Modified Donnan potentials for ion transport through biological ion channels

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In the classical study of ion transport through charged membranes, Donnan potentials are used to approximate the difference between the applied electrostatic potential and the potential at the liquid/membrane interface. For very thin membranes (e.g., biological lipid bilayers), this discontinuous approximation of the potential is not sufficient. Here we derive a modification to the classic Donnan potential for ion transport through a biological ion channel embedded in a lipid bilayer. We also show how to derive the classic Donnan potential without the usual assumptions and estimate the amount of space charge at the liquid/membrane interface.

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I. INTRODUCTION

In the classical study of liquid/membrane interfaces, membranes acquire selectivity because they are charged [1–4]. Such charged membranes allow the movement of counter ions (with charge opposite that of the membrane) and impede the movement of co-ions of the same charge as the membrane. The Donnan potential is the electrical potential difference between the voltage applied to the system and that at the interface. This nonzero potential also changes the concentrations of ions near the membrane away from their values in bulk solution. Classically, the Donnan potential and the associated change in the ion concentrations are generally used as boundary conditions for the drift-diffusion (Nernst-Planck) equations that model the transport through the membrane. In this paper we reexamine these boundary conditions with the goal of improving them for modeling ion transport through biological ion channels.

In general, there are two major kinds of ion transport through membrane: (1) leaky transport through the entire membrane and (2) channel transport through a specialized structure embedded in an otherwise impermeable membrane (a single biological ionic channel in a bilayer membrane, for example). The total current flow is much less through a channel than through a membrane even if the specific conductance of both structures is the same since the channel has so much less area available for current flow. Furthermore, membranes that allow transport through their whole area usually are quite thick in practical applications (say micrometer in thickness) while impermeable lipid bilayers are typically only 2-nm thick. Such differences in length scales fundamentally change the need for accurate boundary conditions. For all charge transport problems it is important to correctly determine the local electrostatic potential and carrier concentrations since they are nonlinearly coupled. For very short membranes this is even more important because the length scales involved amplify any mistakes in the potential and the concentrations; different potentials and concentrations at the edges of the channel give qualitatively different answers both theoretically [5] and experimentally [6].

These differences in total current and length of current path between the two cases require different analyses for the refinement of the Donnan potentials we consider. In this paper we will mainly consider channel transport (case 2) with some discussion of how to apply the results to membrane transport in general (case 1) at the end. Because of this, our analysis will focus on the area around and inside the ''channel'' through the membrane; we call it a channel whether or not it is a biological ion channel.

A. Biological ion channels

Ion channels are cylindrical, hollow proteins that regulate the movement of ions (mainly Na^+, K^+, Ca^{2+}, and Cl^-) across nearly all biological membranes. Since these membranes are otherwise impermeable to charged particles, the only way ions can cross is through the pore that runs down the long axis of a channel. This property has been exploited by evolution to produce many varied phenomena necessary for life: channels are responsible for the initiation and continuation of the electrical signals in the nervous system; in the kidneys, lungs, and intestines, channels coordinate changes in ionic concentration gradients that result in the absorption or release of water; in muscle cells, a group of channels is responsible for the timely delivery of the Ca^{2+} ions that initiate a contraction. Clinically, malfunctioning channels cause cystic fibrosis, cholera, and many other diseases and have recently been implicated in schizophrenia and bipolar disorders. Furthermore, a large number of drugs (including valium and phencyclidine) act directly or indirectly on channels.

To produce such varied and complicated phenomena, channels act in groups, opening and closing at the same time and letting only specific ion types through (for example, selectively passing far more Na^+ ions than K^+ ions). Despite such complex final results, it is possible to remove a single

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channel from the biological system and study it as an isolated physical system. (This is rarely possible to do with other objects in biology and still have interesting results.) To do this, an individual channel is placed in a phospholipid membrane that separates two baths of known ionic concentration. A voltage is applied to the system by electrodes in the baths that are far away from the channel and the amount of current passed by the channel (in the form of ions) is measured. It is this experimental setup that we consider here.

B. Classical Donnan potentials

In classical membrane transport, the Donnan potential is derived by using the drift-diffusion (Nernst-Planck) equation [1,7–9]

$$J_j = -D_j\left(\frac{dc_j}{dx} + z_j c_j \frac{e}{kT}\frac{d\phi}{dx} + c_j \frac{1}{kT}\frac{d\mu_j^{\mathrm{ex}}}{dx}\right) \tag{1}$$

to describe the flux density J_j for ion species j; ϕ is the local electrostatic potential, D_j, c_j, and z_j are the diffusion coefficient, local concentration, and valence of species j, respectively, and k, T, and e are the Boltzmann constant, absolute temperature, and elementary charge, respectively. μ_j^{ex} is the excess chemical potential of species j, which we take to be a step function that has one constant value in the liquid and another constant value in the membrane. The Nernst-Planck equation can also be written as the derivative of the total chemical potential $a_j \exp[(z_j e/kT)\phi]$, where the activity $a_j = \gamma_j c_j$ with the activity coefficient defined as $\gamma_j = \exp(\mu_j^{\mathrm{ex}}/kT)$. The Nernst-Planck equations for all species is coupled with the Poisson equation to describe the electric field:

$$-\frac{d}{dx}\left(\epsilon\frac{d\phi}{dx}\right) = e\sum_j z_j c_j + e q(x), \tag{2}$$

where q is the charge inherent to the membrane ($q=0$ in the liquid). The boundary conditions for this system are

$$c_j(-\infty) = c_j(L), \quad c_j(\infty) = c_j(R), \tag{3}$$

$$\phi(-\infty) = V_L, \quad \phi(\infty) = V_R, \tag{4}$$

$$V = V_L - V_R. \tag{5}$$

V is the voltage applied to the system and $c_j(L)$ and $c_j(R)$ are the left and right bulk concentration of ion species j. For these equations it is not difficult to prove that both the potential ϕ and the activities a_j are continuous if μ_j^{ex} and $q(x)$ have jump discontinuities.

Assuming that the membrane is represented by the interval $[x_L, x_R]$, the left and right Donnan potentials are

$$\Psi_L = \phi(x_L) - V_L, \quad \Psi_R = \phi(x_R) - V_R. \tag{6}$$

To derive values for these quantities, two approximations are usually made [10,1,7]:

(1) Each species is in equilibrium with the liquid: $J_j = 0$.

(2) Charge neutrality holds everywhere: $0 = \Sigma_j z_j c_j(x) + q(x)$ for all x.

Then the Nernst-Planck equations for each species can be integrated to give

$$c_j(x_L) = c_j(L)\exp(\Delta\mu_j^{\mathrm{ex}})\exp(-z_j\Psi_L), \tag{7}$$

where $\Delta\mu_j^{\mathrm{ex}}$ is the difference in the excess chemical potentials of species j in the liquid and within the membrane. With the charge neutrality condition, the Donnan potential can be shown to be the solution to the polynomial

$$0 = \sum_j z_j c_j(L)\exp(\Delta\mu_j^{\mathrm{ex}}) Y_L^{-z_j} + q_L, \tag{8}$$

where $Y_L = \exp(e\Psi_L/kT)]$ and $q_L = q(x_L)$. Similar formulas hold for the right side of the membrane. With these potentials, the concentrations just inside the membrane follow from Eq. (7).

This approximation for the potentials at the edge of the membrane works well for thick membranes, when the change in the potential in the liquid to that in the membrane can be well approximated by a discontinuity. However, when the membrane is very thin, the part of the membrane where charge neutrality does not hold may be of significant size relative to the width of the membrane, and one of the assumptions for deriving the Donnan potential is violated. One example of such a situation is the modeling of ion transport through biological membranes (specifically through open ion channels) where the membrane is only tens of Angstroms wide [11]. In such a case, the charge-neutrality condition used above does not hold and thus the Donnan potentials do not give a good approximation to the potentials at the edges of the membrane. (This is easily verified with numerical solutions to the Poisson-Nernst-Planck system.)

In this paper we derive new formulas for the potentials and concentrations at the interface of a liquid and a thin membrane. Furthermore, we employ neither of the two assumptions normally used.

II. THEORY

We consider a more general model than Eqs. (1) and (2) which we write in nondimensional units:

$$-\varepsilon^2\left[\frac{d}{dx}\left(\epsilon(x)\frac{d\phi}{dx}(x)\right) + \frac{dA/dx}{A(x)}\epsilon(x)\frac{d\phi}{dx}(x)\right] = \sum_j z_j c_j(x) + q(x) \tag{9}$$

$$-\frac{J_j}{D_j(x)A(x)} = \frac{dc_j}{dx}(x) + z_j c_j(x)\frac{d\phi}{dx}(x) + z_j c_j(x)\frac{d\theta_j}{dx}(x), \tag{10}$$

where

$$\varepsilon^2 = \frac{\epsilon_{\mathrm{scale}} kT}{C_{\mathrm{scale}} e^2 d^2}. \tag{11}$$

First introduced in [12], A is an area function (although not necessarily a cross-sectional area) designed to model the bath/channel interface because the entire membrane no longer conducts ions. (We are only considering a small channel attached to a large electrolyte bath.) As explained in [11], $A(x)$ is the area of the equipotential surface in the full three-dimensional problem and is assumed to be equal to the cross-sectional area inside the channel. An explicit formula for $A(x)$ is not known, but our results do not require such a formula. If $A(x)$ is taken to be constant, the usual Poisson-Nernst-Planck equations are obtained with the exception that J_j is a flux instead of a flux density. The expressions involving $A(x)$ used here are derived in [11]. The excess chemical potentials μ_j^{ex} are rewritten as $\mu_j^{\text{ex}}=z_j\theta_j$. Equations (9) and (10) are nondimensional where the electrostatic potential has been scaled with kT/e, the excess chemical potentials with kT, the concentrations/diffusion coefficients/dielectric with the largest concentrations/diffusion coefficients/dielectric in the system ($C_{\text{scale}}/D_{\text{scale}}/\epsilon_{\text{scale}}$), and the length of the system d (which includes the membrane and enough of the liquid to reach bulk concentrations) has been scaled to 1. For simplicity we assume that the dielectric coefficient is continuous everywhere.

