Chapter 2

Electrical Signals of Nerve Cells

Overview

Nerve cells generate electrical signals that transmit information. Although neurons are not intrinsically good conductors of electricity, they have evolved elaborate mechanisms for generating these signals based on the flow of ions across their plasma membranes. Ordinarily, neurons generate a negative potential, called the resting membrane potential, that can be measured by recording the voltage between the inside and outside of nerve cells. The action potential transiently abolishes the negative resting potential and makes the transmembrane potential positive. Action potentials are propagated along the length of axons and are the fundamental signal that carries information from one place to another in the nervous system. Still other types of electrical signals are produced by the activation of synaptic contacts between neurons or by the actions of external forms of energy on sensory neurons. All of these electrical signals arise from ion fluxes brought about by nerve cell membranes being selectively permeable to different ions, and from the non-uniform distribution of these ions across the membrane.

Electrical Potentials across Nerve Cell Membranes

Neurons employ several different types of electrical signal to encode and transfer information. The best way to observe these signals is to use an intracellular microelectrode to measure the electrical potential across the neuronal plasma membrane. A typical microelectrode is a piece of glass tubing pulled to a very fine point (with an opening of less than 1 µm diameter) and filled with a good electrical conductor, such as a concentrated salt solution. This conductive core can then be connected to a voltmeter, such as an oscilloscope, to record the transmembrane voltage of the nerve cell.

The first type of electrical phenomenon can be observed as soon as a microelectrode is inserted through the membrane of the neuron. Upon entering the cell, the microelectrode reports a negative potential, indicating that neurons have a means of generating a constant voltage across their membranes when at rest. This voltage, called the **resting membrane potential**, depends on the type of neuron being examined, but it is always a fraction of a volt (typically –40 to –90 mV).

The electrical signals produced by neurons are caused by responses to stimuli, which then change the resting membrane potential. **Receptor potentials** are due to the activation of sensory neurons by external stimuli, such as light, sound, or heat. For example, touching the skin activates Pacinian corpuscles, receptor neurons that sense mechanical disturbances of the skin. These neurons respond to touch with a receptor potential that changes the resting potential for a fraction of a second (Figure 2.1A). These transient

Figure 2.1   Types of neuronal electrical signals. In all cases, microelectrodes are used to measure changes in the resting membrane potential during the indicated signals. (A) A brief touch causes a receptor potential in a Pacinian corpuscle in the skin. (B) Activation of a synaptic contact onto a hippocampal pyramidal neuron elicits a synaptic potential. (C) Stimulation of a spinal reflex produces an action potential in a spinal motor neuron.
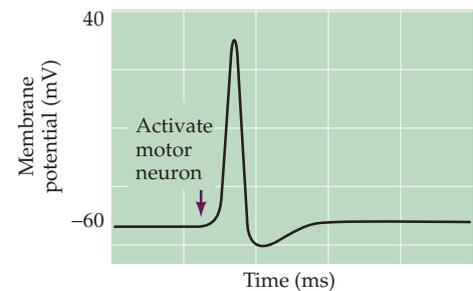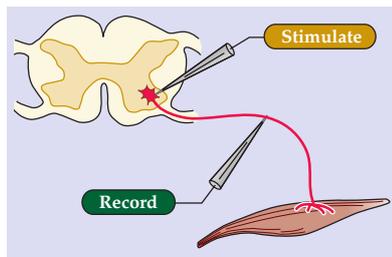
(A) Receptor potential
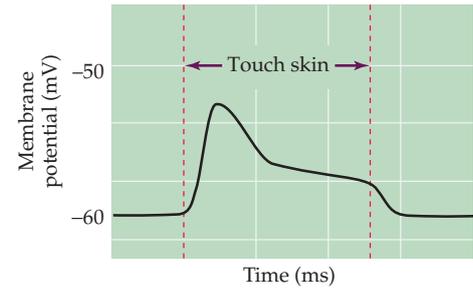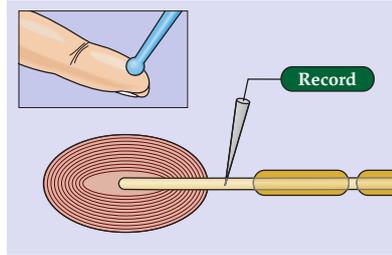
(B) Synaptic potential
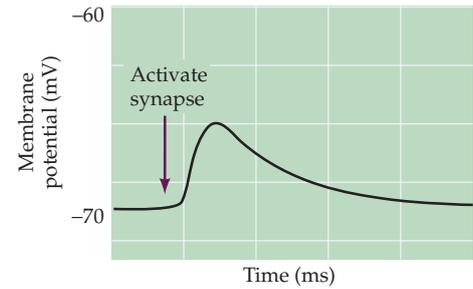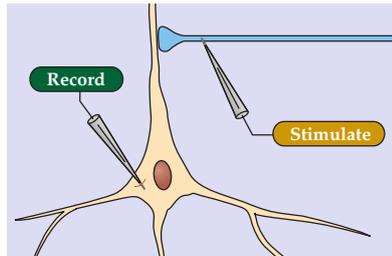
(C) Action potential

changes in potential are the first step in generating the sensation of vibrations (or "tickles") of the skin in the somatic sensory system (Chapter 8). Similar sorts of receptor potentials are observed in all other sensory neurons during transduction of sensory signals (Unit II).

Another type of electrical signal is associated with communication between neurons at synaptic contacts. Activation of these synapses generates **synaptic potentials**, which allow transmission of information from one neuron to another. An example of such a signal is shown in Figure 2.1B. In this case, activation of a synaptic terminal innervating a hippocampal pyramidal neuron causes a very brief change in the resting membrane potential in the pyramidal neuron. Synaptic potentials serve as the means of exchanging information in complex neural circuits in both the central and peripheral nervous systems (Chapter 5).

The use of electrical signals—as in sending electricity over wires to provide power or information—presents a series of problems in electrical engineering. A fundamental problem for neurons is that their axons, which can be quite long (remember that a spinal motor neuron can extend for a meter or more), are not good electrical conductors. Although neurons and wires

are both capable of passively conducting electricity, the electrical properties of neurons compare poorly to an ordinary wire. To compensate for this deficiency, neurons have evolved a "booster system" that allows them to conduct electrical signals over great distances despite their intrinsically poor electrical characteristics. The electrical signals produced by this booster system are called **action potentials** (which are also referred to as "spikes" or "impulses"). An example of an action potential recorded from the axon of a spinal motor neuron is shown in Figure 2.1C.

One way to elicit an action potential is to pass electrical current across the membrane of the neuron. In normal circumstances, this current would be generated by receptor potentials or by synaptic potentials. In the laboratory, however, electrical current suitable for initiating an action potential can be readily produced by inserting a second microelectrode into the same neuron and then connecting the electrode to a battery (Figure 2.2A). If the current delivered in this way makes the membrane potential more negative (**hyperpolarization**), nothing very dramatic happens. The membrane potential simply changes in proportion to the magnitude of the injected current (central part of Figure 2.2B). Such hyperpolarizing responses do not require any unique property of neurons and are therefore called passive electrical responses. A much more interesting phenomenon is seen if current of the opposite polarity is delivered, so that the membrane potential of the nerve cell becomes more positive than the resting potential (**depolarization**). In this case, at a certain level of membrane potential, called the **threshold potential**, an action potential occurs (see right side of Figure 2.2B).

