ELECTROSTATIC RADIUS OF THE GRAMICIDIN CHANNEL DETERMINED FROM VOLTAGE DEPENDENCE OF H^+ ION CONDUCTANCE

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ABSTRACT The results of Decker and Levitt (1987) suggest that the conductance of H^+ ion through the gramicidin channel is limited primarily by diffusion in the bulk solution at the channel mouth. It is assumed in this paper that the H^+ conductance is 100% diffusion limited. This means that all the factors that influence the H^+ flux are external to the channel and are presumed to be known. In particular, the diffusion coefficient of H^+ in this region is assumed to be equal to the bulk solution value and the only force acting on the ion is that due to the applied voltage. A model of the H^+ flux is derived, based on the Nernst-Planck equation. It has three adjustable parameters: the electrostatic radius, the capture distance, and the radius of the H^+ ion. The acceptable range of the parameters was determined by comparing the predictions of the model with the experimental measurements of the H^+ conductance at pH 3.75. The best fit was obtained for an electrostatic radius in the range 2.3–2.7 Å. This is in good agreement with earlier predictions (2.5 Å) based on the assumption that the dielectric constant of the channel water is equal to that of bulk water. The addition of 1 M choline Cl^- (an impermeant) increases the H^+ current at low voltage and decreases it at high voltage. The increase can be explained by the small surface charge that results from the separation of charge produced by exclusion of the large choline cation (relative to Cl^-) from the membrane surface. The decrease at high voltages can be accounted for by the change in the profile of the applied potential produced by the increase in ionic strength.

INTRODUCTION

There have been a number of studies that have tried to obtain information about the structure of the gramicidin channel from measurements of its cation conductance (Hladky and Haydon, 1984; Polymeropoulos and Brickmann, 1985). The success of these studies has been limited, primarily because not enough is known about the kinetics of ion transport within the channel. It should be possible to avoid most of these unknowns by analyzing the conductance of H^+ because it seems to be limited almost entirely by diffusion in the bulk solution (Decker and Levitt, 1987)—a process that is, presumably, quantitatively understood. Here, a theoretical model of the H^+ conductance (in the absence of formic acid) will be developed, based on the assumption that H^+ is 100% diffusion limited. By comparing the predictions of the model with the experimental H^+ conductance, some definite inferences can be made about the structure of the gramicidin channel.

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There are two aspects of the experimental results that must be fit by the model: the absolute conductance and the voltage dependence of the conductance. It is relatively easy to fit the absolute conductance by varying, for example, the "capture" distance of the channel (the position where the ion falls into the attractive energy well of the channel). In order to fit the voltage dependence, part of the drop in the applied voltage must occur in the bulk solution. The magnitude of this voltage drop depends on the "electrostatic radius" of the channel. This is the effective radius of the high dielectric, aqueous region of the channel and is an important parameter for characterizing the gramicidin channel, playing an important role in determining the cation conductance. The electrostatic radius is a function of the dielectric constant and radius of the region occupied by the channel water, and the dielectric constant and thickness of the polar inner wall of the channel, all of which are unknown. The most important result of this comparison between the diffusion-limited model and the experimental H^+ conductance is that it places narrow limits on the possible values of this electrostatic radius.

The exact solution for the voltage profile as a function of the electrostatic radius requires a complicated numerical solution (Jordan, 1982). In addition, it is difficult to incorporate the influence of ionic strength into this exact solution. To avoid these problems, an approximate analyti-

cal solution is derived for the profile of the applied voltage external to the channel. This solution differs from the exact solution by only a few percent and can be easily modified to include ionic strength effects.

Factors That Contribute to a Voltage Drop in the Bulk Solution

There are three different factors that produce a voltage drop and it is useful to clearly define each of them since they are often confused.

Voltage Drop for a Pure Lipid Membrane. For a pure lipid membrane (no ion channels) a fraction of the total voltage drop will be in the bulk solution because the aqueous double-layer behaves like a capacitor. Hainsworth and Hladky (1987) have recently discussed the theoretical and experimental influence of this voltage drop on the gramicidin channel conductance. This effect is important only at very low ionic strength and is relatively insignificant in these studies. The voltage dependence of the experimental H^+ conductance used in this paper (Decker and Levitt, 1987) was measured in ionic solutions with concentrations as high as 27 mM $MgCl_2$, an ionic strength for which this effect is negligible. In these experiments, there was no significant difference of the voltage dependence in the solutions containing 2 mM $MgCl_2$ (plus 5 mM aspartic acid buffer), the lowest ionic strength used, versus 27 mM $MgCl_2$. The results in the figures are the average values from experiments in which Mg^{++} varied from 2 to 27 mM.

