2

FREE DIFFUSION

INTRODUCTION

In this chapter, we consider the simplest of transport processes: the passive diffusion of a solute that occurs when its electrochemical potentials on the two sides of a permeable barrier are different. Indeed, this process is so simple that it fails to represent many aspects of transport in living systems. Nonetheless, it does describe some aspects of biological transport quite well, and it also provides a "base case" whose behavior can be compared against that of more complex transport mechanisms.

This chapter is divided into two sections; the first deals with free diffusion of nonelectrolytes, and the second with that of ionic species. The principal property variable determining the flux of a nonelectrolyte is its permeability, a quantity that can in principle be related to the diffusion coefficient of the solute. Electrolyte diffusion in free solution is most rigorously described by classical electrodiffusion theory. The flux equations provided by this theory are very complex, and they have not seen nearly as much use as have approximations to them. Accordingly, emphasis will be placed here on the principles of electrodiffusion, and on the approximate solutions and special cases that are most commonly used.

The equations of free diffusion can describe a wide variety of transport phenomena, including steady and unsteady transport processes; processes that can be described in one, two or three dimensions and in a variety of geometries; and processes in which chemical reactions and fluid flow take place simultaneously with diffusion. In this chapter, we will discuss a small subset of these, focusing on the tools that are applied to living systems. Comprehensive discussions of diffusional processes can be found in other texts, such as Crank's (1975) classic text, published thirty years ago and still being reprinted! A more limited set of solutions, but with more consistent biological applicability, can be found in Truskey et al. (2004).

M.H. Friedman, *Principles and Models of Biological Transport*,
DOI: 10.1007/978-0-387-79240-8_2,

2.1. FREE DIFFUSION OF NONELECTROLYTES

The first transport process we will consider is the diffusion of a dissolved nonelectrolyte across a membrane or a similar barrier. The diffusive process is driven by the solute's concentration gradient. For now, the nature of the solvent is not particularly important, and it will usually be understood to be water, which is the most common biological solvent. As will be seen in Chapter 7, much of the material developed below is equally applicable to diffusion through a lipid film, such as the hydrophobic region of a cell membrane.

In free diffusion through a membrane, the solute particles move about by random Brownian motion, like that in free solution. The solute flux, which is a measurable and reproducible quantity, is essentially the resultant of these separate motions. Even though the path of a single solute particle cannot be predicted, the consequence of an enormous number of these paths is quite reproducible.

2.1.1. The Teorell Equation

The flux in free diffusion can be written very simply, in a form proposed by Teorell (1953):

$$\text{Flux} = \text{Mobility} \times \text{Concentration} \times \text{Driving force}. \tag{2.1}$$

In the most commonly used units, the flux is the number of mols of solute crossing one square centimeter of membrane per second; it is proportional to the product of the solute mobility, which measures its ease of transport and depends jointly on the barrier/solvent and the solute, as well as the temperature; the solute concentration, which measures the amount of material available to participate in the process; and the driving force for the diffusion of the solute.

The choice of a proper driving force is dictated by thermodynamic considerations that we will not examine until Chapter 6; for now, we will rationalize that choice by analogy with electrical phenomena. First, we recall that, when the chemical potential of the solute is the same in the two phases bounding the membrane, the solute is in equilibrium, and its flux across the membrane is zero. An analogous situation occurs in electrical circuits; when there is no electrical potential difference, there is no current flow. When the electrical potentials at two points are different, the potential gradient defines a field, and charged particles move in response to it. The force acting on the charges is the negative of the *electrical potential gradient*. The analogous driving force for solute flux is the negative of the *chemical potential gradient*:

$$\text{Driving force} = -\nabla\mu_s. \tag{2.2}$$

Almost every transport process with which we will be concerned can be described in terms of a single spatial coordinate perpendicular to the plane of the barrier. Calling that the x-direction, the driving force becomes:

$$\text{Driving force} = -\frac{d\mu_s}{dx}. \tag{2.3}$$

The implicit assumption in this *one-dimensional* treatment of gradients and fluxes is that these vectors are oriented perpendicular to the membrane plane and have negligible components parallel to that plane. This is reasonable if the extent of the membrane is much larger than its thickness, as is usually the case. The Teorell equation can now be written:

$$J_s = U_s c_s \left(-\frac{d\mu_s}{dx}\right), \tag{2.4}$$

where U_s and c_s are the solute mobility and concentration, respectively. The flux J_s is positive in the direction of increasing x.

An integral driving force can also be defined, by integrating Eq. (2.3) across the membrane:

$$\text{Integral driving force} = \int_0^a -\frac{d\mu_s}{dx}dx = \mu_s^{\mathrm{I}} - \mu_s^{\mathrm{II}}. \tag{2.5}$$

In Eq. (2.5), a is the thickness of the membrane. Phase I bathes the face of the membrane at $x = 0$, and Phase II the face at $x = a$. From Chapter 1, the integral driving force is zero at equilibrium.

The integral driving force would appear to be far more convenient than the differential driving force [given by Eq. (2.3)] for describing transport, because it is based on the chemical potentials in the two phases external to the membrane. Chemical potentials *inside* the membrane, which must be known to find the local differential driving force, are generally unmeasurable. Fortunately, with a few reasonable assumptions, Eq. (2.4) can be integrated to give an expression that relates the transmembrane flux to the conditions in the ambient solutions. This we now do.

2.1.2. Integration of the Teorell Equation; Fick's First Law; Solute Permeability

In integrating Eq. (2.4), the temperature is assumed to be uniform and the effect of ressure on the chemical potential of the solute is neglected; these are quite reasonable assumptions for the systems with which we will be dealing. If, in addition, the solutions are assumed to be ideal, then the chemical potential can be written very simply as

$$\mu_s = \text{constant} + RT \ln c_s. \tag{2.6}$$

Differentiating with respect to x,

$$\frac{d\mu_s}{dx} = RT\frac{d\ln c_s}{dx} = RT\left(\frac{1}{c_s}\frac{dc_s}{dx}\right). \tag{2.7}$$

Substituting Eq. (2.7) into (2.4),

$$J_s = -U_s RT \frac{dc_s}{dx}. \tag{2.8}$$

The solute *diffusion coefficient* D_s is related to the solute mobility through the *Nernst–Einstein* relation, $D_s = U_s RT$. D_s is often referred to as the *binary* diffusion coefficient (denoted D_{ij}), as a reminder that its value depends on the identities of both solute and solvent. Introducing the diffusion coefficient into Eq. (2.8), we obtain:

$$J_s = -D_s \frac{dc_s}{dx}. \tag{2.9}$$

Equation (2.9) is known as *Fick's first law of diffusion.* Note that the flux is positive if the concentration gradient is negative.

In the preceding derivation, it was assumed that Eq. (2.6) holds within the membrane, as though transport proceeded through aqueous pores in which the dependence of chemical potential on solution properties was identical to that in the aqueous solutions at the membrane faces. This is the first of several derivations in which the expressions for chemical or electrochemical potential in free solution will be used to describe the thermodynamic state of solute or solvent inside a transport barrier. The state of solutes and solvent inside a complex, heterogeneous biological barrier is not so neatly defined. Accordingly, it is convenient to think of $c_s(x)$ [and $\psi(x)$ when describing electrolyte transport] as the concentration (and potential) of a free aqueous solution in equilibrium with a thin membrane slice at x. The concentration and potential of this equilibrium solution can be quite different from that of the true solution phase at that point in the membrane; however, since the two phases are defined to be in equilibrium, the chemical potentials of the solute and solvent are the same in each.

A notable difference between the concentration of such an equilibrium aqueous solution and the true intramembrane solute concentration arises when the solubility of the solute in the membrane is different from that in the ambient aqueous phases. Such is the case for diffusion through the lipid bilayer of the cell membrane. The relationship between the solute concentration in the lipid and in an equilibrium aqueous solution is expressed in terms of the *partition coefficient* of the solute between the two phases. Diffusion through lipid layers will be described in Chapter 7.

Fick's first law assumes a somewhat more complicated form when the solutions are nonideal. In that case, the solute chemical potential must be written in terms of activity. The activity, in turn, is the product of the concentration and the activity coefficient. Thus, Eq. (2.7) is replaced by:

$$\frac{d\mu_s}{dx} = RT \frac{d \ln a_s}{dx} = RT \left(\frac{d \ln c_s}{dx} + \frac{d \ln \gamma_s}{dx} \right). \tag{2.10}$$

For nonelectrolytes, the activity coefficient of the solute can be assumed to depend on only c_s, which in turn is a function of x. Thus, the following substitution can be made:

$$\frac{d \ln \gamma_s}{dx} = \frac{d \ln \gamma_s}{d \ln c_s} \cdot \frac{d \ln c_s}{dx}. \tag{2.11}$$

Substituting Eq. (2.11) into (2.10):

$$\frac{d\mu_s}{dx} = RT\left(\frac{1}{c_s}\frac{dc_s}{dx}\right)\left(1+\frac{d\ln\gamma_s}{d\ln c_s}\right).$$

The Teorell equation then becomes

$$J_s = -U_s RT\left(1+\frac{d\ln\gamma_s}{d\ln c_s}\right)\frac{dc_s}{dx}. \tag{2.13}$$

Define an augmented diffusion coefficient D_s^* by

$$D_s^* = D_s\left(1+\frac{d\ln\gamma_s}{d\ln c_s}\right). \tag{2.14}$$

For an ideal solution, $D_s^* = D_s$. By substituting Eq. (2.14) into (2.13), a flux equation is obtained that looks almost identical to Eq. (2.9), and can be regarded as a generalization of Fick's first law to nonideal solutions:

$$J_s = -D_s^*\frac{dc_s}{dx}. \tag{2.15}$$

Fick's first law, as generalized above, is now integrated across the membrane to yield an expression for flux in terms of the transmembrane concentration difference. To set up the integration, Eq. (2.15) is rewritten as

$$J_s dx = -D_s^* dc_s. \tag{2.16}$$

In the steady state, the solute flux is independent of x. Assume that the same is true of D_s^*; Eq. (2.16) can then be integrated across the membrane and solved for J_s:

$$J_s = -\frac{D_s^*(c_s^{II} - c_s^{I})}{a}. \tag{2.17}$$

The flux in Eq. (2.17) is based on a unit area of membrane, so it can be continuous at the interfaces $x = 0$ and $x = a$ only if the entire cross-section of the barrier is available for transport. Furthermore, the assumption that the expressions for chemical potential as a function of concentration are the same in both barrier and bath implies that the solute diffuses through the same solvent as that in the ambient phases. The only barrier for which these assumptions hold would be a thin stagnant water film somehow maintained between two well-stirred aqueous baths. The *solute permeability* of such a thin film is defined as the solute flux per unit concentration difference:

$$k_s^0 = \frac{J_s^0}{c_s^{I} - c_s^{II}} = \frac{J_s^0}{\Delta c_s} = \frac{D_s^*}{a}, \tag{2.18}$$

where we have used the superscript "0" to indicate that diffusion takes place through a thin aqueous film.

The *form* of Eq. (2.18) has been adopted to describe solute transport in biological systems. For such systems, the barrier is not a thin aqueous film, and the permeability is not given by D_s^*/a. The solute permeability of a biological barrier is in general an *experimental* property, obtained by dividing the measured flux of a solute by its transmembrane concentration difference:

$$k_s = \frac{J_s'}{\Delta c_s}, \tag{2.19}$$

where J_s' is the measured flux. Radiolabeled tracers are often used to measure permeability; the numerator and denominator of the right-hand side of Eq. (2.19) are replaced by the tracer flux and the transmembrane difference in tracer activity. Even when the solute does not cross the membrane by free diffusion, the experimental permeability is descriptive of the transport behavior of the system. Such empirical permeabilities, though not always easy to interpret in physical terms, are nonetheless useful for comparing solute transport rates and for predicting fluxes under similar conditions.