The action potential, which is an active response generated by the neuron, is a brief (about 1 ms) change from negative to positive in the transmem-

Figure 2.2 Recording passive and active electrical signals in a nerve cell. (A) Two microelectrodes are inserted into a neuron; one of these measures membrane potential while the other injects current into the neuron. (B) Inserting the voltage-measuring microelectrode into the neuron reveals a negative potential, the resting membrane potential. Injecting current through the current-passing microelectrode alters the neuronal membrane potential. Hyperpolarizing current pulses produce only passive changes in the membrane potential. While small depolarizing currents also elict only passive responses, depolarizations that cause the membrane potential to meet or exceed threshold additionally evoke action potentials. Action potentials are active responses in the sense that they are generated by changes in the permeability of the neuronal membrane.
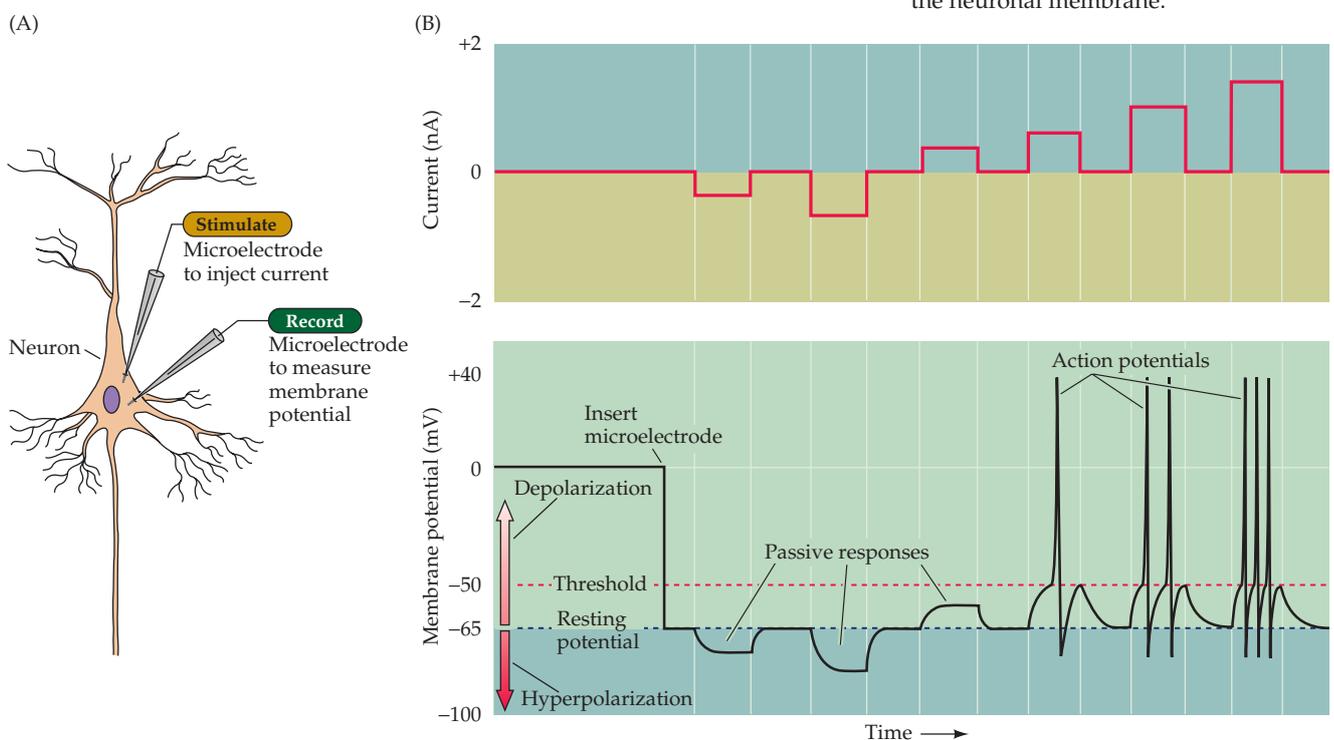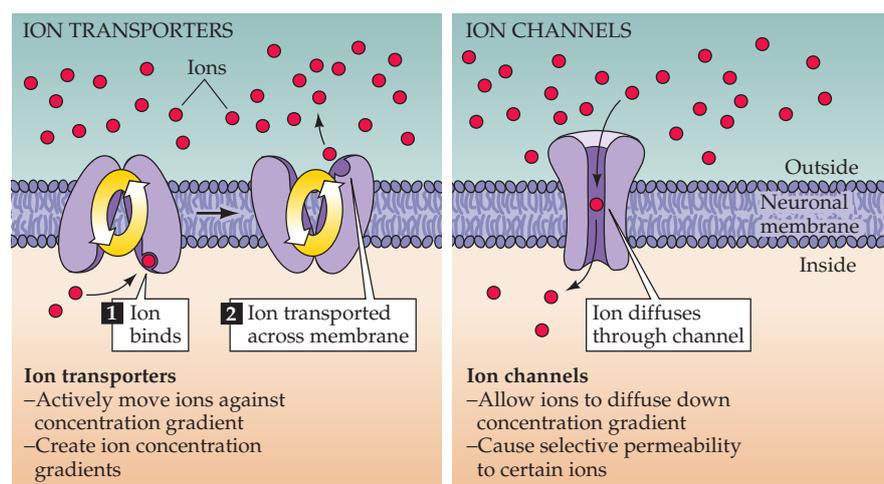
brane potential. Importantly, the amplitude of the action potential is independent of the magnitude of the current used to evoke it; that is, larger currents do not elicit larger action potentials. The action potentials of a given neuron are therefore said to be *all-or-none*, because they occur fully or not at all. If the amplitude or duration of the stimulus current is increased sufficiently, multiple action potentials occur, as can be seen in the responses to the three different current intensities shown in Figure 2.2B (right side). It follows, therefore, that the intensity of a stimulus is encoded in the frequency of action potentials rather than in their amplitude. This arrangement differs dramatically from receptor potentials, whose amplitudes are graded in proportion to the magnitude of the sensory stimulus, or synaptic potentials, whose amplitude varies according to the number of synapses activated and the previous amount of synaptic activity.

Because electrical signals are the basis of information transfer in the nervous system, it is essential to understand how these signals arise. Remarkably, all of the neuronal electrical signals described above are produced by similar mechanisms that rely upon the movement of ions across the neuronal membrane. The remainder of this chapter addresses the question of how nerve cells use ions to generate electrical potentials. Chapter 3 explores more specifically the means by which action potentials are produced and how these signals solve the problem of long-distance electrical conduction within nerve cells. Chapter 4 examines the properties of membrane molecules responsible for electrical signaling. Finally, Chapters 5–7 consider how electrical signals are transmitted from one nerve cell to another at synaptic contacts.

How Ionic Movements Produce Electrical Signals

Electrical potentials are generated across the membranes of neurons—and, indeed, all cells—because (1) there are *differences in the concentrations* of specific ions across nerve cell membranes, and (2) the membranes are *selectively permeable* to some of these ions. These two facts depend in turn on two different kinds of proteins in the cell membrane (Figure 2.3). The ion concentration gradients are established by proteins known as **active transporters**, which, as their name suggests, actively move ions into or out of cells against their concentration gradients. The selective permeability of membranes is

Figure 2.3 Ion transporters and ion channels are responsible for ionic movements across neuronal membranes. Transporters create ion concentration differences by actively transporting ions against their chemical gradients. Channels take advantage of these concentration gradients, allowing selected ions to move, via diffusion, down their chemical gradients.