Equilibrium Voltage Drop at the Mouth of an Ion Channel. The channel represents a high dielectric hole in the low dielectric membrane and this results in a finite voltage drop in the bulk solution at the channel mouth. An analytical expression is derived here for the dependence of this voltage drop on the size of the hole and on the ionic strength. (Andersen [1983] has developed another approximation for this effect.) This is the only factor that is important for the analysis of the H^+ conductance presented here.

Voltage Drop Resulting from Flux of Permeant Ion. The above two factors do not depend on the presence of an ion current and will be present at equilibrium when there is no flux (i.e., no permeable ions). This third factor arises directly from the ion flux that produces a depletion of permeable ions at one end of the channel and an excess at the other end. This nonequilibrium ion distribution produces a voltage gradient. Lauger (1976) derived an approximation for this effect based on an assumption of electroneutrality. Recently, Levitt (1985) has presented an exact solution for the potential (Φ) based on Poisson's equation, modified for the conditions of a high dielectric channel in a low dielectric membrane:

$$\frac{d}{dX}\left(S\frac{d\Phi}{dX}\right) = -(4\pi e/\epsilon_w)\,\Sigma z_i C^i S^i_e. \qquad (1)$$

The sum on the right side is over all the ions, permeable and nonpermeable. For the case considered in this paper where H^+ is the only permeable species, the contribution of the permeable ions to the voltage drop (Eq. 1) is negligible because H^+ is present in such a low concentration (2.7×10^{-4} M). The contribution of the impermeable ions to the voltage gradient (Eq. 1) is important and is included in the ionic strength influence on the equilibrium voltage drop discussed above (see Eq. 5).

Derivation of the Flux Equation

At pH 3.75, only a small fraction of the channels are occupied by H^+ ions (Decker and Levitt, 1987), so that there is no interaction between ions and the flux can be described by the classical Nernst-Planck equation (Levitt, 1986):

$$J = C_0(e^{\psi_0} - e^{-\psi_0})/H \qquad U = \Phi + \Psi; \qquad u = (e/kT)U$$

$$H = \int_{-\infty}^{\infty} h(X)dX; \qquad h(X) = e^u/(DS_e); \qquad (2)$$

where D is the diffusion coefficient, S_e is the "effective" area available to the H^+ ion, $+\psi_0$ and $-\psi_0$ are the bulk values of the applied voltage on the left side and right side, C_0 is the symmetric bulk solution concentration, and u is the dimensionless total potential which is the sum of the applied voltage (ψ) and the intrinsic channel potential (ϕ). The integral H can be divided into an integral over the bulk solutions on the left and right side and over the "channel":

$$H = \int_{-\infty}^{-L-W} h(X)\,dX + \int_{-L-W}^{L+W} h(X)\,dX + \int_{L+W}^{-\infty} h(X)\,dX, \qquad (3)$$

where the physical channel extends from $-L$ to $+L$ and W defines the distance from the channel end where the H^+ ion is "captured" by the channel. The three integrals correspond to the magnitude of the generalized resistances of the three regions, arranged in series.

In general, the potential energy (u), area (S_e), and diffusion coefficient (D) in the channel are unknown, and must be either guessed or determined empirically by fitting the experimental results. However, the transport of H^+ in the gramicidin channel presents a unique case where the transport is nearly bulk solution limited, i.e., the resistance of the bulk solutions (the first and third integral in Eq. 3) represents ~95% of the total resistance (Decker and Levitt, 1987). Thus, the gramicidin channel has a very high H^+ conductance, relative to the bulk solutions, presumably a result of a high diffusion coefficient and/or large negative potential energy in the channel. It will be assumed here

