CHAPTER **TWELVE** **12**

Transport Across Cell Membranes

To survive and grow, cells must be able to exchange molecules with their environment. They must import nutrients such as sugars and amino acids and eliminate metabolic waste products. They must also regulate the concentrations of a variety of inorganic ions in their cytosol and organelles. A few molecules, such as CO_2 and O_2, can simply diffuse across the lipid bilayer of the plasma membrane. But the vast majority cannot. Instead, their movement depends on specialized **membrane transport proteins** that span the lipid bilayer, providing private passageways across the membrane for select substances (**Figure 12–1**).

In this chapter, we consider how cell membranes control the traffic of inorganic ions and small, water-soluble molecules into and out of the cell and its membrane-enclosed organelles. Cells can also selectively transfer large macromolecules such as proteins across their membranes, but this transport requires more elaborate machinery and is discussed in Chapter 15.

We begin by outlining some of the general principles that guide the passage of ions and small molecules through cell membranes. We then examine, in turn, the two main classes of membrane proteins that mediate this transfer: transporters and channels. *Transporters* shift small organic molecules or inorganic ions from one side of the membrane to the other by changing shape. *Channels*, in contrast, form tiny hydrophilic pores across the membrane through which substances can pass by diffusion. Most channels only permit passage of ions and are therefore called *ion channels*. Because these ions are electrically charged, their movements can create a powerful electric force—or voltage—across the membrane. In the final part of the chapter, we discuss how these voltage differences enable nerve cells to communicate—and, ultimately, to shape how we behave.

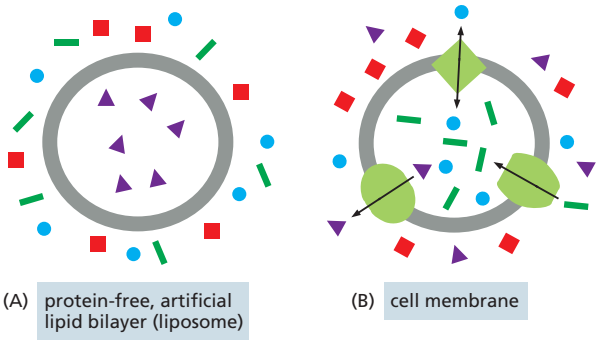


Figure 12–1 Cell membranes contain specialized membrane transport proteins that facilitate the passage of selected small, water-soluble molecules. (A) Protein-free, artificial lipid bilayers such as liposomes (see Figure 11–13) are impermeable to most water-soluble molecules. (B) Cell membranes, by contrast, contain membrane transport proteins (*light green*), each of which transfers a particular substance across the membrane. This selective transport can facilitate the passive diffusion of specific molecules or ions across the membrane (*blue* circles), as well as the active pumping of specific substances either out of (*purple* triangles) or into (*green* bars) the cell. For other molecules, the membrane is impermeable (*red* squares). The combined action of different membrane transport proteins allows a specific set of solutes to build up inside a membrane-enclosed compartment, such as the cytosol or an organelle.

PRINCIPLES OF TRANSMEMBRANE TRANSPORT

As we saw in Chapter 11, the hydrophobic interior of the lipid bilayer creates a barrier to the passage of most hydrophilic molecules, including all ions. These molecules are as reluctant to enter a fatty environment as hydrophobic molecules are reluctant to interact with water. But cells and organelles must allow the passage of many hydrophilic, water-soluble molecules, such as inorganic ions, sugars, amino acids, nucleotides, and other cell metabolites. These molecules cross lipid bilayers far too slowly by *simple diffusion*, so their passage across cell membranes must be accelerated by specialized membrane transport proteins—a process called *facilitated transport*. In this section, we review the basic principles of such facilitated transmembrane transport and introduce the various types of membrane transport proteins that mediate this movement. We also discuss why the transport of inorganic ions, in particular, is of such fundamental importance for all cells.