ION TRANSPORTERS

Ions

1 Ion binds

2 Ion transported across membrane

Ion transporters
–Actively move ions against concentration gradient
–Create ion concentration gradients

ION CHANNELS

Outside
Neuronal membrane
Inside

Ion diffuses through channel

Ion channels
–Allow ions to diffuse down concentration gradient
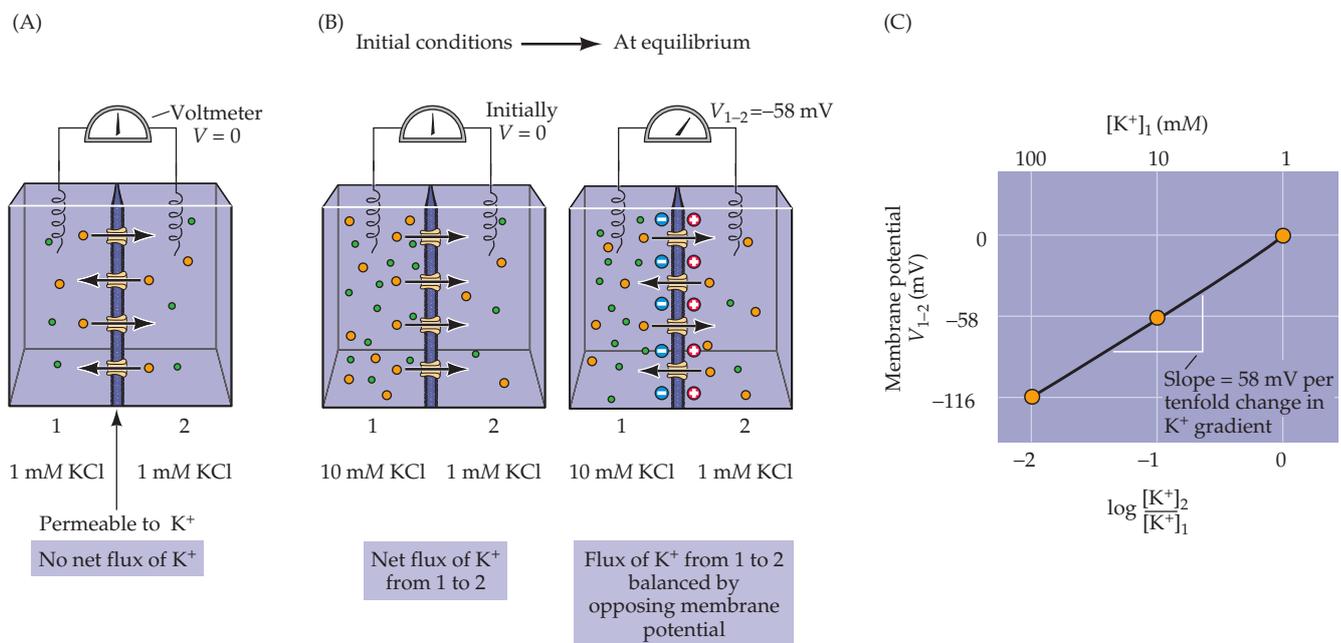–Cause selective permeability to certain ions

due largely to **ion channels**, proteins that allow only certain kinds of ions to cross the membrane in the direction of their concentration gradients. Thus, channels and transporters basically work against each other, and in so doing they generate the resting membrane potential, action potentials, and the synaptic potentials and receptor potentials that trigger action potentials. The structure and function of these channels and transporters are described in Chapter 4.

To appreciate the role of ion gradients and selective permeability in generating a membrane potential, consider a simple system in which an artificial membrane separates two compartments containing solutions of ions. In such a system, it is possible to determine the composition of the two solutions and, thereby, control the ion gradients across the membrane. For example, take the case of a membrane that is permeable only to potassium ions (K^+). If the concentration of K^+ on each side of this membrane is equal, then no electrical potential will be measured across it (Figure 2.4A). However, if the concentration of K^+ is not the same on the two sides, then an electrical potential will be generated. For instance, if the concentration of K^+ on one side of the membrane (compartment 1) is 10 times higher than the K^+ concentration on the other side (compartment 2), then the electrical potential of compartment 1 will be negative relative to compartment 2 (Figure 2.4B). This difference in electrical potential is generated because the potassium ions flow down their concentration gradient and take their electrical charge (one positive charge per ion) with them as they go. Because neuronal membranes contain pumps that accumulate K^+ in the cell cytoplasm, and because potassium-permeable channels in the plasma membrane allow a transmembrane flow of K^+, an analogous situation exists in living nerve cells. A continual resting efflux of K^+ is therefore responsible for the resting membrane potential.

In the hypothetical case just described, an equilibrium will quickly be reached. As K^+ moves from compartment 1 to compartment 2 (the initial conditions on the left of Figure 2.4B), a potential is generated that tends to impede further flow of K^+. This impediment results from the fact that the

Figure 2.4 Electrochemical equilibrium. (A) A membrane permeable only to K^+ (yellow spheres) separates compartments 1 and 2, which contain the indicated concentrations of KCl. (B) Increasing the KCl concentration in compartment 1 to 10 mM initially causes a small movement of K^+ into compartment 2 (initial conditions) until the electromotive force acting on K^+ balances the concentration gradient, and the net movement of K^+ becomes zero (at equilibrium). (C) The relationship between the transmembrane concentration gradient ($[K^+]_2/[K^+]_1$) and the membrane potential. As predicted by the Nernst equation, this relationship is linear when plotted on semi-logarithmic coordinates, with a slope of 58 mV per tenfold difference in the concentration gradient.

(A)

Voltmeter
$V = 0$

1   2

1 mM KCl | 1 mM KCl

Permeable to K^+

No net flux of K^+

(B)

Initial conditions ⟶ At equilibrium

Initially
$V = 0$

$V_{1-2} = -58$ mV

1   2   1   2

10 mM KCl   1 mM KCl   10 mM KCl   1 mM KCl

Net flux of K^+ from 1 to 2

Flux of K^+ from 1 to 2 balanced by opposing membrane potential

(C)

$[K^+]_1$ (mM)

100   10   1

Membrane potential V_{1-2} (mV)

0

−58

−116

Slope = 58 mV per tenfold change in K^+ gradient

−2   −1   0

$\log \dfrac{[K^+]_2}{[K^+]_1}$

potential gradient across the membrane tends to repel the positive potassium ions that would otherwise move across the membrane. Thus, as compartment 2 becomes positive relative to compartment 1, the increasing positivity makes compartment 2 less attractive to the positively charged K^+. The net movement (or flux) of K^+ will stop at the point (at equilibrium on the right of Figure 2.4B) where the potential change across the membrane (the relative positivity of compartment 2) exactly offsets the concentration gradient (the tenfold excess of K^+ in compartment 1). At this **electrochemical equilibrium**, there is an exact balance between two opposing forces: (1) the concentration gradient that causes K^+ to move from compartment 1 to compartment 2, taking along positive charge, and (2) an opposing electrical gradient that increasingly tends to stop K^+ from moving across the membrane (Figure 2.4B). The number of ions that needs to flow to generate this electrical potential is very small (approximately 10^{-12} moles of K^+ per cm^2 of membrane, or 10^{12} K^+ ions). This last fact is significant in two ways. First, it means that the concentrations of permeant ions on each side of the membrane remain essentially constant, even after the flow of ions has generated the potential. Second, the tiny fluxes of ions required to establish the membrane potential do not disrupt chemical electroneutrality because each ion has an oppositely charged counter-ion (chloride ions in the example shown in Figure 2.4) to maintain the neutrality of the solutions on each side of the membrane. The concentration of K^+ remains equal to the concentration of Cl^- in the solutions in compartments 1 and 2, meaning that the separation of charge that creates the potential difference is restricted to the immediate vicinity of the membrane.