that the H^+ is 100% diffusion limited so that the second integral in Eq. 3 can be neglected. It will also be assumed that, in the region of bulk solution external to the capture position, Φ is zero and D is equal to the bulk solution value (D_0). This is an approximation to the actual situation where the attractive potential that "captures" the ion must vary continuously from its value in the channel to zero in the bulk solution. Although the origin of this attractive potential in the gramicidin channel is unknown, it must be of short range since gramicidin is uncharged. The capture distance W should be regarded as an empirical (and adjustable) constant that characterizes the position where this attractive potential becomes significant. Using the antisymetrical property of ψ, H can then be written as:

$$D_0H = e^{\psi_0}\int_{-\infty}^{-L-W} dXe^{\psi-\psi_0}/S_e + e^{-\psi_0}\int_{L+W}^{\infty} dXe^{\psi+\psi_0}/S_e$$
$$\simeq (e^{\psi_0} + e^{-\psi_0})I_1 + (e^{-\psi_0} - e^{\psi_0})I_2$$
$$I_1 = \int_{L+W}^{\infty} dX/S_e; \quad I_2 = \int_{L+W}^{\infty} dX(\psi_0 + \psi)/S_e, \qquad (4)$$

where the second quality results from the expansion of the exponentials, which is valid because <10% of the voltage drop occurs in the bulk solution (see Fig. 2).

An analytical approximation for the applied voltage profile (ψ) has been derived previously (Levitt, 1987). In Eq. 4, only the profile in the bulk solution is needed and the derivation has been slightly modified to increase the accuracy of this result. The modification consists of defining two radii (Fig. 1): the electrostatic radius of the channel (a) and another, larger, radius just outside the channel mouth (d). It is assumed that the field lines are confined to a cylinder of radius a in most of the channel, and spread into a radius d at the channel end (see Levitt, 1987 for details). The radius d was then empirically adjusted to fit the exact numerical calculations of Jordan (1982) for the potential external to the channel. The modified expression for ψ is written in terms of dimensionless variables ($x = X/d$; $\ell = L/d$; $w = W/d$; $c = d^3C$) normalized by the length d (in earlier publications, the length a was used to normalize the variables):

$$1 + \psi/\psi_0 = \begin{cases} Q\exp[-\kappa(x - \ell - 1)]/(x - \ell) & \ell + 1 \le x \le \infty \\ Q\{1 + 2(1 + \kappa)[\pi/4 - \tan^{-1}(x - \ell)]\} & \ell \le x \le \ell + 1 \\ Q\{1 + 2(1 + \kappa)[\pi/4 + (\ell - x)k^2]\} & 0 \le x \le \ell \end{cases} \qquad (5)$$

$$Q = [1 + 2(1 + \kappa)(\pi/4 + \ell k^2)]^{-1}; \quad \kappa^2 = 8\pi\gamma c_i;$$
$$\gamma = e^2/(\epsilon_W kTd); \quad k = d/a,$$

where $\kappa = d/\text{Debye length}$ and c_i is the dimensionless impermeant concentration. Choosing $d = 1.5a$ ($k = 1.5$), this approximation becomes very good, agreeing to within 1% of the exact numerical external ($x > \ell$) potential

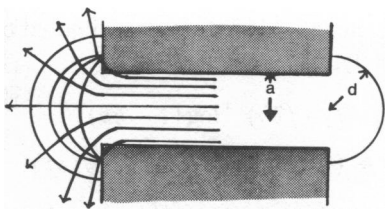

FIGURE 1 Diagram of assumed allignment of field lines used to derive expression for the profile of the applied voltage (Eq. 5). The field lines are constained within the electrostatic radius (a) in the channel and diverge into a larger radius (d) at the channel end. The value of d was adjusted in order to find the best fit to the exact numerical solution for the potential outside the channel. The figure is drawn with $d/a = k = 1.5$, the value that gave the best fit.

(Jordan, 1982) for L/a varying from 1.25 to 10. Fig. 2 shows this voltage profile in the low ionic strength limit for the bulk solution region at the right end ($x > 0$) of the gramidicin channel (half length = L = 12.5 Å) for different values of the electrostatic radius (a). The profile has been rescaled so that the voltage in the bulk solution is zero and $V_{tot} = 2\psi_0$.