There are some cases in which permeability can be estimated from a diffusion coefficient and membrane thickness. If the barrier is a stabilized thin film of a solvent immiscible with water, the permeability of the solute is determined by the partition coefficient, the film thickness and the binary diffusion coefficient of the solute in the solvent that comprises the membrane [see the second paragraph of the note following Eq. (2.9), and Chap. 7]. If the membrane possesses large interstices or pores such that diffusion through them is the same as that in free solution, the permeability is given by $\varphi D_s^*/a$, where φ is the void fraction in the membrane.

Our inability to predict membrane permeability *a priori* reflects our ignorance of many factors that influence the transport of a given solute through a given membrane. Some of these factors, particularly applicable to transport through water-filled passages, are itemized below:

— The void fraction mentioned above, or the fraction of the presented area of a membrane that is occupied by pores, are often unknown.

— If the pores are not highly connected, their resistance to diffusion will depend on their tortuosity; if the solute must diffuse down a tortuous path, it will cross more slowly.

— Transport depends critically on the diameter of the passage along the length of the diffusion path. The walls of pores give rise to a viscous drag that retards the diffusional process, and the degree of retardation remains significant for pores as large as ten times the solute diameter. This effect, which will be discussed in detail in Chapter 7, becomes greater when the diameter of the pore is closer to that of the solute. When the pore is only slightly larger than the solute, the latter must cross the membrane by single-file diffusion, and the augmented diffusion coefficient is no longer the

appropriate measure of solute mobility. Of course, if the pore size is less than the solute size, then the solute does not cross the membrane at all, irrespective its aqueous diffusion coefficient.

— In small pores, there is considerable opportunity for chemical and physical interactions between the solute and the walls. Such interactions include adsorption and binding. Furthermore, if the pore walls are charged, Donnan effects can cause the concentration of charged solutes inside the pore to be markedly different from the concentrations in the adjacent solutions. There can also be interactions between the *solvent* and the walls of the pore, which can cause the effective solvent viscosity to differ from that in free solution, thereby (see §2.1.4), affecting the solute diffusion coefficient.

Once the permeability is specified, the transmembrane flux is predicted by

$$J_s = k_s \Delta c_s. \tag{2.20}$$

Equation (2.20) is the expression most commonly used to describe the passive free diffusion of a nonelectrolyte across a barrier. It can also describe the transport of an ion in the absence of an electrical potential gradient. Some illustrative values of solute permeability are given in Table 2.1.

Table 2.1. Nonelectrolyte Permeabilities of Three Cell Membranes

Ox erythrocyte[a]	cm/s
Urea	7.8×10^{-5}
Glycol	2.1×10^{-6}
Diethylene glycol	7.5×10^{-7}
Glycerol	1.7×10^{-8}
Ehrlich ascites tumor cell (mouse)[b]	**cm/s**
Galactose	2.1×10^{-6}
3-methyl glucose	1.8×10^{-6}
Sorbose	5×10^{-7}
Glucosamine	2.5×10^{-7}
Human erythrocyte[c]	**cm/s**
Ethanol	2.1×10^{-3}
Glycerol	1.6×10^{-7}
Thiourea	1.1×10^{-6}
Urea	7.7×10^{-7}

[a] Davson and Danielli (1952).
[b] Crane et al. (1957).
[c] From summary in Lieb and Stein (1986).

2.1.3. Unstirred Layers

Assume that $c_s^{\mathrm{I}} > c_s^{\mathrm{II}}$; by our conventions, $\Delta c_s > 0$, $J_s > 0$, and solute moves from Phase I to Phase II. Before a solute molecule can cross the membrane, it must first find its way from the bulk of Phase I to the membrane interface at $x = 0$. Two mechanisms are available to accomplish this: diffusion, which is described by equations like those given above, and convection, in which fluid motion carries solute from the main body of the phase to the proximity of the membrane.

If one of the phases is the inside of a cell, convection is limited, and exchange between the bulk of the cytoplasm and the membrane interface is largely by diffusion. Similarly, diffusion is usually the dominant transport mechanism in the extracellular space on the other side of the cell membrane. In many experimental situations, however, convection can be introduced by stirring. The influence of stirring does not extend unattenuated to the membrane–solution interface; a thin, effectively *unstirred layer* adjacent to the membrane remains. Solute crosses this layer only by diffusion, and its flux is properly defined by Eq. (2.18); at $x = 0$,

$$J_s = \frac{D_s^{\mathrm{I}}}{\delta^{\mathrm{I}}}(c_{sb}^{\mathrm{I}} - c_s^{\mathrm{I}}), \tag{2.21}$$

where D_s^i is the solute diffusion coefficient in the ith phase, δ^i is the thickness of the corresponding unstirred layer, and the subscript "sb" denotes the bulk phase concentration of the solute; c_s^i is the solute concentration at the interface between the membrane and the ith phase. For nonideal solutions, the augmented diffusion coefficient would be used in place of D_s.

Similar considerations apply to the transport of solute from $x = a$ to the bulk of Phase II. The solute concentration profile is shown in Figure 2.1. Three resistances in series separate the two bulk phases. The solute flux is equal to the overall concentration difference divided by the sum of these resistances, each of which is inversely proportional to a permeability:

$$J_s = \frac{c_{sb}^{\mathrm{I}} - c_{sb}^{\mathrm{II}}}{\delta^{\mathrm{I}}/D_s^{\mathrm{I}} + 1/k_s + \delta^{\mathrm{II}}/D_s^{\mathrm{II}}}. \tag{2.22}$$

Here, k_s is the true permeability of the membrane. The *apparent permeability* of the membrane, $J_s/\Delta c_{sb}$, is the reciprocal of the denominator in the preceding equation.

The characterization of the unstirred layer (or *diffusion layer*, as it is also known) as a sharply defined boundary layer containing all of the diffusional resistance outside the membrane is clearly an approximation. When the bounding phase is unstirred, there are concentration gradients throughout. In the presence of stirring, convective effects are absent *at* the membrane–solution interface and increase with distance from the membrane surface. Notwithstanding the limitations of the unstirred layer concept, it does provide a convenient means for including diffusional resistances outside the membrane in the equation for solute flux, and for characterizing the magnitude of such resistances. The quantity δ^i can be regarded as the thickness of a layer of the external phase whose resistance to diffusion is the same as that actually present outside the membrane.

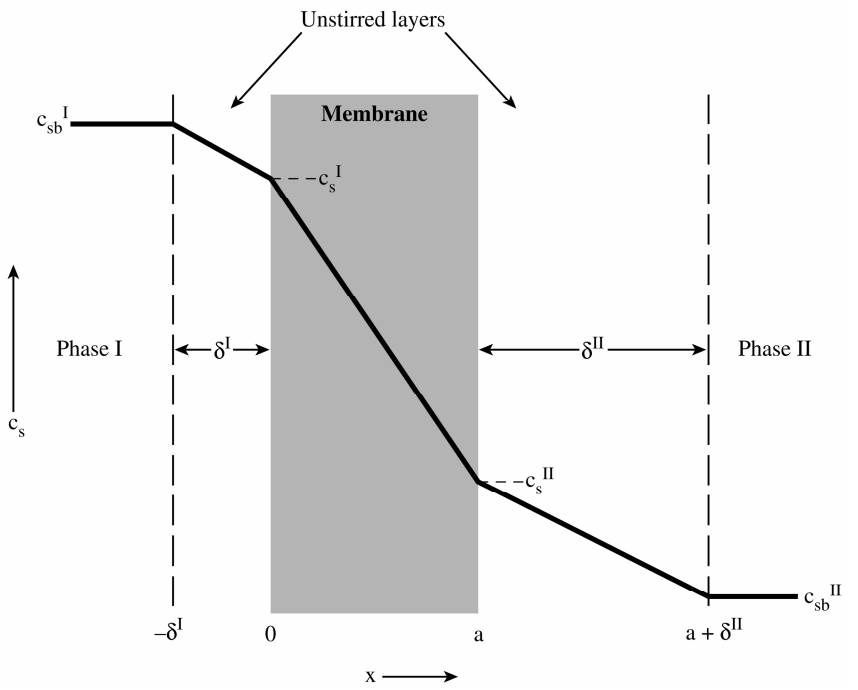

Figure 2.1. Concentration profile in the presence of unstirred layers, and in the absence of solvent flow. The actual transmembrane concentration difference, $c_s^{I} - c_s^{II}$, is less than the overall concentration difference, $c_{sb}^{I} - c_{sb}^{II}$.

It can be seen from Eq. (2.21) that c_s approaches c_{sb} as the thickness of the unstirred layer approaches zero; otherwise, the solute flux would become infinite. When these two concentrations are assumed to be identical (an assumption that is often made in practice, and will be made liberally in the chapters to follow), the phase is said to be *well-stirred.* Although vigorous stirring can reduce the effective thickness of the unstirred layer, it cannot be reduced to zero; the *well-stirred assumption* is always an approximation. The effects of a variety of stirring motions on solute flux are analyzed in Pedley (1983).

In the presence of unstirred layers, the concentration difference driving the transmembrane flux is less than the difference between the bulk phase concentrations (see Fig. 2.1). Solute permeabilities calculated using the latter driving force can be seriously underestimated if the resistance of the unstirred layers is an important fraction of the total interphase resistance. This is more likely to be the case if the membrane permeability is high. The neglect of unstirred layer effects can also lead to errors in the calculated parameters of carrier-based transport systems (Chap. 4).

The diffusion coefficients of small solutes in the cytoplasm are not known very well, so it is difficult to make good estimates of the errors in cell membrane permeability caused by intracellular diffusional resistance. In such cases, it is common to

assume that there is *no* diffusional resistance on the cytoplasmic side. The true cell membrane permeability is underestimated when this approach is used; however, permeabilities that are derived in this way can be compared with the permeabilities of other solutes derived similarly, or used to predict flux, as long as the bulk cytoplasmic concentration of the solute is used in the flux equation.

We will see in Chapter 6 that, for many solute/membrane combinations, a transmembrane concentration difference induces a solvent flow, termed *osmosis*, in the direction of the more concentrated solution. The solute concentration profile in the unstirred layer is curved when osmosis (or any transmembrane solvent flow) is present. The effect of unstirred layers on transport in the presence of osmosis is discussed in Chapter 10.

Equation (2.22) describes the steady-state transport of solute across a series of resistances, for the case in which two of the resistances are unstirred layers and the third is the membrane itself. The δ/D_s ratios in the equation are simply the reciprocals of the permeabilities of the individual unstirred layers. Equation (2.22) can be regarded as a transport equivalent of Ohm's Law for the voltage-driven current through a number of resistors in series; here, the voltage is replaced by the bulk concentration difference, the current by solute flux, and the ohmic resistances by the reciprocals of the permeabilities of each barrier.

This description of the flux through series barriers can be applied to many biological transport processes, such as transport though a single layer of cells, where solute enters across one face of the cell, crosses the cytosol, and then exits across the other face; transport through a cell supported by a permeable layer of extracellular matrix; or transport through a series of cell layers, as in epithelia (Chap. 10). In such cases, the general equation for solute flux is

$$J_s = \frac{\Delta c_s}{\sum_{i=1}^{m} \frac{1}{k_{si}}}, \tag{2.23}$$

where Δc_s is the overall concentration difference and k_{si} is the solute permeability of the ith of m barriers. As above, the reciprocal of the denominator of Eq. (2.23) is the *apparent permeability* of the composite barrier.

2.1.4. Applications of Solution Theory

A considerable body of theory has been developed to describe free diffusion in solution. Most of this theory cannot be directly applied to biological systems, for reasons that have already been presented. One applicable product of solution theory is the *Stokes–Einstein equation*, which identifies the variables that have the greatest influence on the diffusion coefficient. In general, the diffusion coefficient depends on the solute (naturally), the solvent, the concentration of the solute (or composition, for a multicomponent solution), and temperature.