The Forces That Create Membrane Potentials

The electrical potential generated across the membrane at electrochemical equilibrium, the **equilibrium potential**, can be predicted by a simple formula called the **Nernst equation**. This relationship is generally expressed as

$$E_X = \frac{RT}{zF} \ln \frac{[X]_2}{[X]_1}$$

where E_X is the equilibrium potential for any ion X, R is the gas constant, T is the absolute temperature (in degrees on the Kelvin scale), z is the valence (electrical charge) of the permeant ion, and F is the Faraday constant (the amount of electrical charge contained in one mole of a univalent ion). The brackets indicate the concentrations of ion X on each side of the membrane and the symbol ln indicates the natural logarithm of the concentration gradient. Because it is easier to perform calculations using base 10 logarithms and to perform experiments at room temperature, this relationship is usually simplified to

$$E_X = \frac{58}{z} \log \frac{[X]_2}{[X]_1}$$

where log indicates the base 10 logarithm of the concentration ratio. Thus, for the example in Figure 2.4B, the potential across the membrane at electrochemical equilibrium is

$$E_K = \frac{58}{z} \log \frac{[K]_2}{[K]_1} = 58 \log \frac{1}{10} = -58 \text{ mV}$$

The equilibrium potential is conventionally defined in terms of the potential difference between the reference compartment, side 2 in Figure 2.4, and the other side. This approach is also applied to biological systems. In this case,

the outside of the cell is the conventional reference point (defined as zero potential). Thus, when the concentration of K^+ is higher inside than out, an inside-negative potential is measured across the K^+-permeable neuronal membrane.

For a simple hypothetical system with only one permeant ion species, the Nernst equation allows the electrical potential across the membrane at equilibrium to be predicted exactly. For example, if the concentration of K^+ on side 1 is increased to 100 mM, the membrane potential will be –116 mV. More generally, if the membrane potential is plotted against the logarithm of the K^+ concentration gradient ($[K]_2/[K]_1$), the Nernst equation predicts a linear relationship with a slope of 58 mV (actually $58/z$) per tenfold change in the K^+ gradient (Figure 2.4C).

To reinforce and extend the concept of electrochemical equilibrium, consider some additional experiments on the influence of ionic species and ionic permeability that could be performed on the simple model system in Figure 2.4. What would happen to the electrical potential across the membrane (the potential of side 1 relative to side 2) if the potassium on side 2 were replaced with 10 mM sodium (Na^+) and the K^+ in compartment 1 were replaced by 1 mM Na^+? No potential would be generated, because no Na^+ could flow across the membrane (which was defined as being permeable only to K^+). However, if under these ionic conditions (10 times more Na^+ in compartment 2) the K^+-permeable membrane were to be magically replaced by a membrane permeable only to Na^+, a potential of +58 mV would be measured at equilibrium. If 10 mM calcium (Ca^{2+}) were present in compartment 2 and 1 mM Ca^{2+} in compartment 1, and a Ca^{2+}-selective membrane separated the two sides, what would happen to the membrane potential? A potential of +29 mV would develop, because the valence of calcium is +2. Finally, what would happen to the membrane potential if 10 mM Cl^- were present in compartment 1 and 1 mM Cl^- were present in compartment 2, with the two sides separated by a Cl^--permeable membrane? Because the valence of this anion is –1, the potential would again be +58 mV.

The balance of chemical and electrical forces at equilibrium means that the electrical potential can determine ionic fluxes across the membrane, just as the ionic gradient can determine the membrane potential. To examine the influence of membrane potential on ionic flux, imagine connecting a battery across the two sides of the membrane to control the electrical potential across the membrane without changing the distribution of ions on the two sides (Figure 2.5). As long as the battery is off, things will be just as in Figure 2.4, with the flow of K^+ from compartment 1 to compartment 2 causing a negative membrane potential (Figure 2.5A, left). However, if the battery is used to make compartment 1 initially more negative relative to compartment 2, there will be less K^+ flux, because the negative potential will tend to keep K^+ in compartment 1. How negative will side 1 need to be before there is no net flux of K^+? The answer is –58 mV, the voltage needed to counter the tenfold difference in K^+ concentrations on the two sides of the membrane (Figure 2.5A, center). If compartment 1 is initially made more negative than –58 mV, then K^+ will actually flow from compartment 2 into compartment 1, because the positive ions will be attracted to the more negative potential of compartment 1 (Figure 2.5A, right). This example demonstrates that both the direction and magnitude of ion flux depend on the membrane potential. Thus, in some circumstances the electrical potential can overcome an ionic concentration gradient.

The ability to alter ion flux experimentally by changing either the potential imposed on the membrane (Figure 2.5B) or the transmembrane concen-
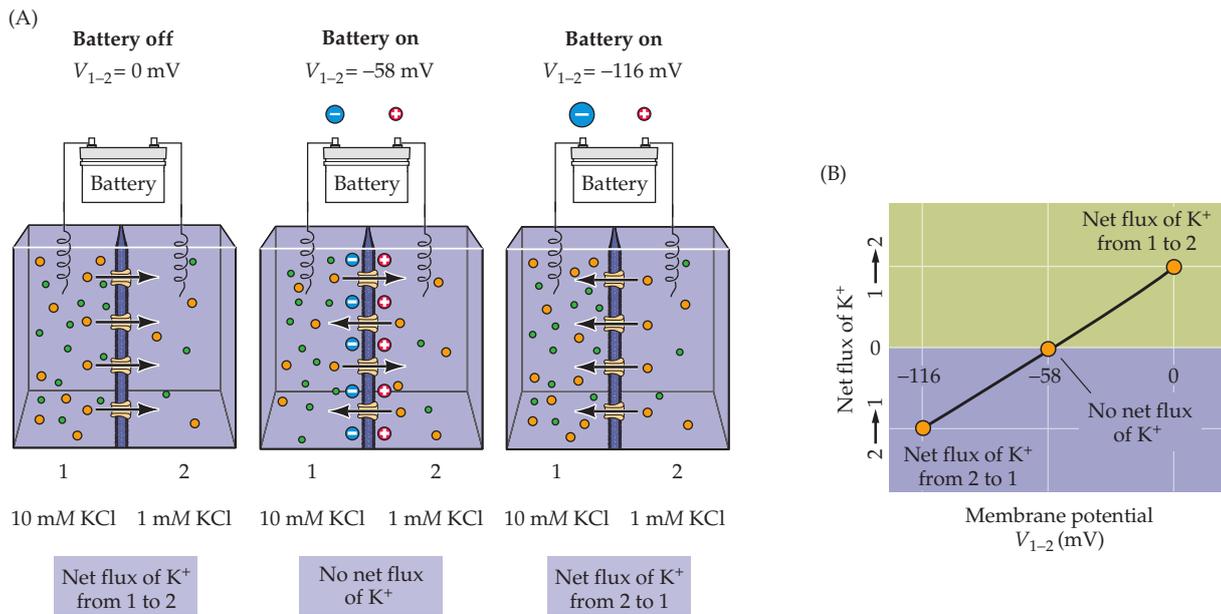
(A)

(B)

Figure 2.5 Membrane potential influences ion fluxes. (A) Connecting a battery across the K⁺-permeable membrane allows direct control of membrane potential. When the battery is turned off (left), K⁺ ions (yellow) flow simply according to their concentration gradient. Setting the initial membrane potential (V_{1-2}) at the equilibrium potential for K⁺ (center) yields no net flux of K⁺, while making the membrane potential more negative than the K⁺ equilibrium potential (right) causes K⁺ to flow against its concentration gradient. (B) Relationship between membrane potential and direction of K⁺ flux.

tration gradient for an ion (see Figure 2.4C) provides convenient tools for studying ion fluxes across the plasma membranes of neurons, as will be evident in many of the experiments described in the following chapters.