An expression for the "effective" area (in terms of the dimensionless variable $s_e = S_e/d^2$) available to the H^+ ion has been derived previously (Levitt, 1987):

$$s_e/\pi = \begin{cases} g + (x - \ell)^2 f & \ell \le x \le \ell + k^{-1} \\ 2(x - \ell)(x - \ell - b) & \ell + k^{-1} \le x \end{cases} \qquad (6)$$
$$g = (k^{-1} - b)^2, \qquad f = 1 - k^2b^2,$$

where B is the "effective" radius of the H^+ ion and $b = B/d$. It is assumed in Eq. 6 that the electrostatic radius (a) is equal to the physical radius available to the H^+ ion. This should not reduce the generality of the result since the effective radius of the H^+ ion ($b = B/d = k^{-1}B/a$) is treated as an arbitrary adjustable parameter in fitting the experimental results.

The integrals I_1 and I_2 are then obtained by carrying out the integrations in Eq. 4, using Eqs. 5 and 6. (In this

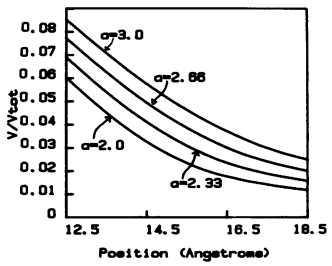

FIGURE 2 Profile of applied voltage (Eq. 5) at the right end of the gramicidin channel (half length = 12.5 Å) relative to the total transmembrane voltage for different values of the electrostatic radius (a) for the case where $\kappa = 0$. The profile of Eq. 5 has been rescaled so that the voltage (V) in the right bulk solution is zero and $V_{tot} = 2\psi_0$.

integration, the $\tan^{-1}[y]$ was approximated by $y - y^3/3$). The final analytical expression is

$$\begin{aligned}
\pi d I_1 &= I_3 - (2b)^{-1}\log(1-kb)\\
\pi d I_2/Q\psi_0 &= (1+\pi t/2)I_3 - t[1+g/(3f)]\\
&\quad \cdot f^{-1}\log[(g+f/k^2)/(g+fw^2)]\\
&\quad + [t/(3f)](k^{-2}-w^2)\\
&\quad + (2b)^{-1}(1+\pi t/2)\log[(1-b)/(1-bk)]\\
&\quad - t(1-b^2/3)\log[(k-bk)/(1-bk)]\\
&\quad + (t/6)(1-k^{-2}) + (tb/3)(1-k^{-1})\\
&\quad + 0.5e^{\kappa}\{Ei(-\kappa) - e^{-b\kappa}Ei[-\kappa(1-b)]/b^2\\
&\quad - [\kappa Ei(-\kappa) + e^{-\kappa}]/b\}\\
t &= 1+\kappa; \quad I_3 = \{\tan^{-1}[k^{-1}(f/g)^{1/2}]\\
&\quad - \tan^{-1}[w(f/g)^{1/2}]\}(gf)^{-1/2}, \qquad (7)
\end{aligned}$$

where *Ei* is the exponential integral function. The H^+ flux is then obtained from Eqs. 2, 4, and 7.

Comparison of Theoretical and Experimental Values of H^+ Conductance at Low Ionic Strength

There are three adjustable parameters in this expression for the H^+ flux: the electrostatic radius (a); the capture distance ($w = W/d$); and the radius of the H^+ ion ($b = B/d$). As b increases, the cross-sectional area available to the ion decreases, decreasing the flux. As a increases, the voltage drop in the bulk solution increases with a corresponding increase in the voltage dependence of the flux. As w increases, the H^+ ion is "captured" further out in the bulk solution so that the ion sees less of the voltage drop and the voltage dependence is decreased. It will be assumed that w ranges from a minimum of 0 to a maximum of 1 ($W = d$). Values of w outside this range do not seem to be physically likely.

Fig. 3 shows a comparison of the experimental (●) and theoretical H^+ current for the entire range of parameters that can fit the data for the gramicidin channel ($2L$ = 25 Å). Each panel is for one value of the electrostatic radius (a), ranging from 2 to 3 Å. For each curve, b was chosen so that the current matched the experimental value at 100 mv and then w was varied to give the best fit to the voltage dependence. For a =2.0 Å, the theoretical curves have too little voltage dependence, even for the extreme value $w = 0$. Similarly, for a = 3 Å, the theoretical curves have too much voltage dependence, even for the extreme value w = 0.9. Good fits to the experimental results can be obtained for values of a ranging from 2.33 Å (w = 0–0.5) to 2.67 Å (w = 0.5–0.8). Although this analysis suggests that the electrostatic radius of the gramicidin channel is probably in the range of 2.3 to 2.7 Å, a radius in the range of 2–3 Å cannot be ruled out, given the number of assumptions that were made in deriving this result.