For the rest of the paper we will show the work on the left side of the channel and merely state the results for the right side; the proofs are similar. To denote the liquid we use the subscript 0 and to denote the channel we use the subscript 1.

To approximate the piecewise constant excess chemical potentials, we first consider them as continuous functions that change rapidly at the interface. Later we take the limit that the derivative becomes infinite. In this way delta functions are avoided. Put mathematically, our assumption about the excess chemical potentials is [13,14]:

$$\theta_{j,0}(x)=\frac{\mu_{j,0}^{\text{ex}}}{z_{j,0}}+\Theta_{j,0}\left(\frac{x_L-x}{\varepsilon}\right), \tag{12}$$

$$\theta_{j,1}(x)=\frac{\mu_{j,1}^{\text{ex}}}{z_{j,1}}+\Theta_{j,1}\left(\frac{x_L-x}{\varepsilon}\right), \tag{13}$$

where the membrane runs from x_L to x_R, 0 denotes the bath, 1 denotes the channel.

$$\mu_{j,i}^{\text{ex}}=\text{const} \quad (i=0,1), \tag{14}$$

and

$$\lim_{\zeta\to\infty}\Theta_{j,i}(\zeta)=0 \quad (i=0,1). \tag{15}$$

When we take the limit of the derivative becoming infinite (which is after all the analysis is complete)

$$\Theta_{j,i}(0)\approx\Theta_{j,i}(\infty)=0. \tag{16}$$

We are interested in the values $\phi(x_L)$ and $c_j(x_L)$. To find approximate values we analyze the system of equations with singular perturbation (SP) theory [13,14,11]. SP is a well-proven approximation technique used for problems that have a small parameter and involves breaking up the solution into parts where the function is rapidly changing (the so-called boundary layers) and parts where the function behaves ''nicely'' (the so-called outer solutions). Since we are interested in the areas near the edge of the membrane where both the concentrations and the potential are changing rapidly, we will focus mainly on the boundary layers there.

We start by stating the assumptions we use throughout:

(1) The diffusion coefficient for each ion species in the liquid and the membrane are different but constant in each region.

(2) We require that the concentrations and potential in the baths are constant except in the immediate vicinity of the channel. This is the condition that (usually) excludes the leaky membrane [case (1)] since in that case the concentrations generally change linearly in the baths. In the case of channels, this condition is usually true because the baths are coupled only through a small hole that cannot sustain much flow. Furthermore, the diffusion coefficients of all ions are significantly smaller inside the channel than in bulk solution because of the higher friction produced by geometrical constraints and special chemical and physical conditions inside the narrow channel (for example, waters are oriented and ions diffuse in predominantly one direction). This condition ensures that concentrations do not vary much across the electrolyte solutions surrounding a membrane containing just one channel; this is mathematically proven in [11] and can easily be verified with numerical solutions of the equations.

Next we break the analysis into two intervals: $(0,x_1=x_L)$ for the liquid and (x_1,x_2) for the membrane where x_2 is located somewhere within the membrane. We consider each section separately and use the index i to indicate the segment being considered; $i=0$ is the liquid and $i=1$ is the channel. In each segment we assume each solution has the following form:

$$f_0(x)=f_{0,\text{out}}(x)+F_0\left(\frac{x_L-x}{\varepsilon}\right) \quad (f=\phi,c_j;F=\Phi,C_j), \tag{17}$$

$$f_1(x)=f_{1,\text{out}}(x)+F_1\left(\frac{x\text{-}x_L}{\varepsilon}\right) \quad (f=\phi,c_j;F=\Phi,C_j), \tag{18}$$

where the capital letters are for boundary layers (which we expect at the membrane edge because of the change in charge from $q_0=0$ in the liquid to $q_1=q_L$ in the membrane). Because ε is small (it is related to the Debye length), the arguments of F_i become large very quickly and therefore are good for modeling rapidly changing functions. The boundary layer functions are assumed to go to zero rapidly [see (27)] so they only describe a thin layer. Substituting these into Eqs. (9) and (10) and multiplying out all the sums, for both $i=0,1$ we define the outer solutions to satisfy the original equations but *not the boundary conditions*:

$$\varepsilon^2\left[\frac{d}{dx}\left(\epsilon\frac{d\phi_{i,\text{out}}}{dx}\right)+\frac{dA/dx}{A}\epsilon\frac{d\phi_{i,\text{out}}}{dx}\right]=\sum_j z_j c_{j,i,\text{out}}+q_i \tag{19}$$

$$-\frac{J_j}{D_{j,i}A}=\frac{dc_{j,i,\text{out}}}{dx}+z_jc_{j,i,\text{out}}\frac{d\phi_{i,\text{out}}}{dx}. \tag{20}$$