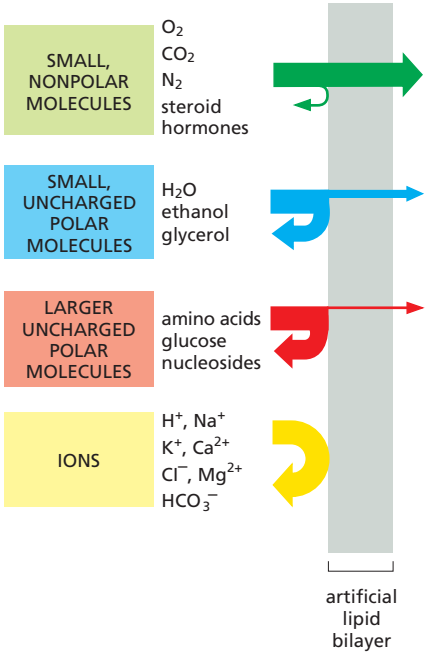


Figure 12–2 The rate at which a solute crosses a protein-free, artificial lipid bilayer by simple diffusion depends on its size and solubility. Many of the organic molecules that a cell uses as nutrients (*red*) are too large and polar to pass efficiently through an artificial lipid bilayer that does not contain the appropriate membrane transport proteins.

Lipid Bilayers Are Impermeable to Ions and Most Uncharged Polar Molecules

Given enough time, virtually any molecule will diffuse across a lipid bilayer. The rate at which it diffuses, however, varies enormously depending on the size of the molecule and its solubility properties. In general, the smaller the molecule and the more hydrophobic, or nonpolar, it is, the more rapidly it will diffuse across the lipid bilayer.

Of course, many of the molecules that are of interest to cells are polar and water-soluble. These *solutes*—substances that, in this case, are dissolved in water—are unable to cross the lipid bilayer without the aid of membrane transport proteins. The relative ease with which a variety of solutes can cross a lipid bilayer that lacks membrane transport proteins is shown in **Figure 12–2**.

1. *Small, nonpolar molecules*, such as molecular oxygen (O_2, molecular mass 32 daltons) and carbon dioxide (CO_2, 44 daltons), dissolve readily in lipid bilayers and therefore diffuse rapidly across them; indeed, cells depend on this permeability to gases for the *cell respiration* processes discussed in Chapter 14.
2. *Uncharged polar molecules* (those with an uneven distribution of electric charge) also diffuse readily across a bilayer, but only if they are small enough. Water (H_2O, 18 daltons) and ethanol (46 daltons), for example, cross at a measurable rate, whereas glycerol (92 daltons) crosses less rapidly. Larger uncharged polar molecules, such as glucose (180 daltons), cross hardly at all.
3. In contrast, lipid bilayers are highly impermeable to all charged substances, including all inorganic ions, no matter how small. The

charges on these solutes, and their strong electrical attraction to water molecules, inhibit their entry into the inner, hydrocarbon phase of the bilayer. Thus protein-free lipid bilayers are a billion (10^9) times more permeable to water, which is polar but uncharged, than they are to even small ions such as Na^+ or K^+.

The Ion Concentrations Inside a Cell Are Very Different from Those Outside

Because lipid bilayers are impermeable to inorganic ions, living cells are able to maintain internal ion concentrations that are very different from the concentrations of ions in the medium that surrounds them. These differences in ion concentration are crucial for a cell's survival and function. Among the most important inorganic ions for cells are Na^+, K^+, Ca^{2+}, Cl^-, and H^+ (protons). The movement of these ions across cell membranes plays an essential part in many biological processes, but is perhaps most striking in the production of ATP by all cells (discussed in Chapter 14) and in the communication of nerve cells (discussed later in this chapter).

Na^+ is the most plentiful positively charged ion (cation) outside the cell, whereas K^+ is the most abundant inside (**Table 12–1**). For a cell to avoid being torn apart by electrical forces, the quantity of positive charge inside the cell must be balanced by an almost exactly equal quantity of negative charge, and the same is true for the charge in the surrounding fluid. The high concentration of Na^+ outside the cell is electrically balanced chiefly by extracellular Cl^-, whereas the high concentration of K^+ inside is balanced by a variety of negatively charged inorganic and organic ions (anions), including nucleic acids, proteins, and many cell metabolites (see Table 12–1).