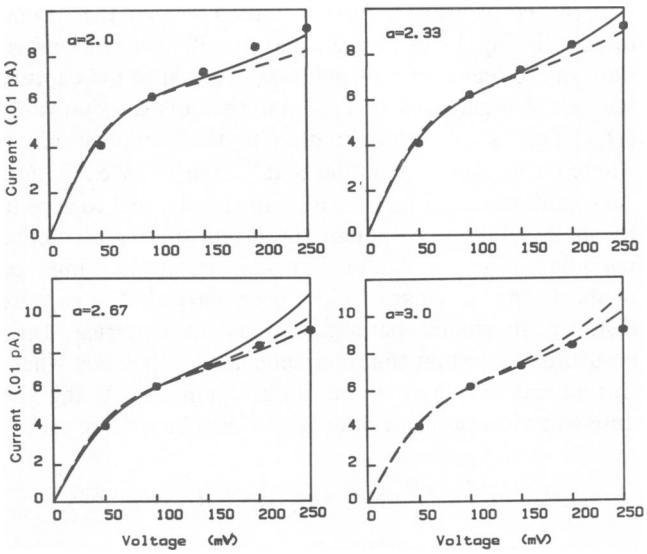

FIGURE 3 Variation of parameters to find the best fit of the theoretical H^+ current (lines) to the experimental results (●) at pH 3.75 and low ionic strength. Each panel is for a different value of the electrostatic radius (a). For each value of w (capture distance), b (H^+ ion radius) was adjusted to fit the data at 100 mV. The theoretical lines correspond to the following sets of parameters: a = 2 Å (w = 0, b = 0.255, ——; w = 0.5, b = 0.605, ---); a = 2.33 (w = 0, b = 0.28, ——; w = 0.5, b = 0.62, ---); a = 2.67 (w = 0, b = 0.3, ——; w = 0.5, b = 0.635, ---; w = 0.8, b = 0.843, —— ——); a = 3 (w = 0.5, b = 0.255, ---; w = 0.9, b = 0.91, —— ——).

Comparison at High Ionic Strength

The theoretical equation for the potential profile (Eq. 5) includes the influence of the ionic strength through the parameter κ (d/Debye length). This expression was derived by assuming that the inert electrolyte was excluded from a hemispherical region of radius d ($=1.5a \simeq 3.7$Å) at the channel mouth (Levitt, 1987), a distance approximately equal to the radius of choline. The presence of inert electrolyte external to this hemisphere screens the potential, reducing the fraction of the applied voltage difference that is external to the channel. Fig. 4 shows the decrease in the external potential produced by the addition of 1 M choline.

The experimental single-channel H^+ current at pH 3.75 in the absence (●) and presence (*) of 1 M choline Cl^- is shown in Fig. 5. It can be seen that choline increases the conductance at low voltages and decreases it at high voltages. The decrease at high voltage is expected since the increase in ionic strength will decrease the voltage drop in the bulk solution (see Eq. 5 and Fig. 4). The increase at low voltages is more surprising and probably results from the large size of the choline ion relative to the Cl^- ion (Carnie and McLaughlin, 1983). The small Cl^- can get closer to the membrane, creating a negative surface charge that

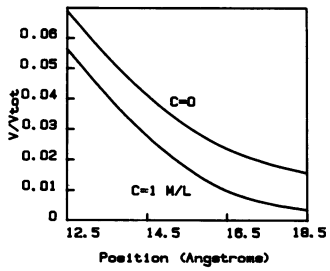

FIGURE 4 Profile of applied voltage (V) at end of gramicidin channel for electrostatic radius (a) of 2.33 Å in the absence ($C = 0$) and presence ($C = 1$) of 1 M large impermeant ion, scaled as in Fig. 2.

increases the concentration of H^+ at the channel mouth. This effect is treated approximately in the Appendix where it is shown that it is of the right size to account for the observed increase in conductance. This negative surface charge should increase the H^+ concentration and therefore, the conductance, by some constant factor at all applied voltages. To correct for this surface charge, the experimental H^+ conductance in the presence of choline was scaled so that it was equal to the low ionic strength conductance at 50 mV (as one would expect theoretically, see Fig. 5). This scaled data (o) are shown in Fig. 5.