(Note that the excess chemical potential terms for the outer solutions drop out because they are assumed to be constant inside the channel except at edges of the channel; thus they will be included in the boundary layer equations.) The boundary layers satisfy the remaining terms of this multiplication:

$$-\left[\frac{d}{d\zeta}\left(\hat{\epsilon}_i\frac{d\Phi_i}{d\zeta}\right)+\hat{\epsilon}_i\frac{d\hat{A}/d\zeta}{\hat{A}}\frac{d\Phi_i}{d\zeta}\right]=\sum_j z_jC_{j,i}, \tag{21}$$

$$0=\frac{dC_{j,i}}{d\zeta}+z_jC_{j,i}\frac{d\hat{\phi}_i}{d\zeta}+z_jC_{j,i}\left(\frac{d\Phi_i}{d\zeta}+\frac{d\Theta_{j,i}}{d\zeta}\right)+z_j\hat{c}_{j,i}\left(\frac{d\Phi_i}{d\zeta}+\frac{d\Theta_{j,i}}{d\zeta}\right), \tag{22}$$

where $\zeta=(-1)^{i+1}(x-x_L)/\varepsilon$ and

$$\hat{f}_i(\zeta)=f_i(x_L+(-1)^{i+1}\varepsilon\zeta)\quad(f=\epsilon,\phi,c_j). \tag{23}$$

For these functions, $d\hat{f}_i/d\zeta=O(\varepsilon)$ where we use the Big-*O* order notation [14].

Next, all of the functions are expanded in powers of ε:

$$f_{\text{out}}(x)=f^{(0)}(x)+\varepsilon f^{(1)}(x)+\cdots\quad(f=\phi_i,c_{j,i}), \tag{24}$$

$$F(\zeta)=F^{(0)}(\zeta)+\varepsilon F^{(1)}(\zeta)+\cdots\quad(F=\Phi_i,C_{j,i}), \tag{25}$$

$$J_j=J_j^{(0)}+\varepsilon_j^{(1)}+\cdots. \tag{26}$$

These functions will be solved to satisfy

$$\lim_{\zeta\to\infty}F^{(k)}(\zeta)=\lim_{\zeta\to\infty}(dF^{(k)}/d\zeta)(\zeta)=0\quad(F=\Phi_i,C_{j,i};k\geqslant 0) \tag{27}$$

as described before Eq. (19). The outer solutions and boundary layers together will need to satisfy the following boundary conditions on the intervals $(0,x_L)$ and (x_L,x_2):

$$\phi_0^{(0)}(x_L)+\Phi_0^{(0)}(0)=\phi(x_L)\equiv\beta_\phi, \tag{28a}$$

$$\phi_0^{(k)}(x_L)+\Phi_0^{(k)}(0)=0\quad(k\geqslant 1), \tag{28b}$$

$$c_{j,0}^{(0)}(x_L)+C_{j,0}^{(0)}(0)=\lim_{x\searrow x_L}c_{j,0}(x)\equiv\beta_{j,0}, \tag{28c}$$

$$c_{j,0}^{(k)}(x_L)+C_{j,0}^{(k)}(0)=0\quad(k\geqslant 1), \tag{28d}$$

$$\phi_1^{(0)}(x_L)+\Phi_1^{(0)}(0)=\phi(x_L)\equiv\beta_\phi, \tag{28e}$$

$$\phi_1^{(k)}(x_L)+\Phi_1^{(k)}(0)=0\quad(k\geqslant 1), \tag{28f}$$

$$c_{j,1}^{(0)}(x_L)+C_{j,1}^{(0)}(0)=\lim_{x\searrow x_L}c_{j,1}(x)\equiv\beta_{j,1} \tag{28g}$$

$$c_{j,1}^{(k)}(x_L)=C_{j,1}^{(k)}(0)=0\quad(k\geqslant 1). \tag{28h}$$

In this way the leading-order terms on each segment take the values of the unapproximated functions at the point x_L while the lower-order terms contribute nothing to the series at x_L. We make a distinction between the limit with $x<x_L$ $[\lim_{x\nearrow x_L}c_{j,0}(x)]$ and the limit with $x<x_L$ $[\lim_{x\searrow x_L}c_{j,1}(x)]$ because if the excess chemical potential is discontinuous, so is the concentration (although the activity is continuous) and so we must consider the limits in each interval separately.

The goal of the paper is to find approximations for β_ϕ and $\beta_{j,1}$, the values of potential and concentration, respectively, just inside the channel.

A. Outer solutions

Substituting Eqs. (24) and (26) into Eqs. (19) and (20) at leading orders gives

$$0=\sum_j z_jc_{j,i}^{(0)}+q_i, \tag{29}$$

$$-\frac{J_j^{(0)}}{D_{j,i}A}=\frac{dc_{j,i}^{(0)}}{dx}+z_jc_{j,1}^{(0)}\frac{d\phi_i^{(0)}}{dx}. \tag{30}$$

Since we are only interested in the transition regions (that is, the boundary layers) we do not explore the solutions to these equations. The following results depend only on the charge neutrality result (29) of the leading-order terms of the concentrations. Note that Eq. (29) is not a physical assumption like in the usual Donnan potential derivation, but a mathematical construction. (The outer solutions and boundary layers are only mathematical functions; they do not physically exist, but, as the solution of the transport equations, their *sum* does.)