Einstein (1908) used Stokes' Law to derive the following approximate expression for the diffusion coefficient of a spherical solute:

$$D \approx \frac{RT}{6\pi\eta sN}, \tag{2.24}$$

where η is solvent viscosity, s is solute radius, and N is Avogadro's number. Stokes' Law describes the drag on a sphere moving through a homogeneous fluid of infinite extent. Implicit in this application of Stokes' Law are the assumptions that solute molecules are much larger than those of the solvent, and that the influence of the solution boundaries (e.g., the walls of a pore) is negligible. Equation (2.24) shows that the most important solute property affecting the diffusion coefficient is its size (and shape; the equation is more complex for nonspherical solutes), and the most important solvent property is its viscosity.

Equation (2.24) predicts that the diffusion coefficient is inversely proportional to the solute radius; that is, the sD product is constant. This condition is met by the data in Table 2.2, even though the solute molecules are not much larger than those of the solvent. In biological systems, this simple inverse relation applies only to diffusion through large passages. When the size of the pore is not much greater than that of the solute, the permeability depends on *pore radius* as well as solute radius. The effect of pore size on solute permeability will be discussed in Chapter 7.

Table 2.2. Test of the Stokes–Einstein Equation

Solute	Solute radius, s[a] nm	Diffusion coefficient in aqueous solution at 25°C, D[a] cm^2/s	$sD \times 10^5$ nm-cm^2/s
Methanol	0.20	1.3×10^{-5}	0.26
Urea	0.24	1.16×10^{-5}	0.28
Glucose	0.39	6.8×10^{-6}	0.26
Glycerol	0.31	8.3×10^{-6}	0.27
Sucrose	0.45	5.5×10^{-6}	0.25
Raffinose	0.58	4.2×10^{-6}	0.24

[a] Data from Schafer and Barfuss (1980).

The predicted effect of solvent viscosity on the diffusion coefficient has often been used to interpret and extrapolate experimental permeability data. From the Stokes–Einstein equation, the diffusion coefficient is expected to vary inversely with solvent viscosity. If the temperature dependence of permeability parallels that of the reciprocal of the viscosity of water, this is taken as evidence that the solute crosses the membrane via water-filled pores. The permeabilities of other diffusional transport routes (e.g., across the lipid phase of the cell membrane) are considerably more sensitive to temperature than is the permeability of an aqueous pore. Similarly, if it is known that a solute uses an aqueous pore to cross a membrane, then the temperature

dependence of the viscosity of water can be used to predict the solute permeability at one temperature from the measured permeability at a different temperature.

2.1.5. Fick's Second Law and Convective Diffusion

Fick's first law is one of the equations most commonly used to describe biological transport by free diffusion. It can readily be generalized to any coordinate system:

$$\boldsymbol{J}_s = -D_s \nabla \boldsymbol{c}_s. \tag{2.25}$$

where $\boldsymbol{J}_s$ is the flux vector in three-space. In the steady state, the law of mass conservation applied to the species s is

$$\nabla \bullet \boldsymbol{J}_s = 0. \tag{2.26}$$

Substituting Eq. (2.25) into (2.26),

$$\nabla \bullet (D_s \nabla \boldsymbol{c}_s) = 0. \tag{2.27}$$

Equation (2.27) is the steady-state form of *Fick's second law of diffusion*, also known as the *diffusion equation*. When the diffusion coefficient is uniform, the equation simplifies further to

$$\nabla^2 c_s = 0. \tag{2.28}$$

The diffusion equation has been solved in numerous geometries, for a wide variety of boundary conditions. Table 2.3 summarizes some useful forms of the steady-state diffusion equation.

Table 2.3. Some Forms of the Steady-State Diffusion Equation

1. Cartesian coordinates (x,y,z)
   a) 1-dimensional: $D_s \dfrac{d^2 c_s}{dx^2} = 0$
   b) 3-dimensional: $D_s \left(\dfrac{\partial^2 c_s}{\partial x^2} + \dfrac{\partial^2 c_s}{\partial y^2} + \dfrac{\partial^2 c_s}{\partial z^2} \right) = 0$
2. Cylindrical coordinates (r =radial coordinate, z =longitudinal coordinate, no azimuthal variation)
   a) r-variation only: $\dfrac{D_s}{r} \dfrac{d}{dr} \left(r \dfrac{dc_s}{dr} \right) = 0$
   b) r- and z-variation, different diffusion coefficients in r- and z-directions: $\dfrac{D_{sr}}{r} \dfrac{\partial}{\partial r} \left(r \dfrac{\partial c_s}{\partial r} \right) + D_{sz} \dfrac{\partial^2 c_s}{\partial z^2} = 0$
3. Spherical coordinates, r-variation only: $\dfrac{D_s}{r^2} \dfrac{d}{dr} \left(r^2 \dfrac{dc_s}{dr} \right) = 0$

An additional contribution to the solute flux arises if the solution itself is moving. Let us return to the one-dimensional case described by Eq. (2.9). If the solution is flowing in the x-direction at a velocity v, the solute flux is augmented by a convective term equal to the product of the solution velocity and the local concentration:

$$J_s = -D_s \frac{dc_s}{dx} + vc_s . \tag{2.29}$$

The three-dimensional equivalent of Eq. (2.29) is

$$\boldsymbol{J}_s = -D_s \nabla \boldsymbol{c}_s + \mathrm{c}_s \boldsymbol{v}, \tag{2.30a}$$

where $\boldsymbol{v}$ is now a vector. Substituting Eq. (2.30a) into Eq. (2.26), the equation for steady-state *convective diffusion* with a uniform diffusion coefficient becomes

$$D_s \nabla^2 c_s - v \bullet \nabla c_s - c_s \nabla v = 0 .$$

Biological solutions can be regarded as incompressible, and it can be shown that incompressibility implies $\nabla \boldsymbol{v} = 0$, so the steady-state convective diffusion equation becomes

$$D_s \nabla^2 c_s - v \bullet \nabla c_s = 0 . \tag{2.30b}$$

When $\boldsymbol{v}$ is uniform (as it would be, in the one-dimensional case), Eq. (2.29) can be integrated to give an expression relating flux, the concentration boundary conditions, and velocity. It is easy to add a solute convection term to the more general forms of the diffusion equation given in Table 2.3, but it is not easy to solve the equations that result. Numerical simulation is usually required.

The use of Eq. (2.26) to describe mass conservation in the steady state implies that the diffusing species is neither produced nor consumed in the region of interest. This assumption will generally apply throughout this text. In the Appendix to Chapter 10, the one-dimensional convective diffusion equation [Eq. (2.29)] will be extended to include changes in species concentration resulting from fluxes across the region boundaries. In Chapter 11, Fick's second law, with [Eq. (2.30b)] and without [Eq. (2.27)] convection, will be generalized to include chemical reactions *within* the region and time-dependent behavior.

2.1.6. Justification of the Steady-State Assumption: Time Scales in Biological Transport

Virtually all of the transport processes described in this text are *steady-state* processes; that is, the concentrations in the system — both the external boundary conditions and the conditions inside the barrier — are assumed to be independent of time. When there is neither production nor consumption of the species of interest inside the membrane, mass conservation implies that the steady state flux satisfies Eq. (2.26). In the common one-dimensional description of membrane transport, Eq. (2.26) becomes

simply $dJ_s/dx = 0$; that is, the flux based on a unit membrane surface area is the same at every cross-section in the membrane. We used this fact to integrate Fick's first law, and we will use it again.

Another implication of the steady-state assumption is that the flux is constant in time. But this creates an apparent contradiction: how can the boundary conditions remain constant in the face of a perpetual flux? Clearly, they cannot, and the easy fix is to postulate, at least for the purposes of analysis, that the bounding solutions are infinite in extent, so their compositions do not change even when solute is lost or gained. Infinite systems are convenient to postulate but rare in the real world. Happily, most biological membranes do experience a relatively stable milieu because of *homeostasis* — the tendency of living systems to maintain a constant "internal environment," which includes, for instance, the composition of the extracellular fluid that defines the boundary conditions for solute transport into and out of cells. The maintenance of reasonably constant boundary conditions for biological transport is thus accomplished by other agencies (such as the kidneys) outside the system under study.

Of course, living systems do experience changes in their environment that cause changes in transport rates; these may reflect a failure of homeostatic mechanisms or a sudden challenge to the system occasioned externally or by the behavior of the organism. Some biological processes, such as regulatory events, are inherently dynamic. In such cases, the boundary conditions for transport cannot be regarded as constant, and steady-state solutions would no longer seem to apply.

The proper approach to describing transport under these circumstances depends on the rate at which the boundary conditions change. If they change slowly compared to the rate at which the transport process can adapt to that change, transport can be regarded as *quasisteady*; that is, the transport rate at any time is equal to the steady-state flux corresponding to the boundary conditions *at that time*. If the boundary conditions change more rapidly, a full unsteady-state solution of the diffusion equation *within the membrane* is necessary.

The time for the transport rate to adapt to changing boundary conditions is the time needed for the concentration profile in the membrane to change to the profile appropriate to the new boundary condition. Using a Fourier series solution in slab geometry, Weiss (1996a) obtained a time constant, $t_d = a^2/(\pi^2 D_s)$, for the approach to the steady state *inside* a homogeneous membrane with an arbitrary initial concentration profile, exposed at $t = 0$ to new concentration boundary conditions at each face. As might be expected, t_d is shorter for thinner membranes and for more rapidly diffusing solutes.

One transport process for which the boundary conditions depend on time is the diffusion of a solute into or out of a closed compartment, such as a cell. The rate at which the concentration in the cell changes is related to the solute flux and the surface area and volume of the cell. Assume the interior of the cell is well-mixed and is Phase II, so flux into the cell is positive. The rate of change of the number of moles of solute in the cell, n_s^{II}, is given by

$$\frac{dn_s^{\mathrm{II}}}{dt} = J_s S, \tag{2.31}$$

where S is the surface area of the cell. The concentration of solute in the cell is equal to the number of mols of solute per unit volume: $c_s^{II} = n_s^{II}/V$, where V is the cell volume (we assume for simplicity that all portions of the cell are accessible to the solute). Combining these two equations and Eq. (2.17), with D_s instead of D_s*, we obtain

$$V\frac{dc_s^{II}}{dt} = \frac{D_s(c_s^{I} - c_s^{II})S}{a}. \tag{2.32}$$

Eq. (2.32) is readily integrated for a constant ambient concentration, c_s^{I}. The intracellular concentration follows a decaying exponential in time, with a time constant $t_c = aV/(D_sS)$. The time constant is shorter when the solute passes through the membrane more readily (low thickness, high diffusion coefficient) and when the surface-to-volume ratio of the cell is large.

The quasisteady approximation is appropriate if $t_d << t_c$, or $a << \pi^2 V/S$. Interestingly, the diffusion coefficient does not appear in the criterion, because it affects the time constants of both processes similarly: even as a high membrane diffusion coefficient allows the concentration profile inside the membrane to adapt more quickly, it also causes the intracellular concentration to change more rapidly.

Generally, the quasisteady approach has proven adequate for describing biological transport in the presence of changing boundary conditions. It is an important assumption underlying time-dependent applications of *compartmental analysis*, a modeling technique for complex systems that will be discussed in Chapter 8. Of course, changes in the boundary conditions are not the only tool by which living systems elicit changes in flux. As we shall see, such changes — particularly, rapid changes — are in most cases obtained by altering the transport properties of the membranes themselves.