The lines in Fig. 5 are the theoretical H^+ current in the absence ($C = 0$) and presence ($C = 1$) of 1 M choline Cl^- and for the case where there is no voltage drop in bulk solution (C = infinite). The same set of parameters ($a = 2.33$ Å, $w = 0.2$, $b = 0.4$) was used for all three curves. The agreement between the scaled experimental data (o) and the theory for 1 M choline is quite good, providing additional support for the validity of the model and the accuracy of the estimate of the electrostatic radius (a).

DISCUSSION

The electrostatic radius is an important parameter for characterizing the ion transport properties of an ion channel. This radius and the pore length completely define the magnitude of the electrostatic potentials of the channel; including the Born image potential, the potential from a fixed charge in the channel, the ion–ion interaction potential, and the profile of the applied voltage (Levitt, 1987). The dielectric constant of the channel should vary continually as one moves out from the center of the water-filled pore, through the polar inner and nonpolar outer peptide regions of the gramicidin channel. Even the concept of a continuum dielectric constant is questionable on the local scale of this problem. This complicated system is usually replaced by an "equivalent" electrostatic radius (defined as the radius of a channel with the dielectric constant of bulk water ($\epsilon = 78$) in a lipid ($\epsilon = 2$) membrane) that mimics the electrostatic potential on the axis of the real channel (Jordan, 1984). The electrostatic radius (a) determined here from the H^+ conductance corresponds to this "equivalent" radius.

The major uncertainty about the magnitude of this equivalent radius is the value that should be used for the dielectric constant of water in the channel. It is not clear whether this single file of oriented water molecules should have the same dielectric constant as bulk water. Jordan (1984) estimated that the equivalent radius should be ~2.5 Å, based on the assumption that the 2 Å radius hole in the channel was filled with water that had a dielectric constant equal to that of bulk water, along with an estimate of the additional effect of the polar gramicidin wall. This value is remarkably close to the value determined here from the H^+ flux (best estimate 2.3–2.7 Å). However, one must retain some skepticism about this agreement, considering the number of assumptions that have gone into these calculations.

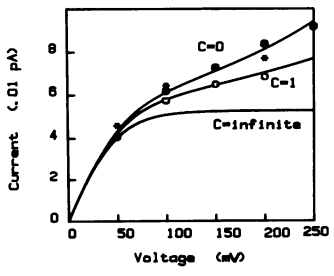

FIGURE 5 Experimental H^+ current in absence (●) and presence (*) of 1 M choline chloride. The results in the presence of 1 M choline have been scaled (O) so that the current at 50 mv is equal to that in the absence of choline. The solid lines correspond to the theoretical current for $a = 2.33$ Å, $w = 0.2$ and $b = 0.4$ in the absence ($C = 0$) and presence ($C = 1$) of 1 M large impermeant ion, and for the case where there is no voltage drop in the bulk solution (C = infinite).

APPENDIX

Surface Potential Resulting from Addition of Large Cation

This surface potential arises from the separation of charge that results from the exclusion of the large cation (and resultant excess of anions) in a region close to the membrane surface. It will be assumed that there is a region of thickness p (equal to the difference between the cation and anion radius) at the membrane surface from which cations are excluded and in which anions are distributed according to their Boltzmann distribution.

The dimensionless form of Poisson's equation can be written as (Carnie and McLaughlin, 1983):

$$\frac{d^2\phi}{d\xi^2} = -\frac{1}{2}\Sigma z_i c_i; \qquad \phi = e\Phi/kT; \qquad c = C/C_0$$

$$\xi = X/\lambda \qquad \lambda^2 = kT\epsilon_w/(8\pi e^2 C_0) = 9.47/M_0, \quad (1A)$$

where λ is the Debye length, C_0 is the bulk solution concentration in number/(length)^3, and, in the last equality, λ is in Angstroms and M_0 is the molar concentration. In Region I ($p \le X \le \infty$; $p/\lambda \le \xi \le \infty$), both ions have a Boltzmann distribution:

$$c_+ = e^{-\phi}; \qquad c_- = e^{\phi}. \quad (2A)$$

The Poisson-Boltzmann equation is then:

$$\frac{d^2\phi}{d\xi^2} = -\frac{1}{2}(e^{-\phi} - e^{\phi}) \simeq \phi. \quad (3A)$$