The assumption about the bath concentrations stated above can now be put mathematically:

$$c_{j,0}^{(0)}(x_L)\approx c_{j,0}^{(0)}(0)=c_j(L), \tag{31}$$

$$c_{j,1}^{(0)}(x_R)\approx c_{j,1}^{(0)}(1)=c_j(R), \tag{32}$$

$$\phi_0^{(0)}(x_L)\approx\phi_0^{(0)}(0)=V_L, \tag{33}$$

$$\phi_1^{(0)}(x_R)\approx\phi_1^{(0)}(1)=V_R. \tag{34}$$

B. Boundary layers

As with the outer solutions, we get a hierarchy of equations for the boundary layers after substituting Eq. (25) into Eqs. (21) and (22), multiplying the series, and equating like powers of ε. At leading order the equations are

$$-\epsilon(x_L)\frac{d^2\Phi_i^{(0)}}{d\zeta^2}=\sum_j z_jC_{j,i}^{(0)}, \tag{35}$$

$$0=\frac{dC_{j,i}^{(0)}}{d\zeta}+z_jC_{j,i}^{(0)}\left(\frac{d\Phi_i^{(0)}}{d\zeta}+\frac{d\Theta_{j,i}}{d\zeta}\right)$$
$$+z_jc_{j,i}^{(0)}(x_L)\left(\frac{d\Phi_i^{(0)}}{d\zeta}+\frac{d\Theta_{j,i}}{d\zeta}\right) \quad (36)$$

This last equation is a linear, first-order ordinary differential equation for $C_{j,i}^{(0)}$ that is solved by

$$C_{j,i}^{(0)}=c_{j,i}^{(0)}(x_L)\{\exp[-z_j(\Phi_i^{(0)}+\Theta_{j,i})]-1\} \quad (37)$$

where the constant of integration was found using Eq. (27). Evaluating this at $\zeta=0$ gives

$$\beta_{j,i}=c_{j,i}^{(0)}(x_L)\exp[-z_j(\beta_\phi-\phi_i^{(0)}(x_L))] \quad (38)$$

where we have used Eqs. (28) and (16).

III. RESULTS

Theorem 1. *The outer solutions just inside the membrane interface* $\phi_1^{(0)}(x_L)$ *and* $c_{j,1}^{(0)}(x_L)$ *are the usual Donnan potential and concentrations*:

$$\phi_1^{(0)}(x_L)=V_L+\ln(Y), \quad (39)$$

$$c_{j,1}^{(0)}(x_L)=c_j(L)\rangle\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})Y^{-z_j}, \quad (40)$$

where Y satisfies

$$0=\sum_j z_jc_j(L)\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})Y^{-z_j}+q_L. \quad (41)$$

Note that Eqs. (40) *and* (41) *are the same as* Eqs. (7) *and* (8).

Proof. Let

$$A_{j,i}=c_{j,i}^{(0)}(x_L)\exp(\mu_{j,i}^{\rm ex}). \quad (42)$$

Since the activities are continuous, we have

$$\beta_{j,0}\exp(\mu_{j,0}^{\rm ex})=\beta_{j,1}\exp(\mu_{j,1}^{\rm ex})\equiv\alpha_j. \quad (43)$$

Using Eq. (38) we get

$$A_{j,0}^{z_k}A_{k,0}^{-z_j}=\alpha_j^{z_k}\alpha_k^{-z_j}=A_{j,1}^{z_k}A_{k,1}^{-z_j} \quad (44)$$

and thus

$$\left(\frac{A_{j,0}}{A_{j,1}}\right)^{1/z_k}=\left(\frac{A_{k,0}}{A_{k,1}}\right)^{1/z_j} \quad (45)$$

for any j and k. Dividing Eq. (38) for the left and right sides of x_L, we get

$$\frac{\alpha_k}{\alpha_j}=\frac{A_{k,0}}{A_{j,0}}\exp[(-z_k+z_j)\{\beta_\phi-\phi_0^{(0)}(x_L)\}] \quad (46)$$

$$=\frac{A_{k,1}}{A_{j,1}}\exp[(-z_k+z_j)\{\beta_\phi-\phi_1^{(0)}(x_L)\}]. \quad (47)$$

Thus

$$\exp[\phi_1^{(0)}(x_L)-\phi_0^{(0)}(x_L)]=\left(\frac{A_{j,0}}{A_{j,1}}\frac{A_{k,1}}{A_{k,0}}\right)^{1/(-z_k+z_j)}. \quad (48)$$

Equation (45) gives

$$\left(\frac{A_{j,0}}{A_{j,1}}\right)^{1/z_j}=\left(\frac{A_{1,0}}{A_{1,1}}\right)^{1/z_1}\equiv Y \quad (49)$$

for all j. Solving for $A_{j,1}$ we get

$$A_{j,1}=a_{j,0}Y^{-z_j}. \quad (50)$$

Putting this back in terms of concentrations, we get

$$c_{j,1}^{(0)}(x_L)=c_{j,0}^{(0)}(x_L)\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})Y^{-z_j}. \quad (51)$$

Next we sum Eq. (51) over j and apply the electroneutrality of outer solutions (29)

$$0=\sum_j z_jc_{j,1}^{(0)}(x_L)+q_L \quad (52)$$

$$=\sum_j z_jc_{j,0}^{(0)}(x_L)\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})Y^{-z_j}+q_L. \quad (53)$$

Therefore the Y needed to find $c_{j,1}^{(0)}(x_L)$ is the solution to this polynomial. Then Eq. (53) is the same as Eq. (7) with $Y=Y_L$ after using approximation (31). Lastly, by substituting Eq. (45) into Eq. (48) and using Eq. (33) we have

$$\phi_1^{(0)}(x_L)=\phi^{(0)}(x_L)+\ln(Y)\approx V_L+\ln(Y) \quad (54)$$

and the last result follows. ■

This result is not surprising; this part of the singular perturbation expansion is a more mathematical way to do the derivation of the Donnan potential. Note, however, that we did not use the assumptions normally used to derive the Donnan potential.