Electrochemical Equilibrium in an Environment with More Than One Permeant Ion

Now consider a somewhat more complex situation in which Na⁺ and K⁺ are unequally distributed across the membrane, as in Figure 2.6A. What would happen if 10 mM K⁺ and 1 mM Na⁺ were present in compartment 1, and 1 mM K⁺ and 10 mM Na⁺ in compartment 2? If the membrane were permeable only to K⁺, the membrane potential would be –58 mV; if the membrane were permeable only to Na⁺, the potential would be +58 mV. But what would the potential be if the membrane were permeable to both K⁺ and Na⁺? In this case, the potential would depend on the relative permeability of the membrane to K⁺ and Na⁺. If it were more permeable to K⁺, the potential would approach –58 mV, and if it were more permeable to Na⁺, the potential would be closer to +58 mV. Because there is no permeability term in the Nernst equation, which only considers the simple case of a single permeant ion species, a more elaborate equation is needed that takes into account both the concentration gradients of the permeant ions and the relative permeability of the membrane to each permeant species.

Such an equation was developed by David Goldman in 1943. For the case most relevant to neurons, in which K⁺, Na⁺, and Cl⁻ are the primary permeant ions, the **Goldman equation** is written

$$V = 58 \log \frac{P_K[K]_2 + P_{Na}[Na]_2 + P_{Cl}[Cl]_1}{P_K[K]_1 + P_{Na}[Na]_1 + P_{Cl}[Cl]_2}$$

where V is the voltage across the membrane (again, compartment 1 relative to the reference compartment 2) and P indicates the permeability of the

(A)

Voltmeter

1	2
10 mM KCl	1 mM KCl
1 mM NaCl	10 mM NaCl

Variable permeability
to Na$^+$ and K$^+$

(B)

Na$^+$ permeable
K$^+$ permeable

E_{Na}

$P_{Na} \gg P_K$

$\uparrow P_{Na}$ $\downarrow P_{Na}$

0

Resting
potential

Action
potential

Repolarization

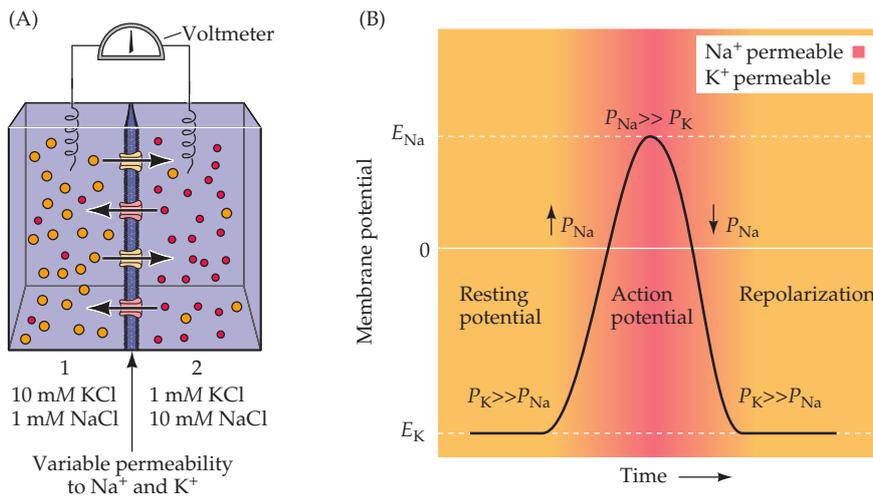$P_K \gg P_{Na}$ $P_K \gg P_{Na}$

E_K

Time

Figure 2.6 Resting and action potentials entail permeabilities to different ions. (A) Hypothetical situation in which a membrane variably permeable to Na$^+$ (red) and K$^+$ (yellow) separates two compartments that contain both ions. For simplicity, Cl$^-$ ions are not shown in the diagram. (B) Schematic representation of the membrane ionic permeabilities associated with resting and action potentials. At rest, neuronal membranes are more permeable to K$^+$ (yellow) than to Na$^+$ (red); accordingly, the resting membrane potential is negative and approaches the equilibrium potential for K$^+$, E_K. During an action potential, the membrane becomes very permeable to Na$^+$ (red); thus the membrane potential becomes positive and approaches the equilibrium potential for Na$^+$, E_{Na}. The rise in Na$^+$ permeability is transient, however, so that the membrane again becomes primarily permeable to K$^+$ (yellow), causing the potential to return to its negative resting value. Notice that at the equilibrium potential for a given ion, there is no net flux of that ion across the membrane.

membrane to each ion of interest. The Goldman equation is thus an extended version of the Nernst equation that takes into account the relative permeabilities of each of the ions involved. The relationship between the two equations becomes obvious in the situation where the membrane is permeable only to one ion, say, K$^+$; in this case, the Goldman expression collapses back to the simpler Nernst equation. In this context, it is important to note that the valence factor (z) in the Nernst equation has been eliminated; this is why the concentrations of negatively charged chloride ions, Cl$^-$, have been inverted relative to the concentrations of the positively charged ions [remember that $-\log(A/B) = \log(B/A)$].

If the membrane in Figure 2.6A is permeable to K$^+$ and Na$^+$ only, the terms involving Cl$^-$ drop out because P_{Cl} is 0. In this case, solution of the Goldman equation yields a potential of –58 mV when only K$^+$ is permeant, +58 mV when only Na$^+$ is permeant, and some intermediate value if both ions are permeant. For example, if K$^+$ and Na$^+$ were equally permeant, then the potential would be 0 mV.

With respect to neural signaling, it is particularly pertinent to ask what would happen if the membrane started out being permeable to K$^+$, and then temporarily switched to become most permeable to Na$^+$. In this circumstance, the membrane potential would start out at a negative level, become positive while the Na$^+$ permeability remained high, and then fall back to a negative level as the Na$^+$ permeability decreased again. As it turns out, this last case essentially describes what goes on in a neuron during the generation of an action potential. In the resting state, P_K of the neuronal plasma membrane is much higher than P_{Na}; since, as a result of the action of ion transporters, there is always more K$^+$ inside the cell than outside (Table 2.1), the resting potential is negative (Figure 2.6B). As the membrane potential is depolarized (by synaptic action, for example), P_{Na} increases. The transient increase in Na$^+$ permeability causes the membrane potential to become even more positive (red region in Figure 2.6B), because Na$^+$ rushes in (there is much more Na$^+$ outside a neuron than inside, again as a result of ion pumps). Because of this positive feedback loop, an action potential occurs. The rise in Na$^+$ permeability during the action potential is transient, however; as the membrane permeability to K$^+$ is restored, the membrane potential quickly returns to its resting level.