Differences in the Concentration of Inorganic Ions Across a Cell Membrane Create a Membrane Potential

Although the electrical charges inside and outside the cell are generally kept in balance, tiny excesses of positive or negative charge, concentrated in the neighborhood of the plasma membrane, do occur. Such electrical imbalances generate a voltage difference across the membrane called the **membrane potential**.

When a cell is "unstimulated," the movement of anions and cations across the membrane will be precisely balanced. In such steady-state

TABLE 12–1 A COMPARISON OF ION CONCENTRATIONS INSIDE AND OUTSIDE A TYPICAL MAMMALIAN CELL

Ion	Intracellular Concentration (mM)	Extracellular Concentration (mM)
Cations		
Na^+	5–15	145
K^+	140	5
Mg^{2+}	0.5*	1–2
Ca^{2+}	10^{-4}*	1–2
H^+	7×10^{-5} ($10^{-7.2}$ M or pH 7.2)	4×10^{-5} ($10^{-7.4}$ M or pH 7.4)
Anions**		
Cl^-	5–15	110

*The concentrations of Mg^{2+} and Ca^{2+} given are for the free ions. There is a total of about 20 mM Mg^{2+} and 1–2 mM Ca^{2+} in cells, but most of these ions are bound to proteins and other organic molecules and, for Ca^{2+}, stored within various organelles.
**In addition to Cl^-, a cell contains many other anions not listed in this table. In fact, most cell constituents are negatively charged (HCO_3^-, PO_4^{3-}, proteins, nucleic acids, metabolites carrying phosphate and carboxyl groups, and so on).

conditions, the voltage difference across the cell membrane—called the *resting membrane potential*—holds steady. But it is not zero. In animal cells, for example, the resting membrane potential can be anywhere between –20 and –200 millivolts (mV), depending on the organism and cell type. The value is expressed as a negative number because the interior of the cell is more negatively charged than the exterior.

The membrane potential allows cells to power the transport of certain metabolites, and it provides cells that are excitable with a means to communicate with their neighbors. As we discuss shortly, it is the activity of different membrane transport proteins, embedded in the bilayer, that enables cells to establish and maintain their characteristic membrane potential.

Cells Contain Two Classes of Membrane Transport Proteins: Transporters and Channels

Membrane transport proteins occur in many forms and are present in all cell membranes. Each provides a private portal across the membrane for a particular small, water-soluble substance—an ion, sugar, or amino acid, for example. Most of these membrane transport proteins allow passage of only select members of a particular type: some permit transit of Na^+ but not K^+, others K^+ but not Na^+, and so on. Each type of cell membrane has its own characteristic set of transport proteins, which determines exactly which solutes can pass into and out of that cell or organelle.

As discussed in Chapter 11, most membrane transport proteins have polypeptide chains that traverse the lipid bilayer multiple times—that is, they are multipass transmembrane proteins (see Figure 11–24). When these transmembrane segments cluster together, they establish a continuous protein-lined pathway that allows selected small, hydrophilic molecules to cross the membrane without coming into direct contact with the hydrophobic interior of the lipid bilayer.

Cells contain two main classes of membrane transport proteins: transporters and channels. These proteins differ in the way they discriminate between solutes, transporting some but not others (**Figure 12–3**). *Channels* discriminate mainly on the basis of size and electric charge: when the channel is open, only ions of an appropriate size and charge can pass through. A *transporter*, on the other hand, transfers only those molecules or ions that fit into specific binding sites on the protein. Transporters bind their solutes with great specificity, in the same way an enzyme binds its substrate, and it is this requirement for specific binding that gives transporters their selectivity.

