

**1** Today we will begin by discussing the ionic basis of the resting membrane potential. Previously I told you that the resting potential of neurons varies between -40 and -90 mV and that the reason for this difference between the inside and outside of the cell is the differential distribution of ions. Dr. Colvin has already shown you a table of ion concentrations and I show you here Table 5.1 from your book. As we can see sodium, chloride and calcium are more abundant outside the cell and potassium is concentrated intracellularly. In addition there are organic anions [A<sup>-</sup>] that are more highly concentrated inside the cell.

Suppose the membrane of a neuron were permeable to only one ion. If the concentration of this ion were much different inside than outside the cell, ions would flow down the concentration gradient. However, as ions diffuse from one side of the membrane to the other, the voltage would change, and so would the electrical gradient for ion flow.

**(OH 1 bottom)** Let's take potassium as an example. Potassium is more highly concentrated inside the cell and would naturally diffuse outside. However as it does so, the inside becomes more hyperpolarized. Because the voltage inside the cell is more negative than outside, K<sup>+</sup> ions would be drawn against their concentration gradient, back inside the cell. At some point the diffusive force of the concentration gradient will be balanced by the electrostatic attraction and there will be no net flow of ions. The membrane potential where this occurs is called the equilibrium potential and is calculated by the Nernst equation.

**2** The Nernst equation is:  $E_{ion} = \frac{RT}{zF} \ln \frac{C_o}{C_i}$  where C<sub>o</sub> is the concentration of the ion outside the cell, C<sub>i</sub> is the concentration inside, R is the gas constant, T is temperature, F is Faraday's constant and z is the valence of the ion (+1 for Na; +1 for K; +2 for Ca; -1 for Cl). I won't derive this. At room temperature  $\frac{RT}{F}$  is about 25 mV and the conversion from natural log to log base 10 is 2.3 so the Nernst equation becomes  $E_{ion} = \frac{58}{z} \log \frac{C_o}{C_i}$ . What this says is that the equilibrium potential for a monovalent ion changes 58 mV for a 10-fold change in the concentration gradient.

Table 1 says that in squid giant axon, K inside is 400 mM, K outside is 20 mM, and the equilibrium potential for K as computed with the Nernst equation is -76 mV. What this means is that there is a steep concentration gradient for K to diffuse from the inside to the outside of the cell, but that this flow is balanced by electrical attraction to the inside of the cell at -76 mV.

## QUIZ TIME

If the voltage is **-76 mV**, is the net movement of K ions inward or outward?  
There is no net movement at the equilibrium potential, also called the reversal potential.

If the voltage is **-66 mV**, is the net movement of K ions inward or outward?  
ANS. Outward. The concentration gradient is outward. If K leaves the cell down its concentration gradient, what happens to the voltage? The voltage hyperpolarizes. So K will continue to leave the cell and the voltage will continue to hyperpolarize until when? When the voltage reaches the equilibrium potential.

If the voltage is **-86 mV**, is the net movement of K ions inward or outward?  
ANS. Inward. The concentration gradient is still outward, but will the ions move outward? If they do the voltage would hyperpolarize more so this does not happen. The electrical attraction to the negative potential will draw more K inside the cell.

**3** Why doesn't the movement of ions change the concentration gradient and thus change the equilibrium potential? Shouldn't we have to recalculate the Nernst potential when ions move? How many ions have to leave the cell to generate an electrical force that counteracts the diffusive force? Not many. Large changes in concentration are needed to change the Nernst potential for an ion, but only a relatively small change is needed to change voltage across the cell.

Let's calculate how much charge is transferred to change the membrane potential of a 20  $\mu\text{m}$  diameter neuron whose membrane has a specific membrane capacitance  $C_m$  of (what is the typical value I mentioned earlier?)  $1.0 \mu\text{F}/\text{cm}^2$  by 100 mV. Remember the formula for the amount of charge that is separated by a capacitor? What is it?  $Q=CV$ .  $V = 100 \text{ mV}$ ,  $C_m = 1.0 \mu\text{F}/\text{cm}^2$  so the amount of charge is  $100 \text{ nFV}/\text{cm}^2$  and since volt \* Farad = coulomb, we get  $1 \times 10^{-7}$  coulombs/ $\text{cm}^2$ . Since there are about  $10^5$  coulombs/mole (Faraday's constant), this corresponds to about  $1 \times 10^{-12}$  moles/ $\text{cm}^2$ .

The membrane area is  $4\pi r^2$  or  $4\pi (10 \times 10^{-4} \text{ cm})^2$  or  $12.5 \times 10^{-6} \text{ cm}^2$ . So for this area we see a transfer of  $12.5 \times 10^{-18}$  moles.

The volume of the sphere is  $4/3 \pi r^3$  or  $4/3 \pi (10 \times 10^{-4} \text{ cm})^3$  or  $4.2 \times 10^{-9} \text{ cm}^3$  or  $4.2 \times 10^{-12}$  liters.