2.2. FREE DIFFUSION OF ELECTROLYTES

The free diffusion of electrolytes is considerably more complex than that of nonelectrolytes. The basic flux equation for electrolytes is the electrodiffusion equation. This nonlinear equation is soluble, but the general solutions are so complex that they have rarely been applied to biological systems. A general solution of the electrodiffusion equation, and a number of special cases, are given below.

2.2.1. Differences between Electrolyte and Nonelectrolyte Diffusion

There are two principal differences between the diffusion of electrolytes and nonelectrolytes:

1. Charged solutes are subject to electrical forces when electrostatic potential gradients are present. Accordingly, the driving force for electrolyte transport is the gradient of the *electrochemical potential* rather than that of the chemical potential.
2. Since any electrolyte solution must contain at least one anion and one cation, there are always at least two solute species. The existence of mul-

tiple species — and, correspondingly, multiple fluxes — leads to two concepts that arise only when electrolyte transport is considered.

The first of these concepts is *electroneutrality*: the concentration of positive charges in a small sample volume equals the concentration of negative charges. This condition can be written as follows:

$$\sum_i z_i c_i = 0 . \tag{2.33}$$

The second concept is *ionic current*. Ions moving in solution carry current just as electrons do in metal conductors. The contribution of each species to the current density is equal to the product of the species' flux and its charge. The current density is obtained by summing these contributions:

$$I = \sum_i I_i = \sum_i z_i J_i . \tag{2.34}$$

where $I_i = z_i J_i$ is the contribution of the ith ion to the current. Note that the units of I as given above are *mols* of charge per square centimeter of transport area per second. If the right-hand side of Eq. (2.34) is multiplied by the Faraday, the units become coulombs per square centimeter per second; that is, amperes per square centimeter.

2.2.2. The Electrodiffusion Equation

The flux of the ith ion in free solution, like that of the nonelectrolytes in the preceding section, is equal to the product of the mobility of the ion, its concentration, and the appropriate driving force, which in this case is $-d\tilde{\mu}_i / dx$. The driving force can be written in terms of the chemical and electrostatic potential gradients:

$$-\frac{d\tilde{\mu}_i}{dx} = -\frac{d\mu_i}{dx} - z_i \mathfrak{F} \frac{d\psi}{dx} . \tag{2.35}$$

The chemical potential gradient is treated as in the previous section, and the flux equation becomes

$$J_i = U_i c_i \left(-\frac{RT}{c_i}\frac{dc_i}{dx} - z_i \mathfrak{F} \frac{d\psi}{dx} \right) = -U_i RT \frac{dc_i}{dx} - U_i c_i z_i \mathfrak{F} \frac{d\psi}{dx} . \tag{2.36}$$

Equation (2.36) is the *electrodiffusion equation*, which is the most common starting point for describing free diffusion in electrolyte solutions. It is also known as the *Nernst–Planck equation*. As in the analysis of the Donnan equilibrium, concentrations are used rather than activities, to facilitate the use of the electroneutrality condition in solving the equation. As a consequence, the solutions of this equation neglect direct ion–ion interactions during the transport process, and the analysis to follow strictly holds only for solutions more dilute than those found in living systems.

The electrodiffusion equation defines the dependence of ionic flux on the *gradients* in concentration and electrostatic potential in the membrane or barrier across which transport takes place. These gradients are not generally measurable. It is therefore desirable to integrate the equation, so that the fluxes can be related to the conditions at the membrane surfaces.

The results of this integration will reveal a much more complex dependence of flux on boundary conditions than was the case when nonelectrolytes were considered. In dilute solutions, the flux of an *un*charged species depends on only the concentration of that solute in the bathing solutions; this is so even when other solutes are present. In electrolyte solutions, however, the flux of each ion depends on the concentrations of all ions in the bounding solutions, and not in a simple fashion. In addition, for any particular pair of bounding compositions, the fluxes, and hence the transmembrane current density, depend on the transmembrane potential difference. This is illustrated in Figure 2.2a.

Shown in Figure 2.2b are two common experimental situations.

1. **Short Circuit**. Electrodes in the two bathing solutions are connected by an external circuit, "shorting out" the membrane and bringing the potential difference across it to zero. The current density measured under short-circuit conditions is called the *short-circuit current*. Passive ion fluxes at short circuit are driven by only concentration gradients and can therefore be described by the same equations as are used to describe the flux of nonelectrolytes. Accordingly, ionic fluxes at short-circuit can be expressed in terms of the membrane permeabilities of the ions, following Eq. (2.20). It is easy to calculate the short-circuit current from the membrane properties and bounding compositions, since at short-circuit the fluxes of the ions are independent of one another and the flux of each can be calculated from Eq. (2.20).
2. **Open Circuit**. At open circuit, there is no net transport of charge across the membrane; that is, $I = 0$. The zero-current condition is more typical of unmanipulated biological systems. The external path between the two sides of the barrier in Figure 2.2b does not ordinarily exist, and electroneutrality demands that equal amounts of positive and negative charge cross the barrier. At open circuit, the quantity of experimental interest is the potential difference across the membrane. The term "*diffusion potential*" is also used to describe the potential difference that develops across a membrane when the current is zero and all flux is passive. Under certain assumptions, the electrodiffusion equation can be integrated to compute the open-circuit potential and fluxes from the compositions of the solutions on the two sides of the membrane and the ionic mobilities within it; the more general solutions are presented first.

Before proceeding, it should be remarked that many cellular and intracellular membranes contain active transport systems that generate a net ionic flux and corresponding active ionic current, I^a. The zero-current condition in this case is $I^a + I = 0$, where I is the *passive* current described by the electrodiffusion equation.

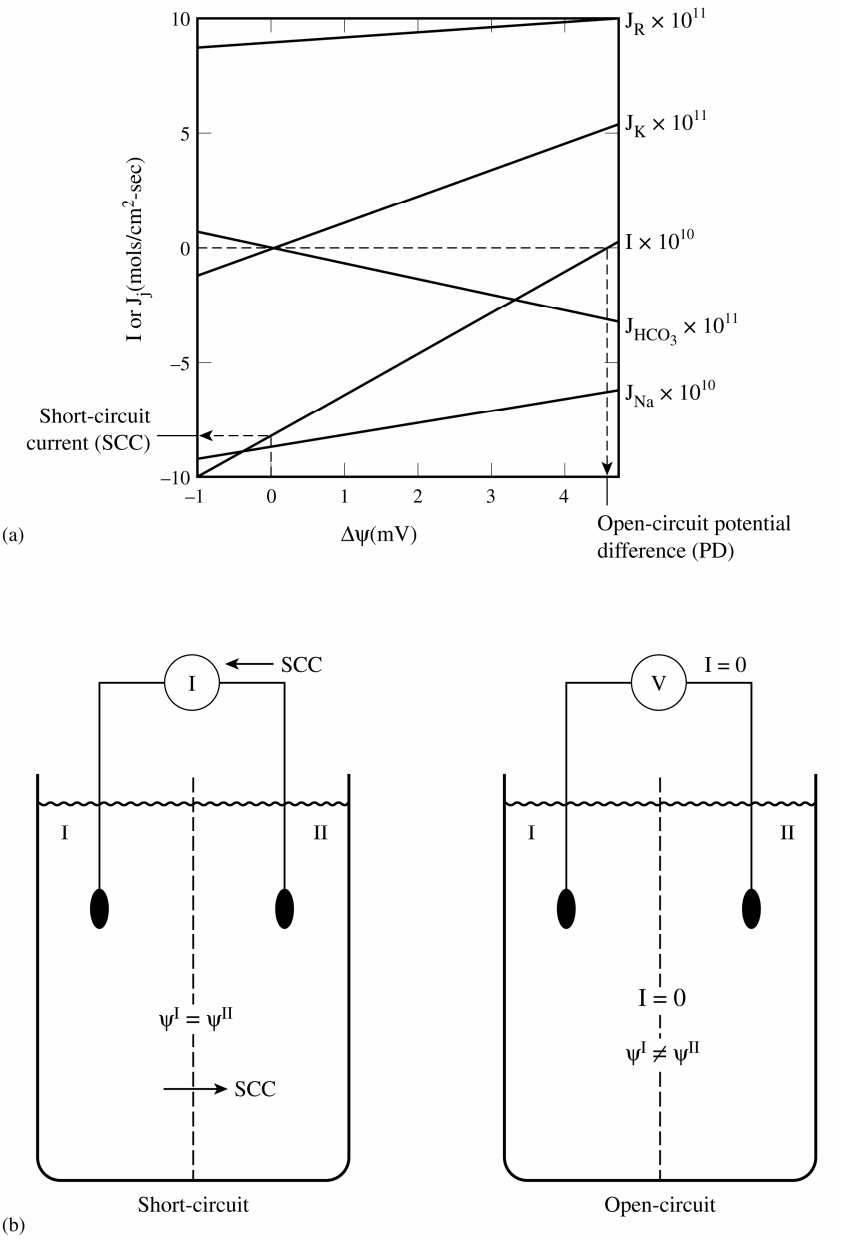

Figure 2.2. (**a**) A solution of the electrodiffusion equation, for an uncharged membrane at 25ºC. The composition of the solution in Phase I is 68 mM NaCl, 15 mM $KHCO_3$, and 68 mM RCl, where R is a large cation whose mobility is one-tenth that of Na; in Phase II, the RCl has been replaced by NaCl. The mobilites of the ions are taken from Table 2.4, and the membrane is modeled as an aqueous film 1 cm thick. Dashed lines denote the short-circuited ($\Delta\psi = 0$) and open circuited ($I = 0$) conditions. After Friedman (1970). (**b**) Short-circuit and open-circuit conditions. In the former, the ammeter measures the short-circuit current; in the latter, the voltmeter measures the open-circuit potential difference.

2.2.3. Integration of the Electrodiffusion Equation

The integration of the electrodiffusion equation is complicated by the nonlinearity of the equation. The nonlinearity arises from the second term on the right-hand side, because the ionic concentration *and* potential gradient are functions of location in the membrane. The second term was absent when nonelectrolytes were considered. In the late nineteenth century, a number of investigators, including Planck (1890) and Behn (1897), reported solutions of the electrodiffusion equation. We will not reproduce these derivations here, limiting ourselves instead to the assumptions and final result. Both investigators made the same assumptions:

1. There are n ions in the system, and Eq. (2.36) holds for each of them; the mobility of each ion is independent of position (x) or local composition. The phases bounding the two faces of the membrane, whose thickness is a, are denoted Phase I and Phase II. The concentration of the ith ion in the jth phase is denoted c_i^j.
2. All ions are univalent. This restriction can be omitted, but the solution is more complicated when the valences of the ions are not all the same.
3. At every point in the membrane, the local composition is electroneutral. This assumption is strictly false whenever the electric field $E = -d\psi/dx$ is nonuniform, but the deviation from electroneutrality is almost always trivial.

   The relation between the nonuniformity of the electric field and the departure of the solution from electroneutrality arises as follows: a volume of solution that is not electrically neutral contains a net charge called the *space charge*, ρ, whose local concentration is equal to $\Sigma_i z_i c_i$. Thus, when the electroneutrality condition [Eq. (2.33)] is satisfied, $\rho = 0$.

   The space charge concentration is related to the gradient of the electric field through the *Poisson equation*: $dE/dx = (\widetilde{\gamma\delta}/\varepsilon)\rho$, where ε is the permittivity of the barrier. Thus, the electroneutrality assumption is strictly correct (i.e., $\rho = 0$) only when the field is uniform (i.e., $dE/dx = 0$).

   When electroneutrality is assumed and the electrodiffusion equation is solved accordingly, the calculated electrostatic potential ψ is not generally a linear function of x; hence the field is not uniform and the electrolyte solution cannot be electrically neutral. For biologically relevant boundary conditions, this inconsistency is unimportant. Using the electric field gradient obtained by solving the electrodiffusion equation with the electroneutrality assumption, the space charge density can be computed from the Poisson equation. This value of ρ is inevitably orders of magnitude less than the concentration of the electrolyte solution itself.