Next we concentrate on the corrections to the Donnan potential and concentrations that are new results. By Eq. (38) and Theorem 1 we already have

$$\beta_{j,1}=c_j(L)\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})\exp[-z_j(\beta_\phi-V_L)] \quad (55)$$

so the only thing missing is a formula for β_ϕ that is given in the next result.

Theorem 2. β_ϕ *is given by the implicit formulation*

$$q_L(\beta_\phi-V_L)+\sum_j c_j(L)[1-\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})]$$
$$\times\exp[-z_j(\beta_\phi-V_L)]$$
$$=q_L\Psi_L+\sum_j c_j(L)[1-\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})\exp(-z_j\Psi_L)], \quad (56)$$

where $\Psi_L=\phi_0^{(0)}(x_L)-V_L\approx\ln(Y)$ *is the usual, but nondimensionalized, Donnan potential on the left side of the membrane. When there is no difference in the excess chemical potentials between the liquid and the membrane for all species* (*that is,* $\mu_{j,0}=\mu_{j,1}$ *for all j*), *then this formula is explicit*:

$$\beta_\phi-V_L=\Psi_L+\Psi_L', \tag{57}$$

where

$$\Psi_L'=\frac{1}{q_L}\sum_j c_j(L)[1-\exp(-z_j\Psi_L)] \tag{58}$$

is the correction to the usual Donnan potential Ψ_L. *In this case*,

$$\beta_{j,0}=\beta_{j,1}=c_j(L)\exp[-z_j(\Psi_L+\Psi_L')]. \tag{59}$$

Proof. Multiplying Eq. (35) by $d\Phi_i^{(0)}/d\zeta$, substituting in Eq. (37), integrating from 0 to ∞, and using Eq. (27) gives

$$\begin{aligned}\frac{\epsilon(x_L)}{2}&\left(\frac{d\Phi_i^{(0)}}{d\zeta}(0)\right)^2\\ &=\sum_j z_j c_{j,i}^{(0)}(x_L)\int_0^\infty \frac{d\Phi_i^{(0)}}{d\zeta}\exp[-z_j(\Phi_i^{(0)}+\Theta_{j,i})]\\ &\quad\times d\zeta-q_i\Phi_i^{(0)}(0).\end{aligned} \tag{60}$$

It is at this stage that we need the fact that the excess chemical potentials we are approximating are step functions. In that case the excess chemical potentials of all species change much more rapidly than the electrostatic potential and we may approximate the integrals above by

$$\begin{aligned}&\int_0^\infty \frac{d\Phi_i^{(0)}}{d\zeta}\exp[-z_j(\Phi_i^{(0)}+\Theta_{j,i})]d\zeta\\ &\quad\approx\exp[-z_j\Theta_{j,i}(\infty)]\int_0^\infty \frac{d\Phi_i^{(0)}}{d\zeta}\exp(-z_j\Phi_i^{(0)})d\zeta\\ &\quad=\frac{1}{z_j}[\exp\{-z_j\Phi_i^{(0)}(0)\}-1].\end{aligned} \tag{61}$$

This expression is exact if there is no difference in the excess chemical potentials in the liquid and the membrane; that is, if the activity coefficients for permeating ions are the same in the bulk solution and the channel. Setting the $[(d\Phi_0^{(0)}/d\zeta)(0)]^2$ equal to $[(d\Phi_1^{(0)}/d\zeta)(0)]^2$ because ϕ must have a continuous first derivative gives

$$\begin{aligned}&\sum_j c_j(L)\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})\exp[-z_j(\beta_\phi-V_L)]\\ &\quad-\sum_j c_j(L)\exp(\mu_{j,0}^{\rm ex}-\mu_{j,1}^{\rm ex})Y^{-z_j}-q_L[\beta_\phi-V_L-\ln(Y)]\\ &\quad=\sum_j c_j(L)\exp[-z_j(\beta_\phi-V_L)]-\sum_j c_j(L).\end{aligned} \tag{62}$$

The result follows since $\Psi_L=\ln(Y)$ by Theorem 1. ■

Lastly, we describe the amount of space charge that is contained in the boundary layers; that is, how far the areas just inside and just outside of the channel are away from charge neutrality.

Theorem 3. *The amount of space charge in the bath is*

$$\begin{aligned}&-\mathrm{sgn}(q_L)\varepsilon A(x_L)\sqrt{2\epsilon(x_L)}\\ &\quad\times\left[\sum_j c_j(L)\{\exp[-z_j(\beta_\phi-V_L)]-1\}\right]^{1/2}.\end{aligned} \tag{63}$$

Overall the system is charge neutral so that the space charge just inside the channel is equal in size and opposite in sign to that in the bath.