The exponentials have been expanded because ϕ is small. The solution to this equation, subject to the boundary condition that both ϕ and $d\phi/dx$ are zero at $\xi = \infty$, is

$$\phi_I = Be^{-\xi}. \quad (4A)$$

In Region II ($0 \le X \le p$; $0 \le \xi \le p/\lambda$), the cation is excluded and the Poisson Boltzmann equation becomes

$$\frac{d^2\phi}{d\xi^2} = \frac{1}{2}e^{\phi} \simeq \frac{1}{2}(1 + \phi). \quad (5A)$$

Since it is assumed that there is no surface charge at the membrane surface, the boundary condition is $d\phi/d\xi = 0$ at $\xi = 0$ (Carnie and McLaughlin, 1983). The solution to this equation is

$$\phi_{II} = -1 + A(e^{+\xi/\sqrt{2}} + e^{-\xi/\sqrt{2}}). \quad (6A)$$

Solving for A using the condition that both ϕ and $d\phi/dx$ must be continuous at $\xi = p/\lambda$:

$$A = \sqrt{2}[e^{-(p/\lambda)/\sqrt{2}}(1 + \sqrt{2}) + e^{-(p/\lambda)/\sqrt{2}}(\sqrt{2} - 1)]^{-1}. \quad (7A)$$

The potential at the membrane surface is then

$$\phi_{II}(0) = -1 + 2A. \quad (8A)$$

For $p = 1.5$ Å (assuming a 3.3 Å radius for choline and a 1.8 Å radius for Cl^-) and $M_0 = 1$ M; $A = 0.38$ and $\phi_{II}(0) = -0.23$ (in units of $kT/e \simeq 25$ mV). This surface potential should increase the H^+ ion concentration (and, therefore, the H^+ current) at the membrane surface by a factor of $e^{-\phi} = 1.26$. Experimentally, the H^+ current at 50 mV is increased by a factor of 1.13 by the addition of 1 M choline Cl^-. Since the H^+ ion is captured at some distance from the membrane surface where the potential will be less than the surface potential, this experimental result is consistent with the theory.

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REFERENCES

Andersen, O. S. 1983. Ion movement through gramicidin A channels. Interfacial polarization effects on single-channel current measurements. *Biophys. J.* 41:135–146.

Carnie, S., and S. McLaughin. 1983. Large divalent cations and electrostatic potentials adjacent to membranes. A theoretical calculation. *Biophys. J.* 44:325–332.

Decker, E. R., and D. G. Levitt. 1988. Use of weak acids to determine the bulk diffusion limitation of H^+ Ion conductance through the gramicidin channel. *Biophys. J.* 53:25–32.

Hainsworth, A. H., and S. B. Hladky. 1987. The effects of double-layer polarization on ion transport. *Biophys. J.* 91:27–36.

Hladky, S. B., and D. A. Haydon. 1984. Ion movements in gramicidin channels. *Curr. Top. Membr. Transp.* 21:327–372.

Jordan, P. C. 1982. Electrostatic modeling of ion pores. Energy barriers and electric field profiles. *Biophys. J.* 39:157–164.

Jordan, P. C. 1984. The total electrostatic potential in a gramicidin channel. *J. Membr. Biol.* 78:91–102.

Lauger, P. 1976. Diffusion-limited ion flow through pores. *Biochim. Biophys. Acta.* 455:493–509.

Levitt, D. G. 1985. Strong electrolyte continuum theory solution for equilibrium profiles, diffusion limitation, and conductance in charged ion channels. *Biophys. J.* 48:19–31.

Levitt, D. G. 1986. Interpretation of biological ion channel flux data-Reaction-rate versus continuum theory. *Annu. Rev. Biophys. Biophys. Chem* 15:29–57.

Levitt, D. G. 1987. Exact continuum solution for a channel that can be occupied by two ions. *Biophys. J.* 52:455–466.

Polymeropoulos, E. E., and J. Brickmann, 1985. Molecular dynamics of ion transport through transmembrane model channels. *Annu. Rev. Biophys. Biophys. Chem.* 14:315–330.