Solutes Cross Membranes by Either Passive or Active Transport

Transporters and channels allow small, hydrophilic molecules and ions to cross the cell membrane, but what controls whether these substances move into the cell (or organelle)—or out of it? In many cases, the direction

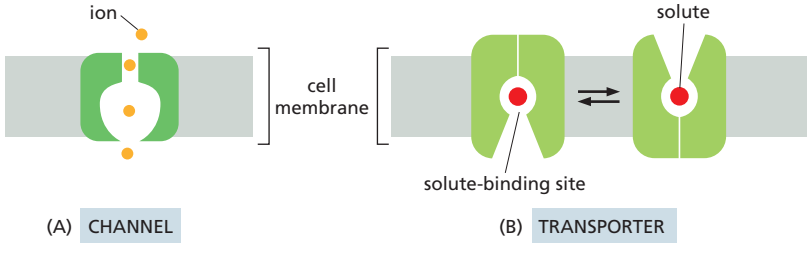


Figure 12–3 Inorganic ions and small, polar organic molecules can cross a cell membrane through either a transporter or a channel. (A) A channel forms a pore across the bilayer through which specific inorganic ions or, in some cases, polar organic molecules can diffuse. Ion channels can exist in either an open or a closed conformation, and they transport only in the open conformation, as shown here. Channel opening and closing is usually controlled by an external stimulus or by conditions within the cell. (B) A transporter undergoes a series of conformational changes to transfer small solutes across the lipid bilayer. Transporters are very selective for the solutes that they bind, and they transfer them at a much slower rate than do channels.

of transport depends only on the relative concentrations of the solute on either side of the membrane. Substances will spontaneously flow "downhill" from a region of high concentration to a region of low concentration, provided a pathway exists. Such movements are called passive, because they need no additional driving force. If, for example, a solute is present at a higher concentration outside the cell than inside, and an appropriate channel or transporter is present in the plasma membrane, the solute will move into the cell by **passive transport**, without expenditure of energy by the membrane transport protein. This is because even though the solute can move in either direction across the membrane, more solute will move in than out until the two concentrations equilibrate. All channels—and many transporters—act as conduits for such passive transport.

To move a solute against its concentration gradient, however, a membrane transport protein must do work: it has to drive the flow of the substance "uphill" from a region of low concentration to a region of higher concentration. To do so, it couples the transport to some other process that provides an input of energy (as discussed in Chapter 3). The movement of a solute against its concentration gradient in this way is termed **active transport**, and it is carried out by special types of transporters called *pumps*, which harness an energy source to power the transport process (**Figure 12–4**). As discussed later, this energy can come from ATP hydrolysis, a transmembrane ion gradient, or sunlight.

Both the Concentration Gradient and Membrane Potential Influence the Passive Transport of Charged Solutes

For an uncharged molecule, the direction of passive transport is determined solely by its concentration gradient, as we have outlined above. But for electrically charged substances, whether inorganic ions or small organic molecules, an additional force comes into play. As mentioned earlier, most cell membranes have a voltage across them—a difference in charge referred to as a membrane potential. This membrane potential exerts a force on any substance that carries an electric charge. The cytosolic side of the plasma membrane is usually at a negative potential relative to the extracellular side, so the membrane potential tends to pull positively charged ions and molecules into the cell and drive negatively charged solutes out.

At the same time, a charged solute—like an uncharged one—will also tend to move down its concentration gradient. The net force driving a charged solute across a cell membrane is therefore a composite of two forces, one due to the concentration gradient and the other due to the membrane potential. This net driving force, called the solute's **electrochemical gradient**, determines the direction in which each solute will flow across the membrane by passive transport.