Proof. The amount of charge contained in both boundary layers is given by

$$\begin{aligned}&\int_0^{x_2}A(x)\sum_j\{z_jc_j(x)+q(x)\}dx\\ &\quad\approx\int_0^{x_L}A(x)\sum_j z_j\{c_{j,0}^{(0)}(x)+C_{j,0}^{(0)}(\zeta)\}dx\\ &\qquad+\int_{x_L}^{x_2}A(x)\sum_j[z_j\{c_{j,1}^{(0)}(x)+C_{j,1}^{(0)}(\zeta)\}+q_L]dx.\end{aligned} \tag{64}$$

The first integral on the right-hand side is the charge contained in the boundary layer in the bath just outside the channel and the second is the charge just inside the channel. Since the outer solutions are charge neutral by Eq. (29), this leaves

$$\begin{aligned}&\int_0^{x_2}A(x)\sum_j\{z_jc_j(x)+q(x)\}dx\\ &\quad\approx\int_0^{x_L}A(x)\sum_j z_jC_{j,0}^{(0)}(\zeta)dx\\ &\qquad+\int_{x_L}^{x_2}A(x)\sum_j\{z_jc_{j,1}^{(0)}(\zeta)\}dx\end{aligned} \tag{65}$$

$$\begin{aligned}&\approx\varepsilon A(x_L)\left(\int_0^\infty\sum_j z_jC_{j,0}^{(0)}(\zeta)d\zeta\right.\\ &\qquad\left.+\int_0^\infty\sum_j z_jC_{j,1}^{(0)}(\zeta)d\zeta\right),\end{aligned} \tag{66}$$

where we have used the approximation that the boundary layers change more rapidly than the area function A, an assumption that we have already made in order to derive Eq. (35). $A(x_L)$ is then the area of the left side of the channel. By the Poisson equation for the boundary layers (35),

$$\int_0^\infty \sum_j z_j C_{j,i}^{(0)} d\zeta = -\epsilon(x_L) \int_0^\infty \frac{d^2\Phi_i^{(0)}}{d\zeta^2} d\zeta = \epsilon(x_L) \frac{d\Phi_i^{(0)}}{d\zeta}(0). \tag{67}$$

Since $d\Phi/dx$ is continuous and by the two different definitions of ζ for the bath and the channel given after Eq. (22), we have $(d\Phi_0^{(0)}/d\zeta)(0) = -(d\Phi_1^{(0)}/d\zeta)(0)$. Therefore, since ϵ is assumed to be continuous, the total charge in both layers is 0 (that is, overall the system is charge neutral) and in order to find the amount of charge separated it suffices to find $(d\Phi_0^{(0)}/d\zeta)(0)$. In Eqs. (60) and (61) we found $[(d\Phi_0^{(0)}/d\zeta)(0)]^2$ and so only the sign remains to be determined. Because we are considering the uncompensated charge in the bath, it must be the counter ions going into the channel. Thus the sign must be opposite to that of the fixed charge inside the channel q_L. ■

IV. DISCUSSION

We have derived a formula for the electrostatic potential at the edge of a channel (that is, at the interface between the channel and the surrounding ionic solutions) based on the Poisson-Nernst-Planck model of charge transport. In fully dimensional units this potential β_ϕ is given by

$$\begin{aligned} q(\beta_\phi - V) + \frac{kT}{e} \sum_j c_{j,\mathrm{liq}} \left[1 - \exp\left(\frac{\Delta\mu_j^{\mathrm{ex}}}{kT} \right) \right] \\ \times \exp\left[-\frac{z_j e}{kT} (\beta_\phi - V) \right] \\ = q\Psi + \frac{kT}{\epsilon} \sum_j c_{j,\mathrm{liq}} \left[1 - \exp\left(\frac{\Delta\mu_j^{\mathrm{ex}}}{kT} \right) \exp\left(-\frac{z_j e}{kT} \Psi \right) \right], \end{aligned} \tag{68}$$

where Ψ is the usual Donnan potential on the side of the membrane being studied,

$$\Delta\mu_j = \mu_{j,\mathrm{liq}} - \mu_{j,\mathrm{mem}} \tag{69}$$

is the difference in excess chemical potential of species j between the bulk liquid and within the membrane, $c_{j,\mathrm{liq}}$ is the bulk liquid concentration of species j, and q and V are, respectively, the membrane charge concentration and the voltage applied on the side of the membrane being studied. The concentrations $\beta_{j,\mathrm{mem}}$ just inside the membrane are given by

$$\beta_{j,\mathrm{mem}} = c_{j,\mathrm{liq}} \exp\left(\frac{\Delta\mu_j^{\mathrm{ex}}}{kT} \right) \exp\left\{ -\frac{z_j e}{kT} (\beta_\phi - V) \right\}. \tag{70}$$

In the case when $\Delta\mu_j^{\mathrm{ex}} = 0$ for all species j we have derived a simple correction to the usual Donnan potential Ψ:

$$\beta_\phi - V = \Psi + \Psi', \tag{71}$$

where

$$\Psi' = \frac{kT}{e} \sum_j \frac{c_{j,\mathrm{liq}}}{q} \left[1 - \exp\left(-\frac{z_j e}{kT} \Psi \right) \right]. \tag{72}$$