So the change in concentration is  $12.5 \times 10^{-18}/4.2 \times 10^{-12}$  which is about  $3 \mu\text{M}$ . In squid giant axon, the total concentration of all intracellular ions is about 500 mM; thus to change the neuron's membrane potential by 100 mV corresponds to a change in intracellular ion concentration of  $3 \mu\text{M}/500 \text{ mM}$  or 0.0006%. Your book goes through a similar calculation for the change in K concentration specifically.

**4** We use the Nernst equation to calculate equilibrium potentials of individual ion species in membranes permeable to several ions where the ion concentrations are fixed. Since the membrane potential will rarely equal the reversal potential for a given ion, the quantity  $(V - E_{\text{ion}})$  provides what we call the driving force on the ion. If we know the conductance of an ion (1/resistance), then we can calculate the current across the membrane for the ion  $I_{\text{ion}} = g_{\text{ion}}(V - E_{\text{ion}})$ .

**5** Now the membrane is permeable to several ions, Na, K, Ca, and Cl. The membrane potential will typically not be equal to the Nernst potential of any one of these ions, but will be somewhere in between. The steady-state membrane potential for a given set of ion concentrations and permeabilities is given by the Goldman-Hodgkin-Katz equation or

$$V_m = \frac{RT}{F} \ln \frac{p_K [K^+]_o + p_{Na} [Na^+]_o + p_{Cl} [Cl^-]_i}{p_K [K^+]_i + p_{Na} [Na^+]_i + p_{Cl} [Cl^-]_o} \text{ or}$$

$$V_m = 58 \log \frac{p_K [K^+]_o + p_{Na} [Na^+]_o + p_{Cl} [Cl^-]_i}{p_K [K^+]_i + p_{Na} [Na^+]_i + p_{Cl} [Cl^-]_o}$$

The subscripts i and o refer to the concentrations inside and outside the cell. Note that  $[Cl^-]_i$  and  $[Cl^-]_o$  are reversed relative to Na and K because of the difference in charge. Note also that calcium is not included. Calcium complicates things because it is a divalent ion and here there is an implicit  $z=1$  next to the F in the first equation. It can be included, but the result is not pretty! Also the resting permeability to Ca is small (and so contributes little to  $V_m$ ).

In squid giant axon the relative permeabilities at rest are:  $p_K:p_{Na}:p_{Cl} = 1:0.04:45$ . These numbers can be plugged into the GHK equation to compute the expected resting potential. You will do this for homework. The result is a bit depolarized to the actual resting potential because the contribution of the electrogenic Na-K pump will hyperpolarize the cell a few millivolts. 3 Na are pumped out in exchange for 2 K and this is what causes the voltage to hyperpolarize these few mVs.

**6** In general the resting potential will be far from  $E_{Na}$  and  $E_K$  but close to  $E_{Cl}$ . Consequently at rest there are comparatively large Na and K currents. At steady-state, the voltage is not changing and the net total current must equal zero. If we assume that  $E_{Cl} = V_m$  so that there is no net Cl current then at rest, the outward K current must equal the inward Na current despite the much smaller Na permeability. This can occur because the driving force for Na is so much greater than that for K, i.e., the absolute value of  $(V - E_{Na})$  is  $>$  the absolute value of  $(V - E_K)$ .

Now this voltage computed with the GHK equation is a steady-state and not an equilibrium as with the Nernst equation. So if the cell is left by itself, K and Na will diffuse down their concentration gradients, and the gradients would gradually disappear. Eventually  $V_m$  would reach 0. This does not happen does it? Why? The gradients are maintained by ion pumps. Besides the Na-K pump, there are a Ca-ATPase and a Na-Ca exchanger and a Cl-bicarbonate exchanger that operate to keep Ca and Cl at their resting levels. Many cells do not have a Cl pump and in these, Cl is distributed passively. When voltage changes, Cl will diffuse to equilibrate at the new potential.

If a given ion's pump is inhibited or poisoned somehow, the concentration gradient of that ion would decrease and  $V_m$  would change until a new steady-state was achieved. You will do a calculation for an example of this in the problem set.

## 7 QUIZ:

1) Will a change in permeability always change  $V_m$ ?

ANS. Always?--No, Only if  $E_{ion} \neq V_m$

2) Suppose  $E_{ion} = V_m$ . When could the change in permeability have any effect on  $V_m$ ?

ANS. No change normally, unless the permeability of another ion is also changed. Then a change in the permeability of an ion where  $E_{ion} = V_m$  will change the effect a change in permeability of another ion has on  $V_m$ .

## 8 EQUIVALENT CIRCUIT MODEL

The GHK equation needs permeabilities for its calculation. Unfortunately permeabilities are very difficult to measure and this makes application of the equations difficult. So we use conductances.

The alternative model that is used is a simple extension of the conceptual model of the membrane we used earlier—where we represented the membrane as an electric circuit. We divide up the resistance component of the circuit into resistances for each of the ions present as shown here. More typically, we use conductances instead of resistances in our models where  $g_x = 1/r_x$ . Conductance is measured in inverse ohms, sometimes called mho, but these days we call the unit Siemens. Just as before I mentioned input resistance as a parameter to measure, one can just as easily compute input conductance  $G_N$ . In fact  $G_N = 1/R_N$ .