TABLE 2.1
Extracellular and Intracellular Ion Concentrations

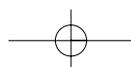| Ion	Concentration (*m*M)	
	Intracellular	*Extracellular*
Squid neuron		
Potassium (K⁺)	400	20
Sodium (Na⁺)	50	440
Chloride (Cl⁻)	40–150	560
Calcium (Ca²⁺)	0.0001	10
Mammalian neuron		
Potassium (K⁺)	140	5
Sodium (Na⁺)	5–15	145
Chloride (Cl⁻)	4–30	110
Calcium (Ca²⁺)	0.0001	1–2

Armed with an appreciation of these simple electrochemical principles, it will be much easier to understand the following, more detailed account of how neurons generate resting and action potentials.

The Ionic Basis of the Resting Membrane Potential

The action of ion transporters creates substantial transmembrane gradients for most ions. Table 2.1 summarizes the ion concentrations measured directly in an exceptionally large nerve cell found in the nervous system of the squid (Box A). Such measurements are the basis for stating that there is much more K⁺ inside the neuron than out, and much more Na⁺ outside than in. Similar concentration gradients occur in the neurons of most animals, including humans. However, because the ionic strength of mammalian blood is lower than that of sea-dwelling animals such as squid, in mammals the concentrations of each ion are several times lower. These transporter-dependent concentration gradients are, indirectly, the source of the resting neuronal membrane potential and the action potential.

Once the ion concentration gradients across various neuronal membranes are known, the Nernst equation can be used to calculate the equilibrium potential for K⁺ and other major ions. Since the resting membrane potential of the squid neuron is approximately –65 mV, K⁺ is the ion that is closest to being in electrochemical equilibrium when the cell is at rest. This fact implies that the resting membrane is more permeable to K⁺ than to the other ions listed in Table 2.1, and that this permeability is the source of resting potentials.

It is possible to test this guess, as Alan Hodgkin and Bernard Katz did in 1949, by asking what happens to the resting membrane potential if the concentration of K⁺ outside the neuron is altered. If the resting membrane were permeable only to K⁺, then the Goldman equation (or even the simpler Nernst equation) predicts that the membrane potential will vary in proportion to the logarithm of the K⁺ concentration gradient across the membrane. Assuming that the internal K⁺ concentration is unchanged during the experiment, a plot of membrane potential against the logarithm of the external K⁺ concentration should yield a straight line with a slope of 58 mV per tenfold change in external K⁺ concentration at room temperature (see Figure 2.4C). (The slope becomes about 61 mV at mammalian body temperatures.)

Box A
The Remarkable Giant Nerve Cells of Squid

Many of the initial insights into how ion concentration gradients and changes in membrane permeability produce electrical signals came from experiments performed on the extraordinarily large nerve cells of the squid. The axons of these nerve cells can be up to 1 mm in diameter—100 to 1000 times larger than mammalian axons. Thus, squid axons are large enough to allow experiments that would be impossible on most other nerve cells. For example, it is not difficult to insert simple wire electrodes inside these giant axons and make reliable electrical measurements. The relative ease of this approach yielded the first intracellular recordings of action potentials from nerve cells and, as discussed in the next chapter, the first experimental measure-

ments of the ion currents that produce action potentials. It also is practical to extrude the cytoplasm from giant axons and measure its ionic composition (see Table 2.1). In addition, some giant nerve cells form synaptic contacts with other giant nerve cells, producing very large synapses that have been extraordinarily valuable in understanding the fundamental mechanisms of synaptic transmission (see Chapter 5).

Giant neurons evidently evolved in squid because they enhanced survival. These neurons participate in a simple neural circuit that activates the contraction of the mantle muscle, producing a jet propulsion effect that allows the squid to move away from predators at a remarkably fast speed. As discussed in
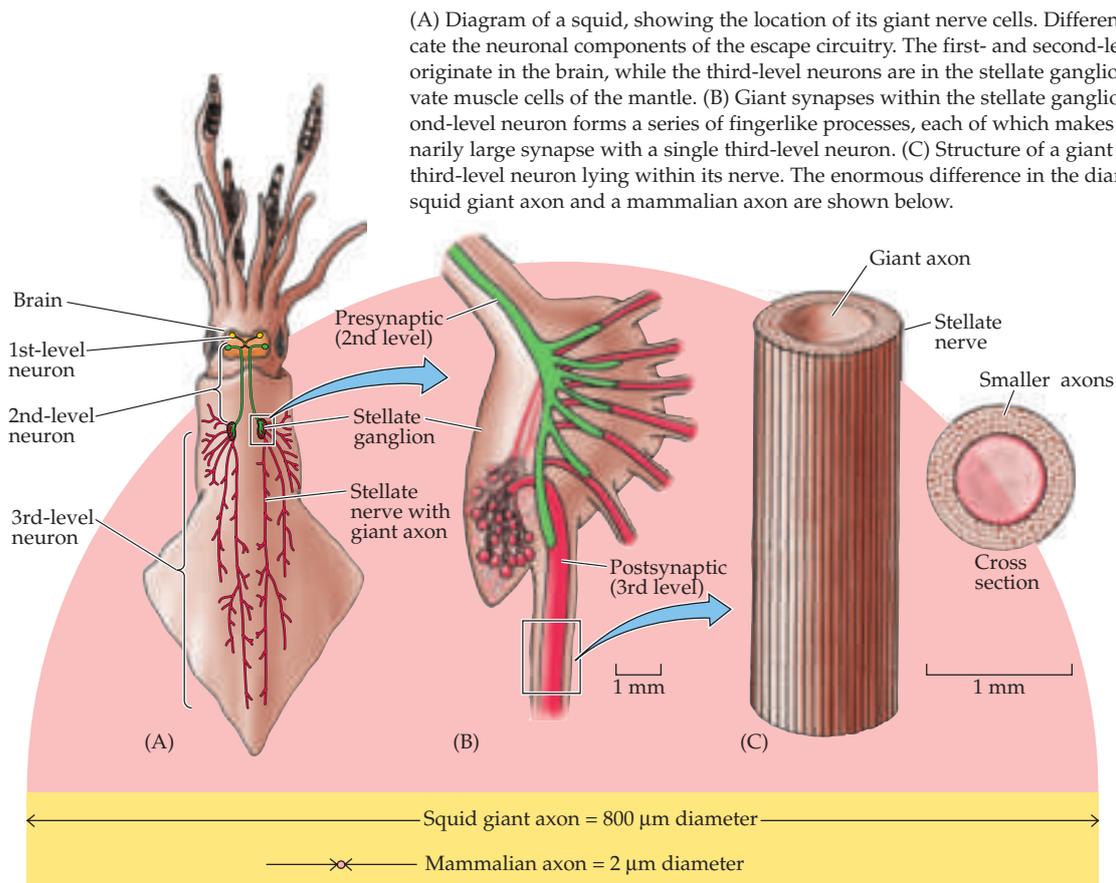
Chapter 3, larger axonal diameter allows faster conduction of action potentials. Thus, presumably these huge nerve cells help squid escape more successfully from their numerous enemies.

Today—nearly 70 years after their discovery by John Z. Young at University College London—the giant nerve cells of squid remain useful experimental systems for probing basic neuronal functions.