4. The system is in the steady state, so all ionic fluxes are independent of x.

The Planck solution gives a transcendental expression for the membrane potential $\Delta\psi = \psi^{I} - \psi^{II}$ as a function of the bounding compositions. The open-circuit potential is obtained by solving

$$\frac{\xi U^{\mathrm{I}} - U^{\mathrm{II}}}{V^{\mathrm{I}} - \xi V^{\mathrm{II}}} = \frac{\ln \dfrac{N^{\mathrm{I}}}{N^{\mathrm{II}}} - \ln \xi}{\ln \dfrac{N^{\mathrm{I}}}{N^{\mathrm{II}}} + \ln \xi} \cdot \frac{\xi N^{\mathrm{I}} - N^{\mathrm{II}}}{N^{\mathrm{I}} - \xi N^{\mathrm{II}}} \tag{2.37}$$

for ξ, where $\xi = \exp(\tilde{\mathfrak{F}}\Delta\psi/RT)$,

$$U^j = \sum_{\text{cations}} U_i c_i^j, \quad V^j = \sum_{\text{anions}} U_i c_i^j,$$

and N^j is the *total concentration* of the jth phase, defined as

$$N^j = \sum_{i=1}^{n} c_i^j .$$

The Behn solution provides the fluxes as well as the membrane potential, and consists of a set of $n + 1$ equations that are solved simultaneously:

$$\left(\frac{\tilde{\mathfrak{F}}}{RT}\right)\Delta\psi = g' \ln \frac{N^{\mathrm{II}}}{N^{\mathrm{I}}}, \tag{2.38a}$$

$$J_i = (g' z_i - 1) \cdot \frac{U_i RT}{a} \cdot \frac{(N^{\mathrm{II}} - N^{\mathrm{I}})(c_i^{\mathrm{II}} \mathrm{e}^{-z_i \tilde{\mathfrak{F}} \Delta\psi / RT} - c_i^{\mathrm{I}})}{N^{\mathrm{II}} \mathrm{e}^{-z_i \tilde{\mathfrak{F}} \Delta\psi / RT} - N^{\mathrm{I}}} \quad (i = 1,\ldots,n). \tag{2.38b}$$

As before, flux from Phase I to Phase II is positive.

If the compositions $\{c_i^{\mathrm{I}}\}$ and $\{c_i^{\mathrm{II}}\}$ are specified, and the mobilities are known, then Eqs. (2.38) constitute $n + 1$ equations in $n + 2$ unknowns: an unspecified constant, g; the membrane potential, $\Delta\psi$; and n fluxes, $\{J_i\}$. Since there is one more unknown than there are equations, one of the unknowns, or a function of them, must be specified. Generally, this is either the membrane potential or the transmembrane current density. If the potential is specified, Eq. (2.38a) can be solved for g', and Eq. (2.38b) gives the fluxes directly; a more difficult iterative procedure is required if the current is given and the membrane potential and fluxes are sought.

Strictly speaking, the superscripts "I" and "II" denote the potentials and concentrations *just inside* the membrane faces. However, in applying the preceding solutions, the concentrations that are generally used are those in the bounding phases, and the membrane potential is measured in the external solutions as well. Often, no harm is done when this approximation is made, but errors can arise if a bounding phase contains charged species that cannot enter the membrane, or if the membrane contains fixed charges that cannot leave (Fig. 2.3). In such cases, the correct (i.e., *intra*membrane) boundary conditions for Eqs. (2.37) and (2.38) are related to the composition and potentials in the bathing solutions by the Donnan equilibrium expressions of the previous chapter. The solutions given above must be further modified when the membrane structure is charged, because the concentration of the charges on the membrane must be included in the electroneutrality condition (see §2.2.6).

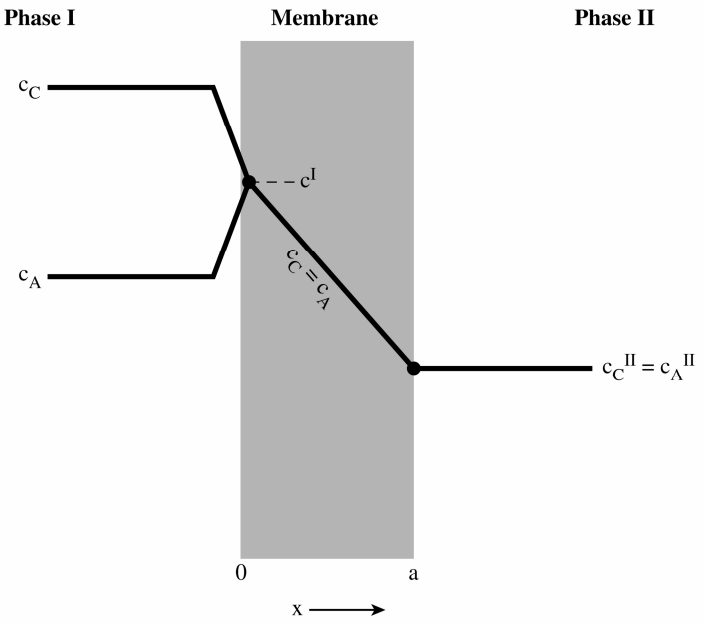

Figure 2.3. Boundary conditions for solutions of the electrodiffusion equation. In the example shown here, there is one mobile cation (*C*) and one mobile anion (*A*) in the system. Both bounding phases are well stirred. Phase I contains a negatively charged species that cannot enter the membrane, so there is a Donnan equilibrium at $x = 0$. Inside the uncharged membrane and impermeant-free Phase II, $c_C = c_A$, by electroneutrality. The correct boundary conditions for the electrodiffusion equation are those at the *filled circles*. The potential difference between Phases I and II equals the sum of the calculated membrane potential and the Donnan potential at the Phase I interface.

Before proceeding to the conditions under which simpler solutions of the electrodiffusion equation can be obtained, we should observe that the equation is linear in c_i; therefore a partial integration can be carried out using an integrating factor. This procedure demonstrates that the ion flux is the product of three terms: $U_i RT$, $c_i^{\mathrm{II}} \exp(-z_i \tilde{\delta} \Delta\psi / RT) - c_i^{\mathrm{I}}$, and the reciprocal of the integral of $\exp[z_i \psi(x)]$ across the membrane. Since $\psi(x)$ is generally not known *a priori*, this equation cannot give flux directly; however, the first term shows that the flux is proportional to the mobility of the ion, and the second demonstrates that the flux of an ion at equilibrium is zero (see also §2.2.4).

2.2.4. Some Special Cases

Equilibrium. By setting $J_i = 0$ in Eq. (2.38b), we obtain the conditions under which the *i*th ion is in equilibrium across the membrane. The flux is zero when the last factor in the numerator is zero:

$$c_i^{\mathrm{II}} e^{-z_i \tilde{\delta} \Delta\psi / RT} - c_i^{\mathrm{I}} = 0 . \tag{2.39}$$

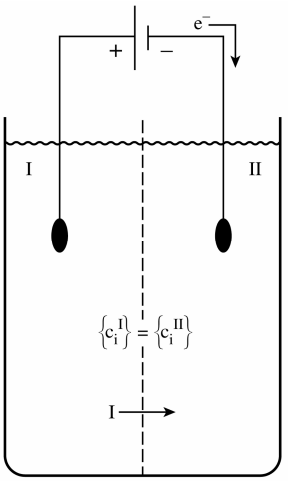

Figure 2.4. Potential-driven current, uniform composition. The electron flow in the external circuit and the current across the membrane are both directed from Phase I to Phase II because the transmembrane current is defined as the flow of *positive* charge [Eq. (2.34)].

It is easy to show that Eq. (2.39) prescribes that the *i*th ion is in equilibrium when its Nernst potential is equal to the membrane potential. This conclusion was also reached in Chapter 1.

Uniform Composition. The solution of the electrodiffusion equation proceeds more directly when the compositions of the solutions on the two sides of the membrane are the same, and the composition inside the membrane is consequently uniform. When a potential is applied across the membrane, the ions migrate across, driven by the electric field, and generate an ionic current; this situation is illustrated in Figure 2.4. The concentration gradient of each ion is zero, so Eq. (2.36) becomes:

$$J_i = U_i c_i z_i \tilde{\mathcal{F}} \left(-\frac{d\psi}{dx} \right). \tag{2.40}$$

Since c_i is independent of x, Eq. (2.40) can easily be integrated to give the ionic flux as a function of the imposed potential difference:

$$J_i = \frac{U_i c_i z_i \tilde{\mathcal{F}} \Delta\psi}{a}. \tag{2.41}$$

Each flux is proportional to the potential difference across the membrane. It follows trivially from Eq. (2.34) that the membrane current density is similarly proportional to the membrane potential; hence, when the composition is uniform, the membrane obeys Ohm's Law. If the fraction of membrane area available for transport is φ, and the transport paths are sufficiently large that the effects of the pore walls on the transport rate can be neglected, the conductance of the membrane is

$$g \text{ (mols/cm}^2\text{-sec-V)} \equiv \frac{I}{\Delta\psi} = \frac{\tilde{\mathcal{F}}\varphi}{a} \sum_i U_i c_i z_i^2 .$$

In electrical units, conductance is measured in *siemens;* 1 S = 1 ohm^{-1} = 1 ampere/volt. The membrane conductance in S/cm^2 is obtained by multiplying the previous value of g by $\mathfrak{F}$.

If, in Eq. (2.40), the electrostatic potential gradient is regarded as the driving force for transport, then, according to the Teorell equation, the product $U_i z_i \mathfrak{F}$ assumes the role of a mobility. Indeed, the absolute value of this product is termed the *electrical (or electrophoretic) mobility* of the ion, $u_i = U_i |z_i| \mathfrak{F}$. The electrical mobilities of several biologically important ions are presented in Table 2.4.

Table 2.4. Electrical Mobilities of Several Biologically Important Ions at 25ºC (Robinson and Stokes, 1965; Davies, 1968)

Ion	Electrical mobility, cm^2/sec-V, $u_i \times 10^4$
H	36.25
Li	4.01
Na	5.19
K	7.62
NH_4	7.62
Mg	5.50
Ca	6.17
Cl	7.92
NO_3	7.41
HCO_3	4.62

Diffusion Potential of a Bi-Ionic System. An explicit solution for the diffusion potential can be obtained from Eqs. (2.38) if the system contains only one anion and one cation, of equal charge. In this case, the anion and cation fluxes are equal, since the current is zero, and the membrane potential is

$$\Delta\psi = \frac{RT}{\mathfrak{F}} \cdot \frac{U_C - U_A}{U_C + U_A} \ln \frac{c^{II}}{c^{I}}, \tag{2.42}$$

where c is electrolyte concentration and the subscripts on the mobilities denote the cation and anion. The diffusion potential is independent of membrane thickness, and depends on only the mobility ratio U_C / U_A (divide numerator and denominator by U_A to see this) and the concentration ratio c^{II}/c^{I}.

The origin of the diffusion potential is easiest to explain for this case in which only two ions are present. If the membrane is permeable to both the anion and cation of a salt whose concentration is different on each side of the membrane, both ions will cross. The ionic fluxes must be equal when the current is zero, even though the mobilities of the two ions are not generally the same. The diffusion potential develops to compensate for this difference in mobility by increasing the electrochemical potential driving force for the ion having the lower mobility, and decreasing that for the more mobile ion. The potential "pulls" the less mobile ion across the membrane, while re-

tarding the flux of the more mobile species. Suppose $c^{II} > c^{I}$ and $U_C > U_A$. Then the salt diffuses from Phase II to Phase I under its concentration gradient, and the potential of Phase I becomes positive relative to that of Phase II (i.e., $\Delta\psi > 0$), so as to retard C and increase the driving force for A.