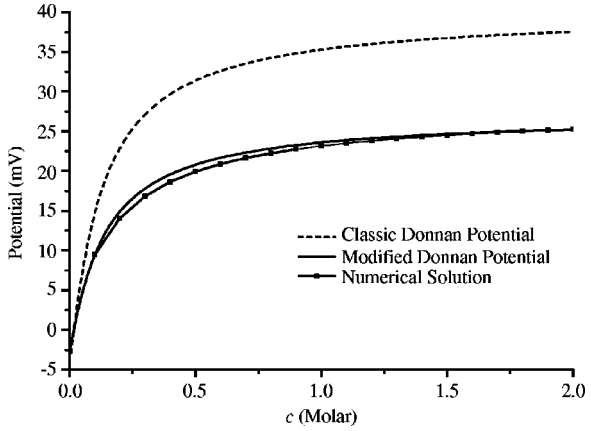

FIG. 1. Plot of the potential (in millivolt) at the liquid/membrane interface as function of bath concentration c (in molar) with $q_1 = -1.0$ M, $z_2 = 2$, $c_2(L) = 0.1$ M, $c_2(R) = 0.1$ M, $\Delta\mu_2 = 37$ mV, $D_{2,\mathrm{bath}} = 3 \times 10^{-9}$ cm^2/sec, $D_{2,\mathrm{ch}} = 1.5 \times 10^{-11}$ cm^2/sec, $z_3 = 1$, $c_3(L) = c$, $c_3(R) = 0.1$ M, $\Delta\mu_3 = 80$ mV, $D_{3,\mathrm{bath}} = 2 \times 10^{-9}$ cm^2/sec, $D_{3,\mathrm{ch}} = 2 \times 10^{-12}$ cm^2/sec, $z_1 = -1$, $c_1(L) = c + 0.2$ M, $c_2(R) = 0.3$ M, $\Delta\mu_1 = 0$ mV, $D_{2,\mathrm{bath}} = 10^{-9}$ cm^2/sec, $D_{2,\mathrm{ch}} = 10^{-11}$ cm^2/sec. The channel was given a radius of 3.5 Å and A was given a constant value of the channel's cross-sectional area. The diffusion coefficients are those used in the numerical solutions, but are not necessary for the theoretical treatment.

The amount of space charge in the bath has magnitude

$$A\left(2kT\epsilon \sum_j c_{j,\mathrm{liq}} \left[\exp\left\{ -\frac{z_j e}{kT} (\beta_\phi - V) \right\} - 1 \right] \right)^{1/2} \tag{73}$$

and has the sign opposite of that of q. Inside the channel, the space charge has the same magnitude, but has the sign the same as that of q. (A and ϵ are the cross-sectional area and dielectric coefficient, respectively, at the liquid/membrane interface.) The results of this analysis for a case of three ion species are shown in Fig. 1.

These modifications to the Donnan potential are valid when the channel is long enough to have the inner part of the channel charge-neutral; that is, the mobile and permanent charge are equal in that region. If there is no such part of the channel, then there are no distinct boundary layers on the edge of the channel and outer solution in the middle and the mathematical assumptions about the structures of the solutions (17), (18), and (27) are no longer true. For biological ion channels, this generally means a channel of length 20 Å or more. For shorter channels, however, the edge potentials given here should still be better than classical Donnan potentials that have been used in the past [15].

A surprising result of our analysis is its confirmation of the validity of the usual simple treatment of the Donnan potential. When our assumptions are satisfied, the usual treatment gives the correct current/voltage relation even though its internal images of the potential and concentration profiles at the membrane edges have serious errors. This is because the classical Donnan potential is the extrapolation of the potential profile across the channel from the charge-neutral center to the channel edge; that is, it ignores the boundary layers. The inclusion in the calculations of the

boundary layers does not change the potential or concentration profiles in the middle of the channel, but only at the edges (assuming the channel has a change-neutral region in the middle). Since the current in this one-dimensional model must be constant, the current calculated in the middle of the channel (away from the boundary layers) is the same regardless of whether the boundary layers are included or not. When dealing with ultrashort channels (<20 Å) this analysis is no longer true; using the usual Donnan potentials and the newly derived potentials would give different current/voltage relations because the center of the channel is never charge neutral. In this case while the newly derived potentials are not exactly correct since the underlying mathematical assumptions are not true, their use must give more accurate results than the application of the usual Donnan potentials since for such channels the new boundary conditions will recover the potential and concentration profiles better. For all types of membranes, the modifications become important when one is interested in the actual shapes of the potential and concentrations across the length of the channel.

Last, in the case of leaky membranes (case 1), the results of this study are still valid when one of the following two conditions is satisfied.

(1) The experiment is set up so that the bath concentrations are known and do not vary significantly with distance from the channel. Experimental precautions are usually taken to ensure this situation for K^+, Na^+, and Cl^- channels. Such a simplification is not possible for Ca^{2+} channels because they normally operate with negligible Ca^{2+} concentration (<1 μM [3]) on one side.

(2) The bath concentrations near the membrane are known. In that case these concentrations should be put into the equations for the modifications of the Donnan potentials and concentrations (68) and (70).

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