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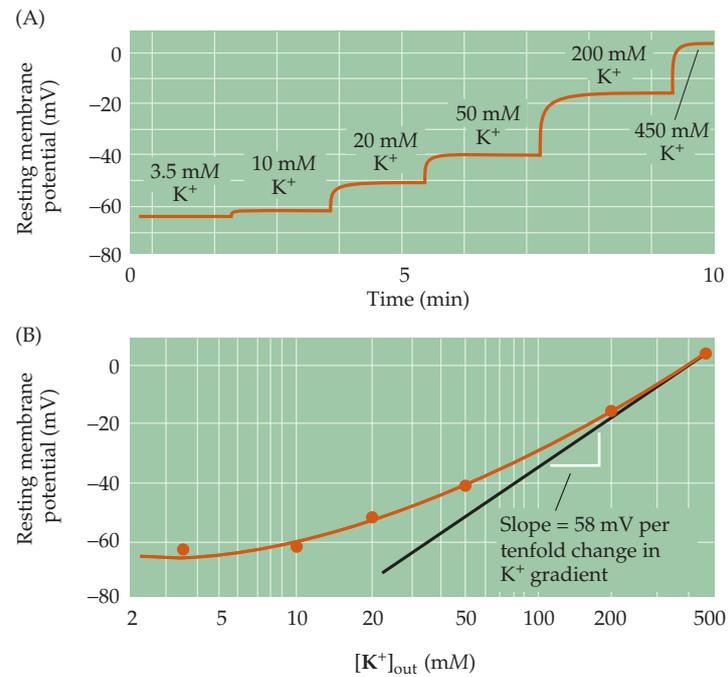
(A) Diagram of a squid, showing the location of its giant nerve cells. Different colors indicate the neuronal components of the escape circuitry. The first- and second-level neurons originate in the brain, while the third-level neurons are in the stellate ganglion and innervate muscle cells of the mantle. (B) Giant synapses within the stellate ganglion. The second-level neuron forms a series of fingerlike processes, each of which makes an extraordinarily large synapse with a single third-level neuron. (C) Structure of a giant axon of a third-level neuron lying within its nerve. The enormous difference in the diameters of a squid giant axon and a mammalian axon are shown below.

Brain

1st-level neuron

2nd-level neuron

3rd-level neuron

(A)

Presynaptic (2nd level)

Stellate ganglion

Stellate nerve with giant axon

Postsynaptic (3rd level)

1 mm

(B)

Giant axon

Stellate nerve

Smaller axons

Cross section

1 mm

(C)

Squid giant axon = 800 μm diameter

Mammalian axon = 2 μm diameter

Figure 2.7   Experimental evidence that the resting membrane potential of a squid giant axon is determined by the K⁺ concentration gradient across the membrane. (A) Increasing the external K⁺ concentration makes the resting membrane potential more positive. (B) Relationship between resting membrane potential and external K⁺ concentration, plotted on a semi-logarithmic scale. The straight line represents a slope of 58 mV per tenfold change in concentration, as given by the Nernst equation. (After Hodgkin and Katz, 1949.)

When Hodgkin and Katz carried out this experiment on a living squid neuron, they found that the resting membrane potential did indeed change when the external K⁺ concentration was modified, becoming less negative as external K⁺ concentration was raised (Figure 2.7A). When the external K⁺ concentration was raised high enough to equal the concentration of K⁺ inside the neuron, thus making the K⁺ equilibrium potential 0 mV, the resting membrane potential was also approximately 0 mV. In short, the resting membrane potential varied as predicted with the logarithm of the K⁺ concentration, with a slope that approached 58 mV per tenfold change in K⁺ concentration (Figure 2.7B). The value obtained was not exactly 58 mV because other ions, such as Cl⁻ and Na⁺, are also slightly permeable, and thus influence the resting potential to a small degree. The contribution of these other ions is particularly evident at low external K⁺ levels, again as predicted by the Goldman equation. In general, however, manipulation of the external concentrations of these other ions has only a small effect, emphasizing that K⁺ permeability is indeed the primary source of the resting membrane potential.

In summary, Hodgkin and Katz showed that the inside-negative resting potential arises because (1) the membrane of the resting neuron is more permeable to K⁺ than to any of the other ions present, and (2) there is more K⁺ inside the neuron than outside. The selective permeability to K⁺ is caused by K⁺-permeable membrane channels that are open in resting neurons, and the

large K$^+$ concentration gradient is, as noted, produced by membrane transporters that selectively accumulate K$^+$ within neurons. Many subsequent studies have confirmed the general validity of these principles.

The Ionic Basis of Action Potentials

What causes the membrane potential of a neuron to depolarize during an action potential? Although a general answer to this question has been given (increased permeability to Na$^+$), it is well worth examining some of the experimental support for this concept. Given the data presented in Table 2.1, one can use the Nernst equation to calculate that the equilibrium potential for Na$^+$ (E_{Na}) in neurons, and indeed in most cells, is positive. Thus, if the membrane were to become highly permeable to Na$^+$, the membrane potential would approach E_{Na}. Based on these considerations, Hodgkin and Katz hypothesized that the action potential arises because the neuronal membrane becomes temporarily permeable to Na$^+$.

Taking advantage of the same style of ion substitution experiment they used to assess the resting potential, Hodgkin and Katz tested the role of Na$^+$ in generating the action potential by asking what happens to the action potential when Na$^+$ is removed from the external medium. They found that lowering the external Na$^+$ concentration reduces both the rate of rise of the action potential and its peak amplitude (Figure 2.8A–C). Indeed, when they examined this Na$^+$ dependence quantitatively, they found a more-or-less linear relationship between the amplitude of the action potential and the logarithm of the external Na$^+$ concentration (Figure 2.8D). The slope of this rela-
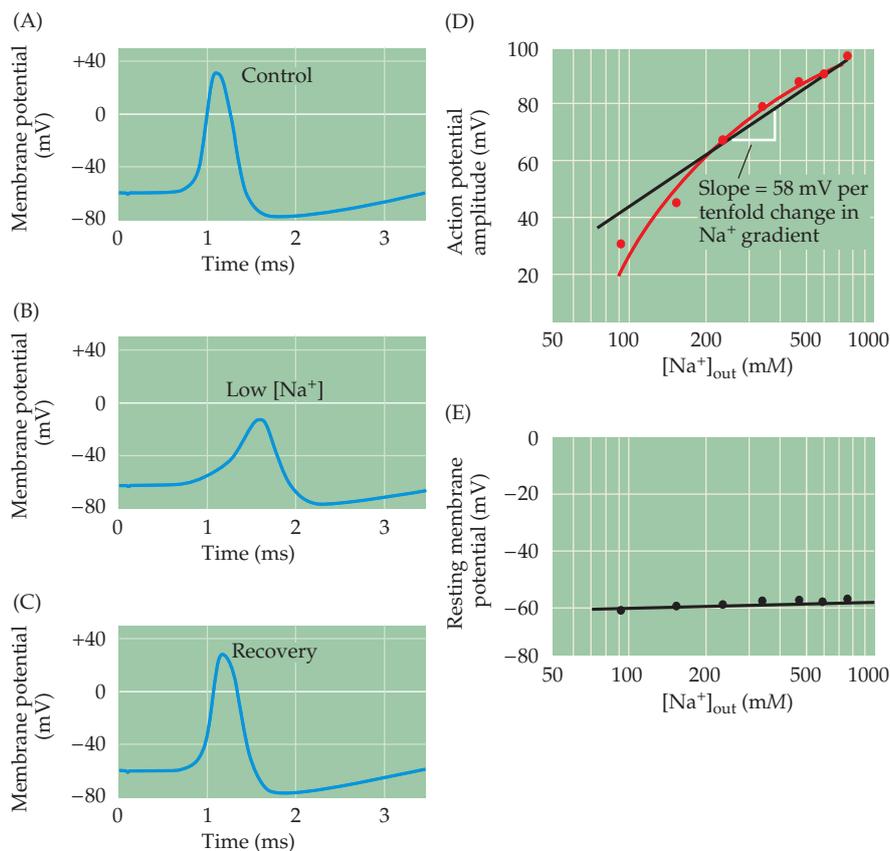

Figure 2.8 The role of sodium in the generation of an action potential in a squid giant axon. (A) An action potential evoked with the normal ion concentrations inside and outside the cell. (B) The amplitude and rate of rise of the action potential diminish when external sodium concentration is reduced to one-third of normal, but (C) recover when the Na$^+$ is replaced. (D) While the amplitude of the action potential is quite sensitive to the external concentration of Na$^+$, the resting membrane potential (E) is little affected by changing the concentration of this ion. (After Hodgkin and Katz, 1949.)