When $U_C = U_A$, there is no mobility difference to compensate for, and the diffusion potential is zero. Diffusion potentials can cause artifacts in certain electrophysiological experiments, and it is desirable to avoid them. Much use is made of concentrated KCl solutions (salt bridges) in such setups, because the mobilities of potassium and chloride are almost identical.

Consider the other extreme, in which the mobility of one ion is much larger than that of the other, say $U_C >> U_A$. In this case, the membrane potential given by Eq. (2.42) approaches $(RT/\mathfrak{F})$ ln c^{II}/c^{I}. Since there is only a single electrolyte in the system, the argument of the logarithm is also c_C^{II}/c_C^{I}; thus the membrane potential approaches the Nernst potential of the cation. We shall see that the *tendency of the membrane potential to approach the Nernst potential of the more (or most) permeable ion* is evident for more complex electrolyte solutions as well. This tendency has been exploited experimentally to "clamp" the membrane potential at a selected value by bathing the two sides of the membrane with solutions containing different concentrations of an ion (often potassium) to which the membrane is particularly permeable.

Active and Passive Exchange with a Closed Compartment. In the steady state, the net rate of entry of any species into a closed compartment equals the rate at which it is consumed; otherwise, its concentration in the compartment would change with time. Similarly, when a compound is synthesized in a closed compartment, the synthesis rate (less any consumption of the material inside the compartment) equals the rate at which the substance leaves the compartment. When the solute is neither consumed nor produced within the compartment, its net entry rate must be zero in the steady state. This is the case for most ions.

As noted earlier, cell membranes are capable of actively transporting ("pumping") ions between the interior of a cell and the extracellular fluid. Suppose that two cationic species with the same valence, z, are exchanged across the cell membrane, such that, for each ion of Species 1 that is pumped from Phase II to Phase I, r ions of Species 2 are pumped from Phase I to Phase II. In the steady state, the net rate at which each ion crosses the cell membrane — the active flux plus the passive flux — must be zero. Hence the passive flux of each ion is the negative of its active flux. Thus, for each ion of Species I moving passively from Phase I to Phase II, r ions of Species 2 move passively from Phase II to Phase 1:

$$J_2 = -rJ_1, \tag{2.43}$$

where r is the *coupling ratio* or *coupling coefficient* of the pump. Substituting Eq. (2.43) into Eq. (2.36), and rearranging,

$$-U_2RT\frac{dc_2}{dx} - U_2c_2z\mathfrak{F}\frac{d\psi}{dx} = rU_1RT\frac{dc_1}{dx} + rU_1c_1z\mathfrak{F}\frac{d\psi}{dx}. \tag{2.44}$$

Solving for $d\psi/dx$,

$$\frac{d\psi}{dx} = -\frac{RT}{z\tilde{\mathcal{F}}}\left(\frac{rU_1\dfrac{dc_1}{dx} + U_2\dfrac{dc_2}{dx}}{rU_1c_1 + U_2c_2}\right). \tag{2.45}$$

The function in the parentheses on the right-hand side is equal to

$$\frac{\dfrac{d(rU_1c_1 + U_2c_2)}{dx}}{rU_1c_1 + U_2c_2} = \frac{d\ln(rU_1c_1 + U_2c_2)}{dx};$$

hence,

$$d\psi = -\frac{RT}{z\tilde{\mathcal{F}}}d\left[\ln(rU_1c_1 + U_2c_2)\right]. \tag{2.46}$$

Integrating across the membrane, and letting $z = 1$,

$$\Delta\psi = \frac{RT}{\tilde{\mathcal{F}}}\ln\frac{U_2c_2^{\mathrm{II}} + rU_1c_1^{\mathrm{II}}}{U_2c_2^{\mathrm{I}} + rU_1c_1^{\mathrm{I}}}. \tag{2.47}$$

This equation, which is obtained directly from the electrodiffusion equation, was used by Mullins and Noda (1963) to relate the membrane potential of skeletal muscle to the stoichiometry of active Na–K exchange across the muscle fiber membrane.

Equal Total Concentrations on the Two Sides of the Membrane: The Constant-Field Equation. Even though the concentrations of individual ions vary widely in the body, the *total* ionic concentration, N, is quite uniform throughout (Table 2.5). The intracellular ionic content is only 10% less than the extracellular value, so the solution of the electrodiffusion equation for $N^{\mathrm{I}} = N^{\mathrm{II}}$ is of some interest. This solution is also simpler — and much more frequently used — than the more general solutions given earlier.

Table 2.5. Typical Ionic Content of Intracellular and Interstitial Fluids, and Blood Plasma[a]

Ion	Plasma	Interstitial fluid	Intracellular fluid
Na	142	139	14
K	4	4	140
Ca	1	1	<<1
Mg	1	1	20
Cl	108	108	4
HCO_3	24	28	10
Phosphates	2	2	65[b]
SO_4	<1	<1	1
Lactate	1	1	2
Total	283	284	256

[a] Concentrations are in mM. Adapted from Guyton and Hall (2000).
[b] Includes larger molecules to which phosphate groups are attached.

The solution for this special case cannot be obtained directly from Eqs. (2.38), which become indeterminate. We begin the derivation by using Eq. (2.36) to construct two sums:

$$\sum_{i=1}^{n} \frac{J_i}{U_i} = -\left(RT \sum_{i=1}^{n} \frac{dc_i}{dx} + \mathfrak{F} \frac{d\psi}{dx} \sum_{i=1}^{n} c_i z_i \right), \tag{2.48}$$

$$\sum_{i=1}^{n} \frac{J_i z_i}{U_i} = -\left(RT \sum_{i=1}^{n} z_i \frac{dc_i}{dx} + \mathfrak{F} \frac{d\psi}{dx} \sum_{i=1}^{n} c_i \right). \tag{2.49}$$

In Eq. (2.49), use has been made of the assumption, as was made in the Planck and Behn solutions, that the ions are monovalent, so $z_i^2 = 1$. The predominance of monovalent ions is evident from Table 2.5.

Consider each of the four sums on the right-hand sides of the two equations just written. The first sum on the right-hand side of Eq. (2.48) is simplified by interchanging the order of summation and differentiation:

$$\sum \frac{dc_i}{dx} = \frac{d}{dx} \sum c_i = \frac{dN}{dx} . \tag{2.50}$$

The second sum on the right-hand side of Eq. (2.48) is zero, by the electroneutrality condition, Eq. (2.33). The first sum on the right-hand side of Eq. (2.49) is also zero, because it is the derivative of a quantity that is uniformly zero:

$$\sum z_i \frac{dc_i}{dx} = \frac{d}{dx} \sum z_i c_i = \frac{d0}{dx} = 0 . \tag{2.51}$$

The second sum on the right-hand side of Eq. (2.49) is N, by definition. Thus, Eqs. (2.48) and (2.49) can be rewritten:

$$\sum \frac{J_i}{U_i} = -RT \frac{dN}{dx} , \tag{2.52}$$

$$\sum \frac{J_i z_i}{U_i} = -\mathfrak{F} N \frac{d\psi}{dx} . \tag{2.53}$$

These equations are not easy to solve in the general case, because the latter is nonlinear. However, the solution proceeds easily when N is the same on both sides of the membrane. First we recall that the steady-state flux is independent of x; if we assume that the ionic mobilities are also uniform, then the left-hand sides of Eqs. (2.52) and (2.53) are constants. Thus the right-hand sides must also be constants, independent of position in the membrane. From Eq. (2.52), dN/dx is constant, so N is a linear function of x. For the special case of interest here, N is the same at both sides of the membrane; therefore, it must be the same throughout. Since N is uniform and the right-hand side of Eq. (2.53) is constant, the electric field, $-d\psi/dx$, is also uniform. If the potential gradient $d\psi/dx$ is the same everywhere in the membrane, it must be equal to

$(\psi^{II} - \psi^{I})/a = -\Delta\psi/a$. The electrodiffusion equation can then be written in the following form:

$$J_i = -U_i RT \frac{dc_i}{dx} + \left(U_i z_i \Im \cdot \frac{\Delta\psi}{a} \right) c_i . \tag{2.54}$$

The coefficient of c_i in Eq. (2.54) is independent of x, so the equation can readily be solved for the flux as a function of the conditions on the two sides of the membrane. The result is one form of the Goldman–Hodgkin–Katz *constant-field equation:*

$$J_i = z_i \Im U_i \cdot \frac{\Delta\psi}{a} \cdot \frac{c_i^{I} - c_i^{II} e^{-z_i \Im \Delta\psi / RT}}{1 - e^{-z_i \Im \Delta\psi / RT}} . \tag{2.55}$$

The constant-field equation is the equation most commonly used to predict the ion fluxes across a membrane, and hence the membrane current, when the membrane potential and bounding compositions are specified. It clearly satisfies the equilibrium condition: the flux of an ion is zero if its Nernst potential equals the membrane potential. The equation can also be used to find the membrane potential when the current and bounding compositions are specified, but this generally requires a numerical solution. An exception is when the current is zero; in this case, the constant-field equation can be manipulated to predict the diffusion potential:

$$\Delta\psi = \frac{RT}{\Im} \ln \frac{\sum_{\text{cations}} U_i c_i^{II} + \sum_{\text{anions}} U_i c_i^{I}}{\sum_{\text{cations}} U_i c_i^{I} + \sum_{\text{anions}} U_i c_i^{II}} . \tag{2.56}$$

As indicated, the sums in Eq. (2.56) include either all cations or all anions.

Equation (2.55) can be written in nondimensional form, in which the nondimensional flux $\phi = \dfrac{J_i a}{U_i RT c_i^{I}}$ is a function of two nondimensional groups: a nondimensional potential $\beta = z_i \Im \Delta\psi / RT$ whose sign depends on the ionic charge, and the transmembrane concentration ratio $C = c_i^{II}/c_i^{I}$:

$$\phi = \beta \cdot \frac{1 - Ce^{-\beta}}{1 - e^{-\beta}} . \tag{2.57}$$

Plots of ϕ vs. β, parameterized by C, are shown in Figure 2.5. The ion flux is zero when the membrane potential equals the Nernst potential (in nondimensional units, $\beta = \ln C$). As Eq. (2.55) indicates and Figure 2.5 demonstrates, plots of flux vs. potential are curved, so the membrane behaves as a rectifier: equal and opposite deviations from the Nernst potential do *not* in general induce equal and opposite ion fluxes.

Although the electric field given by the electrodiffusion equation is independent of x only when $N^{I} = N^{II}$, the explicit solutions for flux and potential given above have seen considerably more use than the more unwieldy Behn or Planck solutions. As noted earlier, the use of the constant-field equation in many biological applications

can be justified by the fortunate fact that N does not vary very much in living systems. What makes the constant-field equation more remarkable is its utility in spite of important differences between ion transport in real biological membranes and the continuum model implied by the electrodiffusion equation. These differences, which will become apparent in the chapters that follow, are more dramatic than a modest nonuniformity in N. An illustration of the use of the constant-field equation to interpret physiologic data is presented in the next subsection.