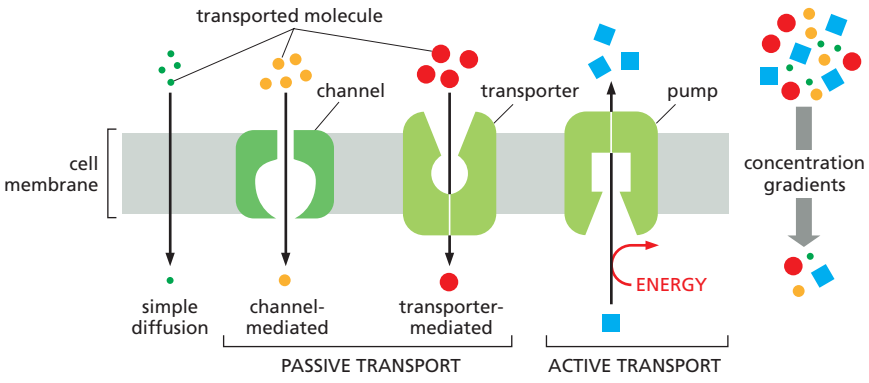


Figure 12–4 Solutes cross cell membranes by either passive or active transport. Some small, nonpolar molecules such as CO_2 (see Figure 12–2) can move passively down their concentration gradient across the lipid bilayer by simple diffusion, without the help of a membrane transport protein. Most solutes, however, require the assistance of a channel or transporter. Passive transport, which allows solutes to move down their concentration gradients, occurs spontaneously; active transport against a concentration gradient requires an input of energy. Only transporters can carry out active transport, and the transporters that perform this function are called pumps.

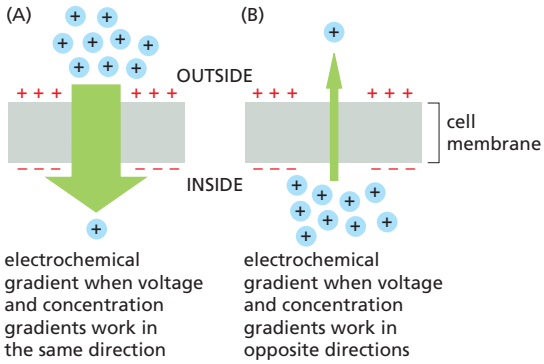


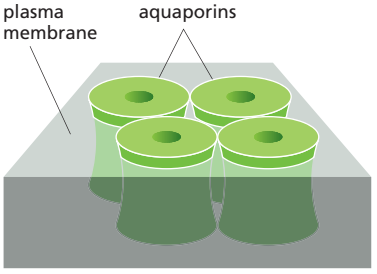
Figure 12–5 An electrochemical gradient has two components. The net driving force tending to move a charged solute across a cell membrane—its electrochemical gradient—is the sum of a force from the concentration gradient of the solute and a force from the membrane potential. The membrane potential is represented here by the + and – signs on opposite sides of the membrane. The width of the *green* arrow represents the magnitude of the electrochemical gradient. (A) The concentration gradient and membrane potential work together to increase the driving force for movement of the solute. Such is the case for Na^+. (B) The membrane potential acts against the concentration gradient, decreasing the electrochemical driving force. Such is the case for K^+.

For some ions, the voltage and concentration gradients work in the same direction, creating a relatively steep electrochemical gradient (**Figure 12–5A**). This is the case for Na^+, which is positively charged and at a higher concentration outside cells than inside (see Table 12–1). Na^+ therefore tends to enter cells when given an opportunity. If, however, the voltage and concentration gradients have opposing effects, the resulting electrochemical gradient can be small (**Figure 12–5B**). This is the case for K^+, which is present at a much higher concentration inside cells, where the resting membrane potential is negative. Because its electrochemical gradient across the plasma membrane of resting cells is small, there is little net movement of K^+ across the membrane even when K^+ channels are open.

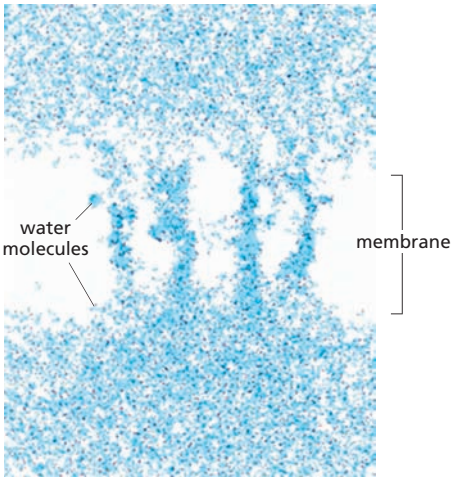
Water Moves Across Cell Membranes Down Its Concentration Gradient—a Process Called Osmosis

Cells are mostly water (generally about 70% by weight), and so the movement of water across cell membranes is crucially important for living things. Because water molecules are small and uncharged, they can diffuse directly across the lipid bilayer (see Figure 12–2). However, this movement is relatively slow. To facilitate the flow of water, some cells contain specialized channels called aquaporins in their plasma membrane (**Figure 12–6** and **Movie 12.1**). For many cells, such as those in the kidney or in various secretory glands, aquaporins are essential for their function.