Box B
Action Potential Form and Nomenclature

The action potential of the squid giant axon has a characteristic shape, or waveform, with a number of different phases (Figure A). During the rising phase, the membrane potential rapidly depolarizes. In fact, action potentials cause the membrane potential to depolarize so much that the membrane potential transiently becomes positive with respect to the external medium, producing an overshoot. The overshoot of the action potential gives way to a falling phase in which the membrane potential rapidly repolarizes. Repolarization takes the membrane potential to levels even more negative than the resting membrane potential for a short time; this brief period of hyperpolarization is called the undershoot.

Although the waveform of the squid action potential is typical, the details of the action potential form vary widely from neuron to neuron in different animals. In myelinated axons of vertebrate motor neurons (Figure B), the action potential is virtually indistinguishable from that of the squid axon. However, the action potential recorded in the cell body of this same motor neuron (Figure C) looks rather different. Thus, the action potential waveform can vary even within the same neuron. More complex action potentials are seen in other central neurons. For example, action potentials recorded from the cell bodies of neurons in the mammalian inferior olive (a region of the brainstem involved in motor control) last tens of milliseconds (Figure D). These action potentials exhibit a pronounced plateau during their falling phase, and their undershoot lasts even longer than that of the motor neuron. One of the most dramatic types of action potentials occurs in the cell bodies of cerebellar Purkinje neurons (Figure E). These potentials have several complex phases that result from the summation of multiple, discrete action potentials.

The variety of action potential waveforms could mean that each type of neuron has a different mechanism of action potential production. Fortunately, however, these diverse waveforms all result from relatively minor variations in the scheme used by the squid giant axon. For example, plateaus in the repolarization phase result from the presence of ion channels that are permeable to Ca^{2+}, and long-lasting undershoots result from the presence of additional types of membrane K^+ channels. The complex action potential of the Purkinje cell results from these extra features plus the fact that different types of action potentials are generated in various parts of the Purkinje neuron—cell body, dendrites, and axons—and are summed together in recordings from the cell body. Thus, the lessons learned from the squid axon are applicable to, and indeed essential for, understanding action potential generation in all neurons.

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(A) The phases of an action potential of the squid giant axon. (B) Action potential recorded from a myelinated axon of a frog motor neuron. (C) Action potential recorded from the cell body of a frog motor neuron. The action potential is smaller and the undershoot prolonged in comparison to the action potential recorded from the axon of this same neuron (B). (D) Action potential recorded from the cell body of a neuron from the inferior olive of a guinea pig. This action potential has a pronounced plateau during its falling phase. (E) Action potential recorded from the cell body of a Purkinje neuron in the cerebellum of a guinea pig. (A after Hodgkin and Huxley, 1939; B after Dodge and Frankenhaeuser, 1958; C after Barrett and Barrett, 1976; D after Llinás and Yarom, 1981; E after Llinás and Sugimori, 1980.)
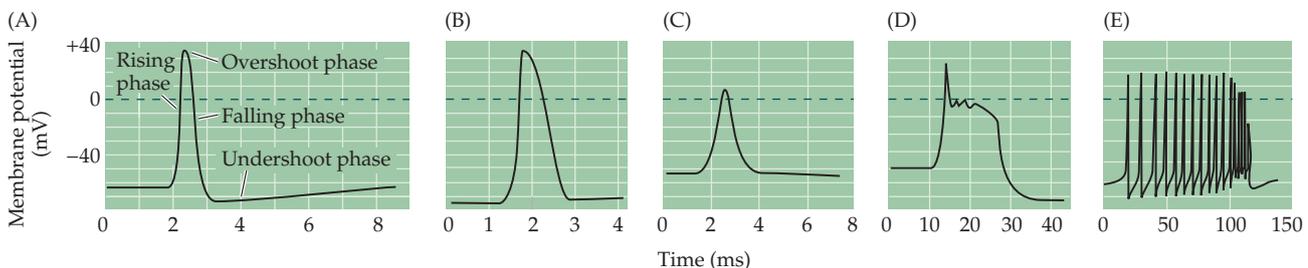
tionship approached a value of 58 mV per tenfold change in Na^+ concentration, as expected for a membrane selectively permeable to Na^+. In contrast, lowering Na^+ concentration had very little effect on the resting membrane potential (Figure 2.8E). Thus, while the resting neuronal membrane is only slightly permeable to Na^+, the membrane becomes extraordinarily permeable to Na^+ during the **rising phase** and **overshoot phase** of the action potential (see Box B for an explanation of action potential nomenclature). This temporary increase in Na^+ permeability results from the opening of Na^+-selective channels that are essentially closed in the resting state. Membrane pumps maintain a large electrochemical gradient for Na^+, which is in much higher concentration outside the neuron than inside. When the Na^+ channels open, Na^+ flows into the neuron, causing the membrane potential to depolarize and approach E_{Na}.

The time that the membrane potential lingers near E_{Na} (about +58 mV) during the overshoot phase of an action potential is brief because the increased membrane permeability to Na^+ itself is short-lived. The membrane potential rapidly repolarizes to resting levels and is actually followed by a transient **undershoot**. As will be described in Chapter 3, these latter events in the action potential are due to an inactivation of the Na^+ permeability and an increase in the K^+ permeability of the membrane. During the undershoot, the membrane potential is transiently hyperpolarized because K^+ permeability becomes even greater than it is at rest. The action potential ends when this phase of enhanced K^+ permeability subsides, and the membrane potential thus returns to its normal resting level.

The ion substitution experiments carried out by Hodgkin and Katz provided convincing evidence that the resting membrane potential results from a high resting membrane permeability to K^+, and that depolarization during an action potential results from a transient rise in membrane Na^+ permeability. Although these experiments identified the ions that flow during an action potential, they did not establish *how* the neuronal membrane is able to change its ionic permeability to generate the action potential, or what mechanisms trigger this critical change. The next chapter addresses these issues, documenting the surprising conclusion that the neuronal membrane potential itself affects membrane permeability.

Summary

Nerve cells generate electrical signals to convey information over substantial distances and to transmit it to other cells by means of synaptic connections. These signals ultimately depend on changes in the resting electrical potential across the neuronal membrane. A resting potential occurs because nerve cell membranes are permeable to one or more ion species subject to an electrochemical gradient. More specifically, a negative membrane potential at rest results from a net efflux of K^+ across neuronal membranes that are predominantly permeable to K^+. In contrast, an action potential occurs when a transient rise in Na^+ permeability allows a net flow of Na^+ in the opposite direction across the membrane that is now predominantly permeable to Na^+. The brief rise in membrane Na^+ permeability is followed by a secondary, transient rise in membrane K^+ permeability that repolarizes the neuronal membrane and produces a brief undershoot of the action potential. As a result of these processes, the membrane is depolarized in an all-or-none fashion during an action potential. When these active permeability changes subside, the membrane potential returns to its resting level because of the high resting membrane permeability to K^+.

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