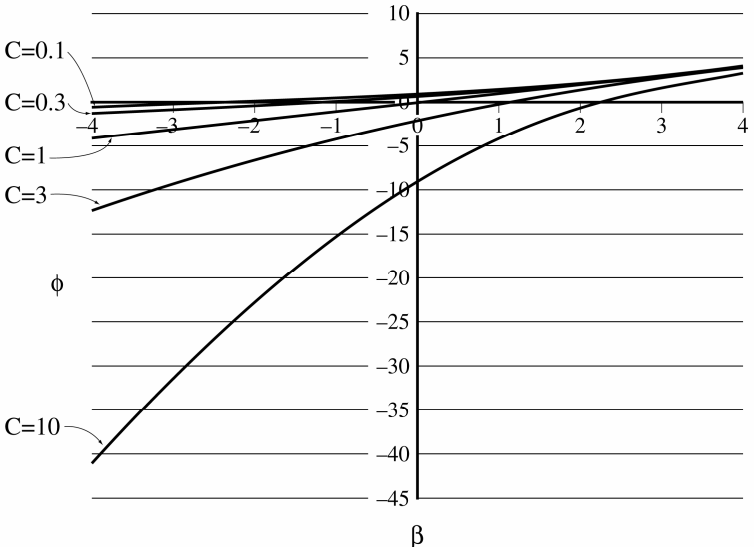

Figure 2.5. Nondimensional representation of transmembrane flux given by the Goldman–Hodgkin–Katz constant-field equation [Eq. (2.55)]. The variables are defined immediately preceding Eq. (2.57).

As was the case for nonelectrolyte transport, the ease with which an ion crosses a biological barrier is generally expressed in terms of its permeability, k_i. The flux equations derived in §§2.2.3 and 2.2.4 — Eqs. (2.38), (2.41), and (2.55) — are based on free solution thermodynamics and are strictly applicable only to transport across a stagnant water film. For such transport, and neglecting nonideal effects, the permeability of the ith ion is related to its diffusion coefficient and mobility in free solution by $k_i^0 = D_i/a = U_iRT/a$, analogous to the relationship for nonelectrolytes. And, as was the case for uncharged solutes, ion permeabilities in biological systems are *experimental* quantities, obtained by measuring the ion flux under known conditions and applying the flux equations presented above, with U_i replaced by k_ia/RT. With this substitution, the first two terms in Eq. (2.55) become $(z_i\tilde{\mathcal{F}}\Delta\psi / RT)k_i$.

An important application of the electrodiffusion equation is prediction of the relationship between ionic permeabilities and the membrane potential. In all such equations derived above — Eqs. (2.37), (2.42), (2.47), and (2.56) — the potential depends on the ratio of linear combinations of mobilities. In free solution, the mobility and permeability are proportional, with the proportionality constant RT/a. If there is a similar proportionality in biological membranes, it is easy to show that the mobilities

in these equations can be replaced by permeabilities. This will be demonstrated in the following section.

2.2.5. Ionic Permeability and the Resting Potential of the Cell

An electrostatic potential difference exists between the interior of biological cells and the extracellular fluid. Generally, the absolute value of this potential is below 100 mV, with the cell interior negative. Comparisons of the Nernst potentials of the primary biological ions — K, Na, and Cl — with the membrane potential show that Cl is generally close to equilibrium across the cell membrane, but the cations are not. The nonequilibrium state of the cations is maintained by *active transport* systems in the cell membrane that pump potassium ions into the cell in exchange for sodium ions, which are pumped out. The pump stoichiometry is such that the number of sodium ions pumped out exceeds the number of potassium ions pumped in; as a consequence, the pump generates an ionic current across the membrane.

Equation (2.56) has been used to estimate the relative cation permeabilities of the cell membrane. As noted earlier, the *total* current across the cell membrane equals the active current due to the Na–K pump *plus* the passive current, which in this case is described by the constant-field equation. The total current across the membrane must be zero, or else charge accumulates in the cell. Thus, if there is an active current, the passive current cannot be zero.

Equation (2.56) was derived under the assumption that the passive current is zero. Even though this is not generally the case for biological cells, the equation has been used to describe the dependence of the cell potential on ionic permeabilities, under the implicit assumption that the passive current is close enough to zero that its effect on the cell potential can be neglected. Considering only the three primary ions given above, and letting Phase I be the inside of the cell and Phase II the outside, Equation (2.56) becomes

$$\Delta\psi_r = \frac{RT}{\mathfrak{F}} \ln \frac{U_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{II}} + U_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{II}} + U_{\mathrm{Cl}} c_{\mathrm{Cl}}^{\mathrm{I}}}{U_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{I}} + U_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{I}} + U_{\mathrm{Cl}} c_{\mathrm{Cl}}^{\mathrm{II}}}, \tag{2.58}$$

where $\Delta\psi_r$ is the cell potential. Since Cl is in equilibrium across the cell membrane, $\Delta\psi_r = E_{\mathrm{Cl}}$:

$$\Delta\psi_r = -\frac{RT}{\mathfrak{F}} \ln \frac{c_{\mathrm{Cl}}^{\mathrm{II}}}{c_{\mathrm{Cl}}^{\mathrm{I}}} = \frac{RT}{\mathfrak{F}} \ln \frac{c_{\mathrm{Cl}}^{\mathrm{I}}}{c_{\mathrm{Cl}}^{\mathrm{II}}}. \tag{2.59}$$

Equating the arguments of the logarithms in the preceding two equations, and rearranging,

$$\frac{c_{\mathrm{Cl}}^{\mathrm{I}}}{c_{\mathrm{Cl}}^{\mathrm{II}}} = \frac{U_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{II}} + U_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{II}}}{U_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{I}} + U_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{I}}}. \tag{2.60}$$

Substituting Eq. (2.60) into (2.59),

$$\Delta\psi_r = \frac{RT}{\mathfrak{F}} \ln \frac{U_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{II}} + U_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{II}}}{U_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{I}} + U_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{I}}}. \tag{2.61}$$

If the permeability and mobility of each cation are related by $U_i = \alpha k_i$, the argument of the logarithm becomes

$$\frac{\alpha k_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{II}} + \alpha k_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{II}}}{\alpha k_{\mathrm{Na}} c_{\mathrm{Na}}^{\mathrm{I}} + \alpha k_{\mathrm{K}} c_{\mathrm{K}}^{\mathrm{I}}}.$$

The proportionality constant cancels out, effectively replacing mobility by permeability. Dividing the numerator and denominator of the argument of the logarithm by k_{K},

$$\Delta\psi_r = \frac{RT}{\mathfrak{F}} \ln \frac{\left(\frac{k_{\mathrm{Na}}}{k_{\mathrm{K}}}\right) c_{\mathrm{Na}}^{\mathrm{II}} + c_{\mathrm{K}}^{\mathrm{II}}}{\left(\frac{k_{\mathrm{Na}}}{k_{\mathrm{K}}}\right) c_{\mathrm{Na}}^{\mathrm{I}} + c_{\mathrm{K}}^{\mathrm{I}}}. \tag{2.62}$$

Equation (2.62) has been used to estimate the Na/K permeability ratio from measurements of the resting potential and intracellular concentrations in solutions of known composition. Values for nerve and muscle at rest range from 0.01 to 0.2; the permeabilities of the two ions in the red cell membrane are closer to one another. The variation of resting potential with the permeability ratio is illustrated in Figure 2.6 for concentrations typical of a nerve fiber.

> Equation (2.62) is derived here as a special case of Eq. (2.56), which relies on the constant-field assumption. However, it can also be derived by setting $r = 1$ in Eq. (2.47), which does not assume a constant field. When the coupling coefficient is unity, there is no active current, so the passive current at open circuit is zero. Indeed, for this particular case, the constant-field assumption is unnecessary.

In this application of equations based on solution theory to transport in real biological systems, an important caveat must be stated. As we have already implied, ions and most other solutes do not cross biological membranes by diffusing down fluid-filled paths that can be regarded as simple extensions of the bounding solution into and through the membrane. Although the constant-field equation and others derived here from solution thermodynamics can describe with some success the effects of boundary conditions and membrane properties on fluxes and potentials, the parameters (such as permeability) that we derive to summarize the experimental results may have a very different *physical* origin than the same parameters when used to describe transport in simple solutions. The permeability ratio of a cell membrane tells us something about how readily various ions cross, but it tells us very little about the physical processes that accompany permeation.

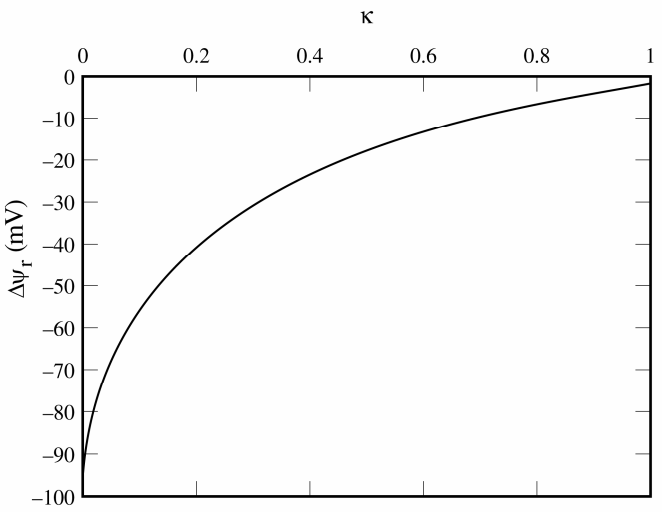

Figure 2.6. Effect of the sodium/potassium permeability ratio κ on cell potential. The internal composition of the cell is 14 mM Na and 140 mM K, and the ambient solution is 142 mM Na and 4 mM K. $T = 37°C$.

2.2.6. Charged Membranes

The membrane matrix can contain dissociated polar groups and consequently possess a net charge. Other membranes, such as the membranes of biological cells, exhibit a surface charge that is due to exposed ionizable groups. We will discuss the first instance here, and the influence of surface charge in the following chapter.

When extending electrodiffusion theory to membranes that contain free charges, the ionizable groups are assumed to be distributed uniformly within the barrier. This assumption is a good one for some systems, such as artificial membranes made from ion exchange resin, or gel-like extracellular structures, including certain connective tissues. It is less applicable to the charged cell membrane pores that we will be discussing in Chapter 4.

The transport process in uniformly charged membranes is described by the model developed by Teorell (1935) and Meyer and Sievers (1936), and which is illustrated in Figure 2.7. When the membrane charge is known, the Donnan equilibrium condition can be used to compute the composition of the solutions just inside each membrane face; these compositions are the boundary conditions for the integration of the electrodiffusion equation across the membrane. The potential difference between the solutions at the two sides of the membrane is equal to the algebraic sum of the Donnan potentials at each face and the transmembrane potential difference obtained from the electrodiffusion equation.

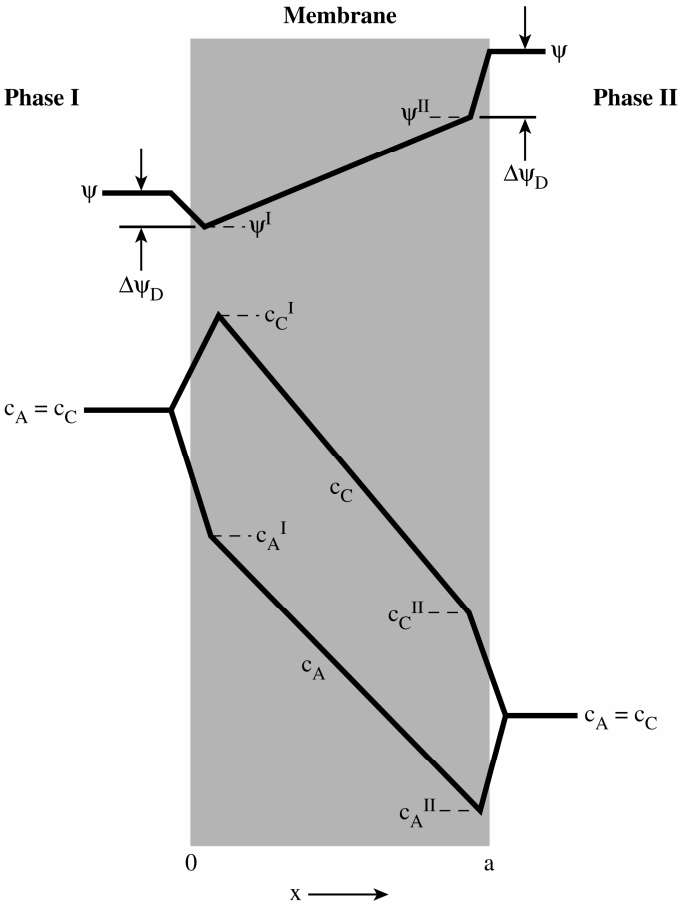

Figure 2.7. The Teorell–Meyer–Sievers (TMS) model. In the example shown here, a negatively charged membrane is bounded by two well-stirred solutions of the same electrolyte, *CA*. The concentrations c_A^{I}, c_C^{I}, c_A^{II}, and c_C^{II}—the boundary conditions for the electrodiffusion equation—are in Donnan equilibrium with the ambient phases. A typical potential profile is also shown, assuming *C* is more mobile than *A*, and $I = 0$. The difference $\psi^{I} - \psi^{II}$ is obtained from the electrodiffusion equation.