If we express our resistances in terms of conductances our circuit model becomes this. (Compare Fig. 7.1 in the text). This circuit model has an additional feature to note and that is the battery in the electric circuit. The battery is just the Nernst potential for the particular ion that flows in that part of the circuit. It is there to tell us which way the current flows in the circuit. As you know, the direction switches at the Nernst potential.

So what is the expression for current flow along one of these paths.

Previously we derived the equation for membrane current in a membrane patch as  $I_m = I_c + I_r = c_m dV/dt + V/r_m$  where the current through the resistor was computed with Ohm's law. We can use Ohm's law in the new circuit to compute the current for an individual ion. We say  $I = \Delta V/R$  or  $\Delta V \cdot G$ . So what is the potassium current in this case?

**9** The potassium current  $I_K = (V - E_K) \cdot g_K$ .  $(V - E_K)$  is the voltage difference across the membrane compensated with the Nernst potential in the battery. We get this by considering  $V_m = E_K + I_K/g_K$  where the voltage is the sum of the voltage difference across the battery and the voltage difference across the resistance (or conductance). As we discussed earlier, the quantity  $(V - E_K)$  is called the driving force and the driving force determines the direction and magnitude of the resulting current.

Similarly we get  $I_{Na} = g_{Na}(V - E_{Na})$  and  $I_{Cl} = g_{Cl}(V - E_{Cl})$ . So in this case our membrane current can be expressed as:

$$I_m = I_c + I_r + I_{pump} \quad (\text{we often use } I_{ion} \text{ instead of } I_r) \quad \text{or}$$

$$I_m = c_m dV/dt + g_K(V - E_K) + g_{Na}(V - E_{Na}) + g_{Cl}(V - E_{Cl}) + I_{pump}$$

**10** In the steady state there is no net current flow and there is no capacitive current. This means that in the steady-state we have (ignoring the pumps for now)

$$0 = g_K(V - E_K) + g_{Na}(V - E_{Na}) + g_{Cl}(V - E_{Cl})$$

From this we can see that if we know the conductances of the various ions and the Nernst potentials for each ion, we can calculate  $V$

We get: 
$$V_m = \frac{g_K E_K + g_{Na} E_{Na} + g_{Cl} E_{Cl}}{g_K + g_{Na} + g_{Cl}}$$

and this allows us to compute the resting potential with the circuit model.

Notes:

1) Do not get confused by the fact that Cl is negatively charged. The equations are in terms of current which is defined as movement of positive charge. A positive (outward) Cl current is actually composed of inward flow of negatively charged Cl.

2) In these equations we could include a term for injected current. BY CONVENTION positive current injected into a neuron is defined as negative in the circuit model equations. So injecting 4 nA of current is expressed as -4 nA and will depolarize the cell.

$$I_{total} = I_c + I_K + I_{Na} + I_{Cl} + I_{pump} + I_{injected}$$

**11** Then solving for  $V_m$  in the steady-state, as we did above, gives

$$V_m = \frac{g_K E_K + g_{Na} E_{Na} + g_{Cl} E_{Cl} - I_{injected}}{g_K + g_{Na} + g_{Cl}} \quad \text{or} \quad V_m = \frac{g_K E_K + g_{Na} E_{Na} + g_{Cl} E_{Cl}}{g_K + g_{Na} + g_{Cl}} - \frac{I_{injected}}{g_K + g_{Na} + g_{Cl}}$$

and from this we can see how  $I_{injected} = -4 \text{ nA}$  would depolarize the cell.

BY CONVENTION (historical accident) (MEMORIZE)

Inward (depolarizing) current is negative (i.e. Na)

Outward (hyperpolarizing) current is positive (i.e. K)

3) The equation for  $V$  is really  $V=IR$  in disguise. Conductances in parallel sum and

$$\frac{1}{g_K + g_{Na} + g_{Cl}} = \frac{1}{\frac{1}{r_K} + \frac{1}{r_{Na}} + \frac{1}{r_{Cl}}} \quad \text{or} \quad \frac{1}{g_m} = \frac{1}{\frac{1}{r_m}} = r_m$$

**12** 4) Be careful in the interpretation of the circuit model. The membrane potential is the result of charge separation by the membrane and the ability of the membrane to store charge (its capacitance). If all channels were suddenly removed from the membrane,  $V_m$  would be the same since it would have the same charge imbalance. Membrane capacitance and charge separation underlie  $V_m$ ; channels provide a method of charging and discharging the membrane and hence changing  $V_m$ .

5) This circuit is very useful and forms the basis of modeling (my computational neuroscience course next quarter). It also allows synaptic currents to be incorporated easily  $I_{syn} = g_{syn}(V - E_{syn})$ .

6) Finally there is an equation relating permeability and conductance. FYI only

$$g_x = P_x F V' \frac{[X]_o - [X]_i \exp(V')}{(1 - \exp(V')(V_m - E_x)} \quad \text{where} \quad V' = \frac{V_m F}{RT}$$