76	–65.032
		KF	01.64	–189.041
		NH_4Cl	01.90	–163.120
5.	0.02	NaCl	04.11	–119.066
		NaF	01.29	–379.350
		KCl	04.76	–102.807
		KF	01.64	–298.391
		NH_4Cl	01.90	–257.558
6.	0.01	NaCl	04.11	–167.883
		NaF	01.29	–534.883
		KCl	04.76	–144.957
		KF	01.64	–420.731
		NH_4Cl	01.90	–363.157

normally do not bind to uncharged phospholipids, such as phosphatidy chalice, while polyvalent cations may do so[7, 9, 10]. The membrane texture and membrane surface potential play a pivotal role in interpreting the ion transport. Membrane permeability of ions increases with the ion size and decreases with the interfacial hydration (Tables 1–4).

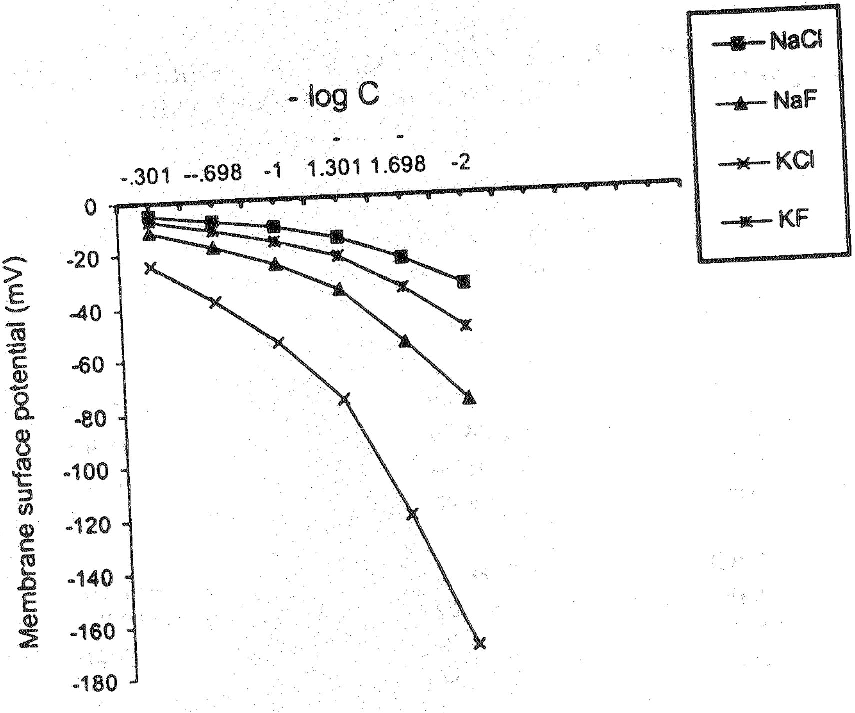

Fig. 1. Plot of membrane surface potential *vs.* –log C

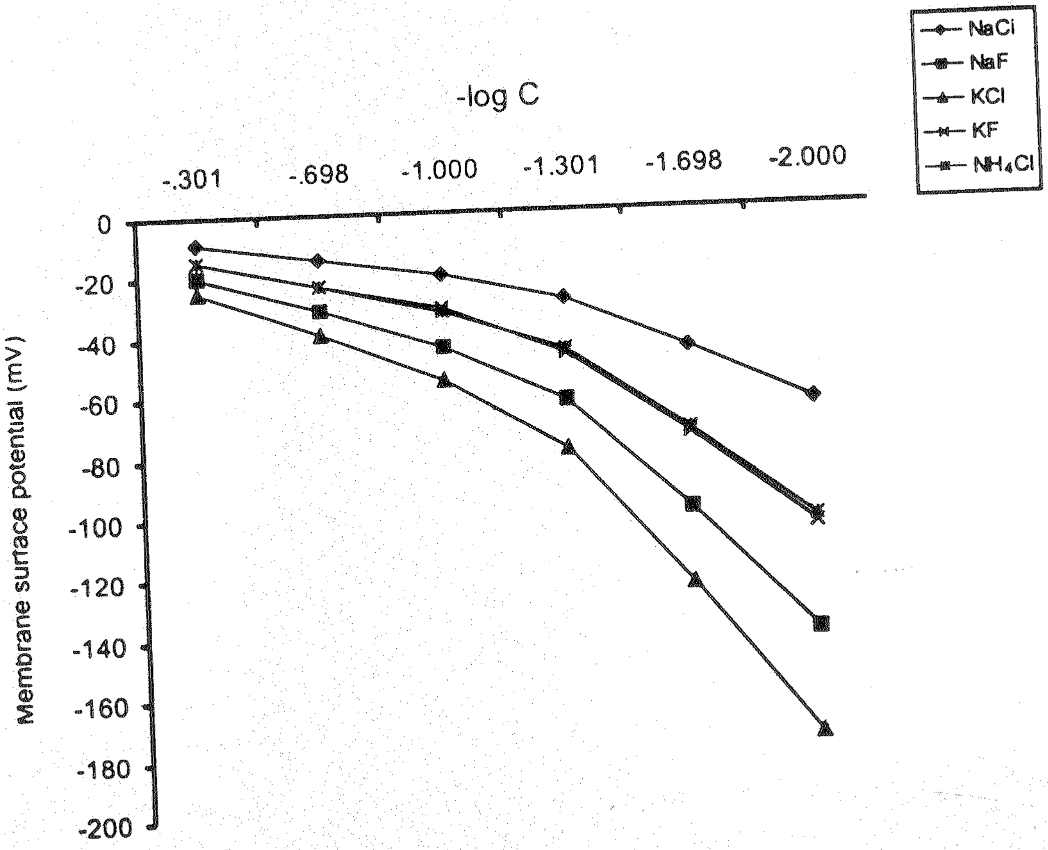

Fig. 2. Plot of membrane surface potential *vs.* –log C

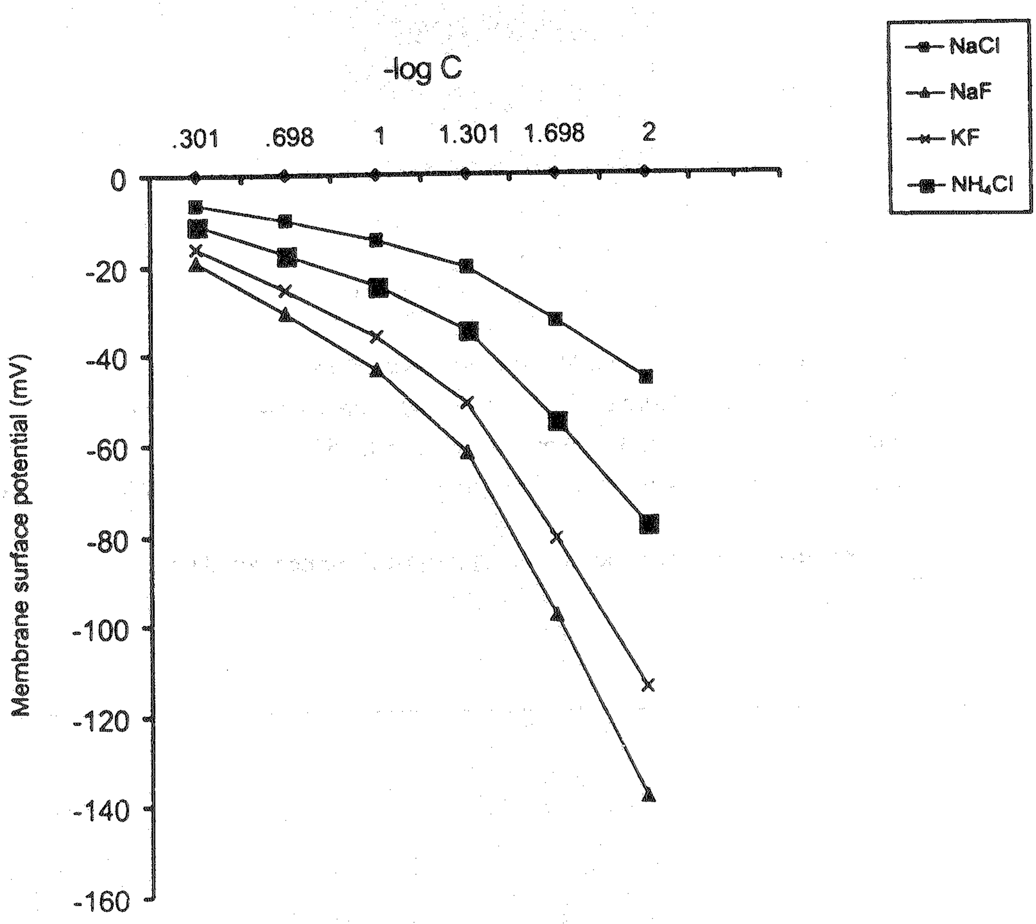

Fig. 3. Plot of membrane surface potential *vs.* –log C

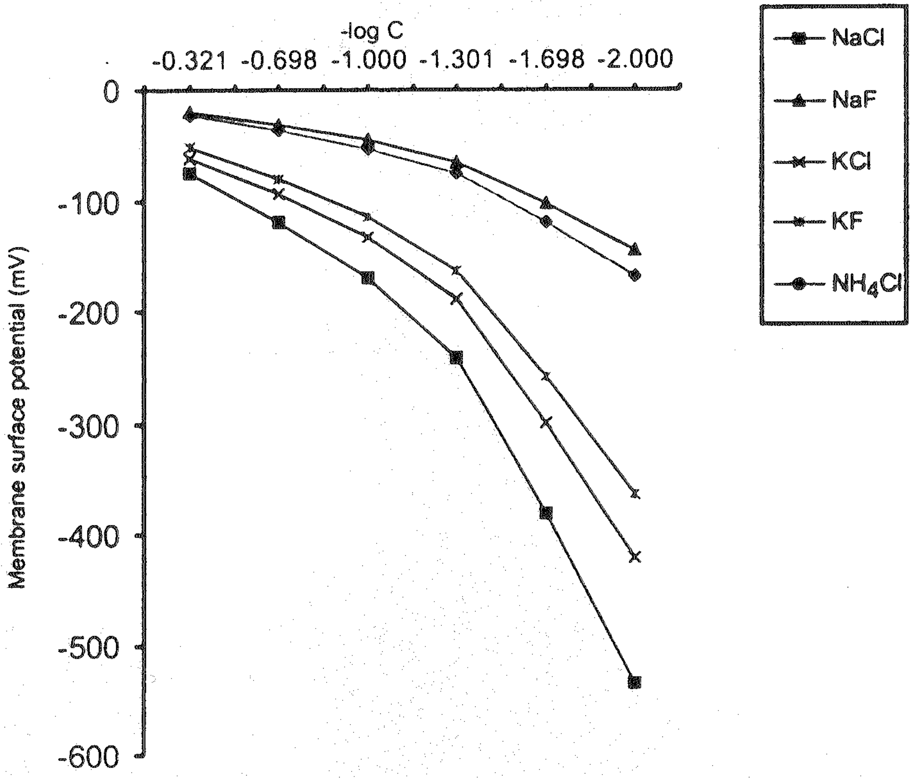

Fig. 4. Plot of membrane surface potential *vs.* –log C

REFERENCES

1. J.R. Buysman and F.T. Koide, *J. Theor. Biol.*, **32**, 1 (1971).
2. M. Nakagaki and R. Takagi, *Chem. Pharm. Bull.*, **34**, 957 (1986).
3. Y.N. Antonenko and L.S. Yaguzhinsky, *J. Bioenerg. Biomembr.*, **14**, 457 (1982).
4. G. Gouy, *J. Phys. Radium (Paris)*, **9**, 457 (1910).
5. D.L. Chapman, *Philos. Mag. (London)*, **25**, 475 (1913).
6. E.J.W. Verwey and J.T.C. Overbeek, Theory of the Stability of Lyophobic Colloids, Elsevier, New York (1948).
7. K.P. Singh, J. Nain and M. Ahmad, *Membrantechnika (Hungary)*, **7**, (2003).
8. M. Ahmad, H. Arif and H.R. Khan, *Bull. Chem. Soc. (Japan)*, **64**, 3115 (1991).
9. J. Westman and L.E. Eriksson, *Biochem. Biophys. Acta*, **557**, 62 (1979).
10. S..\. Tatulian, *Biochem. Biophys. Acta*, **736**, 189 (1983); *Eur. J. Biochem.*, **170**, 413 (1987).

(*Received*: 23 September 2004; *Accepted*: 5 September 2005) AJC-4367