The solution of the electrodiffusion equation is more complicated than before because the electroneutrality condition includes the fixed charge:

$$\sum_i z_i c_i + z_X X = 0\,, \tag{2.63}$$

where X is charge concentration and $z_X = \pm 1$. The electrodiffusion equation was integrated by Behn (1897) for a uniformly charged membrane bathed by solutions all of whose ions have the same valence [Eqs. (2.38) were in fact obtained from Behn's original solution by setting $X = 0$]. The solution is given in Harris (1972); its complexity, coupled with uncertainties regarding both the fixed charge concentration in bio-

logical membranes and the appropriateness of the electrodiffusion equation, have strongly inhibited its application to living systems.

From a biological transport perspective, membrane charge is most relevant to the movement of ions through membrane channels. As already noted, the charge distribution in the membrane cannot be regarded as homogeneous for this transport mechanism, which will be discussed in Chapter 4.

2.2.7. Limitations of the Electrodiffusion Equation and Its Solutions

The electrodiffusion equation and the equations derived from it have seen much use in the description of ion transport across biological membranes. It is important, when using these equations, to recognize their limitations and the limitations of conclusions and parameters obtained by applying them to biological data. These limitations depend on the particulars of the application of the electrodiffusion equation, and can be summarized as follows.

The electrodiffusion equation as initially presented in Eq. (2.36) is already a simplification, since it is written in terms of concentration instead of activity; however, the fact that the activity coefficients of the major ions are far from unity has not limited its use as much as has its complexity. Used without further simplification, it can describe free diffusion in solution or through barriers in which the transport process can plausibly be described as free diffusion (i.e., a continuum process through large aqueous pores). In the case of transport through pores, those limitations noted in §2.1.2 that prohibit *a priori* predictions of nonelectrolyte permeability apply to electrolytes as well.

For the special case of uniform total concentration, the electric field in uncharged barriers is uniform, and the constant-field equation applies. Small deviations in total concentration uniformity, of the order of those seen in biological systems, do not disqualify the constant-field equation. Since the constant-field equation derives from the electrodiffusion equation, the dilute-solution and large-pore restrictions continue to apply. Notwithstanding the former restriction, the constant-field equation is generally used to describe the transport of ions whose activity coefficients can be far from unity.

Many biological transport pathways, such as the channels that ions traverse, are not crossed by free diffusion. The total concentration is neither uniform nor continuous at the membrane boundaries. The continuum assumption can fail too, so even the notion of concentration becomes a statistical concept. There are often multiple pathways in parallel, each selective for a different ion or class of ions. The kinetics of transport through such pathways will be discussed in the chapters to come.

Notwithstanding their limited applicability to many biological transport processes, the equations of free diffusion have seen considerable use in describing such processes. They replicate many empirical features of biological transport, such as rectification and the dependence of membrane potential on permeabilities and ambient concentrations. Thus, they still provide a useful tool for correlating experimental data and predicting behavior under conditions not too dissimilar from those under which the experiments were carried out.

PROBLEMS: CHAPTER 2

1. The permeability of urea in the ox erythrocyte membrane is 7.5×10^{-5} cm/s at 37ºC. The diffusion coefficient of urea in water is 1×10^{-5} cm^2/s at 20ºC. How thin would an unstirred layer have to be, at the temperature at which the ox erythrocyte data were obtained, for its resistance to be one-tenth that of the erythrocyte membrane?

2. The erythrocyte membrane in the previous problem is isolated and put in a chamber to measure its urea permeability, as in Figure 2.1. The solutions on both sides of the membrane are stirred but a layer of thickness δ remains at each face. Sketch the variation of the apparent membrane permeability as the unstirred layer thickness varies from 0 to 500 μm. What are the values of apparent permeability at the two extremes of this range of thicknesses?

3. The passive pathways for ions across cell membranes can sometimes be modeled as water-filled channels. The permeability of chloride across the red cell membrane has been estimated to be 2.5×10^{-4} cm/sec at 24ºC. If the effect of temperature on permeability were strictly a viscosity effect, what would the chloride flux be across short-circuited ($\Delta\psi = 0$) red cell membrane at 37ºC when $\Delta c_{Cl} = 10$ mM?

4. A red blood cell can be modeled as a disc 8 μm in diameter and 2 μm thick. The lipid bilayer (Chap. 3) of the red cell membrane is about 5 nm thick. For diffusion through the bilayer, what is the ratio of the time constants t_d and t_c? Can the quasisteady approximation be used to describe the fluxes across the bilayer that arise when the composition outside the cell is changed?

5. Assuming a constant ambient concentration c_s^{I}, integrate Eq. (2.32) from an initial value of $c_s^{\mathrm{II}}(0) = (c_s^{\mathrm{II}})_0$ to obtain $c_s^{\mathrm{II}}(t)$, and confirm that the time constant for the change in intracellular concentration is that given in the text.

6. Assume that the permeability of a particular solute in a membrane, k_s, is high enough that the quasisteady approximation applies. The membrane area is A, and the membrane is bounded by two finite-sized compartments, one of volume V^{I} in which the initial solute concentration is $(c_s^{\mathrm{I}})_0$, and one of volume V^{II} in which the initial solute concentration is $(c_s^{\mathrm{II}})_0$. Derive an expression for the time constant of the decay of the concentration difference between the two compartments.

7. Urea is diffusing out of a spherical cell that is 10 μm in diameter, through large aqueous pores that occupy 5% of the cell surface. T = 37ºC. The diffusion coefficient of urea in water is about 1.4×10^{-5} cm^2/s at 25ºC. The cell membrane is 5 nm thick.
   (a) Estimate the diffusion constant of urea at 37ºC.

(b) Compute the time constant for the decay of the intracellular urea concentration at 37ºC.
(c) How thick can the cell membrane be before the quasisteady assumption for urea diffusion fails (set $t_d = 0.1t_c$)?

8. Consider a water-filled membrane 100 μm thick. The electrostatic potential in the membrane rises quadratically from zero at one side of the membrane to 90 mV at the other side. Show that the space charge in the membrane is uniform throughout its thickness, and calculate its magnitude. The permittivity of water is about 7×10^{-10} coul^2/N-m^2.

9. Show using the Behn and constant-field solutions that when an ion is in equilibrium across a membrane, the flux of that ion across the membrane is zero.

10. The electrical mobility of the sodium ion in water is 5.19×10^{-4} cm^2/sec-V at 25ºC; what is its diffusion coefficient in cm^2/sec?

11. Explain in physical terms the dependence of membrane conductance on each variable (except the Faraday, which is just a conversion constant) in the expression for g that follows Eq. (2.41). Why does conductance depend on the *square* of each ion's charge?

12. A membrane 1 mm thick separates two 155 mM solutions of NaCl at 25ºC. It contains a square array of 100-μm diameter pores on 400-μm centers. What is the conductance of 1 cm^2 of this membrane?

13. (a) Derive Eq. (2.42) from the Planck and Behn solutions.
(b) Use Eq. (2.42) to compute the diffusion potential across a membrane at 25ºC if Side I of the membrane is bathed by a 100-mM solution of NaX and Side II is bathed by 10 mM NaX. Perform this calculation for two values of the mobility of X:
(i) $u_X = 7.9 \times 10^{-4}$ cm^2/s-V (chloride)
(ii) $u_X = 5.2 \times 10^{-4}$ cm^2/s-V (same as sodium)
(c) Compare the preceding results with the diffusion potential given by the constant-field equation [Eq. (2.56)].
(d) Repeat (b) and (c), assuming that all ionic mobilities are half of the values given above. Explain the result in physical terms.
(e) Assuming ideal solutions, show that the diffusion potential becomes equal to the Nernst potential of the cation when the mobility of the anion is zero. Why is this so?

14. Derive Eq. (2.56) from Eq. (2.55).

15. Side I of a membrane is bathed by a 10-mM solution of AX and Side II is bathed by 100 mM AX; T = 37ºC, the solutions are ideal, and the ions are monovalent.

Using either the Behn or constant-field solution, sketch the variation of diffusion potential as U_A/U_X varies from zero to infinity. What are the asymptotic values of the diffusion potential, and why?

16. The electrical mobility of the sodium ion in water is 5.19×10^{-4} cm^2/sec-V at 25ºC; the corresponding values for K and Cl are 7.62×10^{-4} and 7.92×10^{-4}.
    (a) Using the Goldman equation, find the open-circuit potential across a water film separating a 100-mM KCl solution from a 100-mM NaCl solution.
    (b) Use the Goldman and Planck equations to predict the open-circuit potential across the water film when the concentration of the KCl solution is 90 mM.

17. A membrane is bounded by two solutions of NaCl, 100 mM on Side I and 10 mM on Side II. At open circuit, the absolute potential difference across the membrane is 13 mV and the sodium flux is 1.6×10^{-5} mols/cm^2-sec.
    (a) Is the potential at Side I greater or less than that at Side II? Why? Explain in physical terms.
    (b) Using the constant-field equation, calculate the sodium permeability of the membrane.

18. The intracellular concentrations of the major ions in human erythrocytes are: 135 mM K, 17 mM Na, and 77 mM Cl; the concentrations in plasma are: 4 mM K, 138 mM Na, and 116 mM Cl. Chloride is in equilibrium across the cell membrane.
    (a) What is the cell membrane potential?
    (b) What is the Na/K permeability ratio?
    (c) The composition of the plasma in the reference on which this problem is based differs from that in Table 2.5. Repeat (a) and (b), using the Table 2.5 values, to see how sensitive your results are to plasma composition.

19. Confirm that Eq. (2.47) reduces to Eq. (2.61) when the coupling ratio is unity. Why is this the case?

20. Consider the system described in the caption to Figure 2.6. Assume $U_{Na}/U_K = 0.05$. Now assume that increasing amounts of potassium replace the sodium in the external solution, while maintaining the total concentration of the two ions.
    (a) Plot the membrane potential as a function of external potassium concentration, from $c_K^{II} = 4$ mM ($c_{Na}^{II} = 142$ mM, the condition in Fig. 2.6) to $c_K^{II} = 140$ mM ($c_{Na}^{II} = 6$ mM).
    (b) Can further manipulation of the bathing solution, subject to the same constraint on the total external cation concentration, cause the cell potential to become positive?

21. Some membranes have the ability to exclude virtually all ions bearing a particular charge, so the membrane becomes selective for only anions or only cations. Consider a cation-selective membrane of thickness a bounded on Side I by a solution of the 1–1 salt AX at a concentration, c^{I}, and on Side II by a solution of the 1-1 salt BX at a concentration, c^{II}.
    (a) Write an expression for the short-circuit current in terms of mobilities, membrane thickness and bounding concentrations.
    (b) Using the constant-field equation, write an expression for the open-circuit potential of the membrane, in terms of the same variables.