But for water-filled cells in an aqueous environment, does water tend to enter the cell or leave it? As we saw in Table 12–1, cells contain a high concentration of solutes, including many charged molecules and ions. Thus the total concentration of solute particles inside the cell—also called its *osmolarity*—generally exceeds the solute concentration outside the cell. The resulting osmotic gradient tends to "pull" water into the cell. This movement of water down its concentration gradient—from an area of low solute concentration (high water concentration) to an area of high solute concentration (low water concentration)—is called **osmosis**.



(A)



(B)

Figure 12–6 Water molecules diffuse rapidly through aquaporin channels in the plasma membrane of some cells. (A) Shaped like an hourglass, each aquaporin channel forms a pore across the bilayer, allowing the selective passage of water molecules. Shown here is an aquaporin tetramer, the biologically active form of the protein. (B) In this snapshot, taken from a real-time, molecular dynamics simulation, four columns of water molecules (*blue*) can be seen passing through the pores of an aquaporin tetramer (not shown). The space where the membrane would be located is indicated. (B, adapted from B. de Groot and H. Grubmüller, *Science* 294:2353–2357, 2001.)

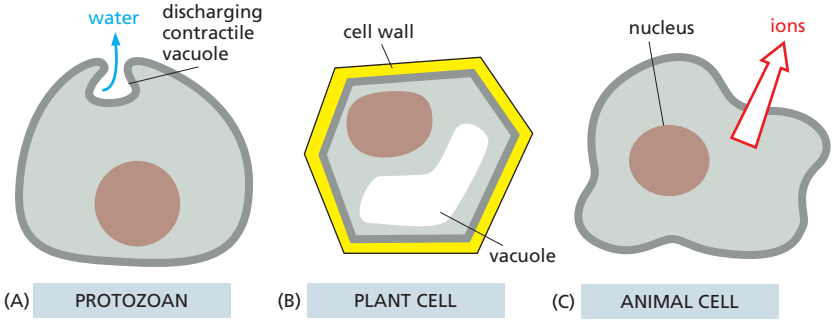


Figure 12–7 Cells use different tactics to avoid osmotic swelling. (A) A freshwater amoeba avoids swelling by periodically ejecting the water that moves into the cell and accumulates in contractile vacuoles. The contractile vacuole first accumulates solutes, which cause water to follow by osmosis; it then pumps most of the solutes back into the cytosol before emptying its contents at the cell surface. (B) The plant cell's tough cell wall prevents swelling. (C) The animal cell reduces its intracellular solute concentration by pumping out ions.

Osmosis, if it occurs without constraint, can make a cell swell. Different cells cope with this osmotic challenge in different ways. Some freshwater protozoans, such as amoebae, eliminate excess water using contractile vacuoles that periodically discharge their contents to the exterior (**Figure 12–7A**). Plant cells are prevented from swelling by their tough cell walls and so can tolerate a large osmotic difference across their plasma membrane (**Figure 12–7B**); indeed, plant cells make use of osmotic swelling pressure, or turgor pressure, to keep their cell walls tense, so that the stems of the plant are rigid and its leaves are extended. If turgor pressure is lost, plants wilt. Animal cells maintain osmotic equilibrium by using transmembrane pumps to expel solutes, such as the Na^+ ions that tend to leak into the cell (**Figure 12–7C**).

TRANSPORTERS AND THEIR FUNCTIONS

Transporters are responsible for the movement of most small, water-soluble, organic molecules and a handful of inorganic ions across cell membranes. Each transporter is highly selective, often transferring just one type of solute. To guide and propel the complex traffic of substances into and out of the cell, and between the cytosol and the different membrane-enclosed organelles, each cell membrane contains a characteristic set of different transporters appropriate to that particular membrane. For example, the plasma membrane contains transporters that import nutrients such as sugars, amino acids, and nucleotides; the lysosome membrane contains an H^+ transporter that imports H^+ to acidify the lysosome interior and other transporters that move digestion products out of the lysosome into the cytosol; the inner membrane of mitochondria contains transporters for importing the pyruvate that mitochondria use as fuel for generating ATP, as well as transporters for exporting ATP once it is synthesized (**Figure 12–8**).

In this section, we describe the general principles that govern the function of transporters, and we present a more detailed view of the molecular mechanisms that drive the movement of a few key solutes.

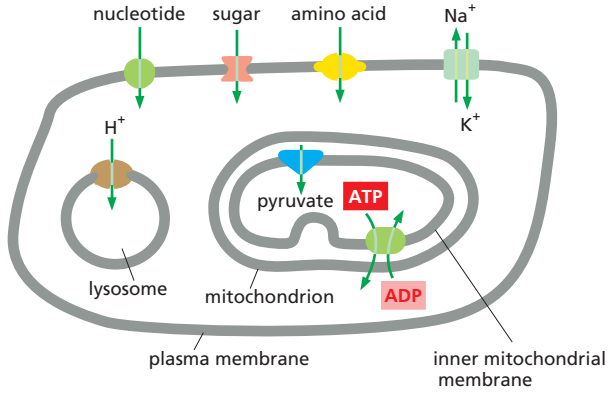


Figure 12–8 Each cell membrane has its own characteristic set of transporters. These transporters allow each membrane to carry out its unique functions. Only a few of these transporters are shown here.

QUESTION 12–1

A simple enzyme reaction can be described by the equation $E + S \leftrightarrow ES \leftrightarrow E + P$, where E is the enzyme, S the substrate, P the product, and ES the enzyme–substrate complex.

A. Write a corresponding equation describing the workings of a transporter (T) that mediates the transport of a solute (S) down its concentration gradient.

B. What does this equation tell you about the function of a transporter?

C. Why would this equation be an inappropriate choice to represent the function of a channel?

Passive Transporters Move a Solute Along Its Electrochemical Gradient

An important example of a transporter that mediates passive transport is the *glucose transporter* in the plasma membrane of many mammalian cell types. The protein, which consists of a polypeptide chain that crosses the membrane at least 12 times, can adopt several conformations—and it switches reversibly and randomly between them. In one conformation, the transporter exposes binding sites for glucose to the exterior of the cell; in another, it exposes the sites to the cell interior.

Because glucose is uncharged, the electrical component of its electrochemical gradient is zero. Thus the direction in which it is transported is determined by its concentration gradient alone. When glucose is plentiful outside cells, as it is after a meal, the sugar binds to the transporter's externally displayed binding sites; if the protein then switches conformation—spontaneously and at random—it will carry the bound sugar inward and release it into the cytosol, where the glucose concentration is low (**Figure 12–9**). Conversely, when blood glucose levels are low—as they are when you are hungry—the hormone glucagon stimulates liver cells to produce large amounts of glucose by the breakdown of glycogen. As a result, the glucose concentration is higher inside liver cells than outside. This glucose can bind to the internally displayed binding sites on the transporter. When the protein then switches conformation in the opposite direction—again spontaneously and randomly—the glucose will be transported out of the cells and made available for import by other, energy-requiring cells. The net flow of glucose can thus go either way, according to the direction of the glucose concentration gradient across the plasma membrane: inward if more glucose is binding to the transporter's externally displayed sites, and outward if the opposite is true.

Although passive transporters themselves play no part in controlling the direction of solute transport, they are highly selective in terms of which solutes they will move. For example, the binding sites in the glucose transporter bind only D-glucose and not its mirror image L-glucose, which the cell cannot use as an energy source.

Pumps Actively Transport a Solute Against Its Electrochemical Gradient

Cells cannot rely solely on passive transport to maintain the proper balance of solutes. The active transport of solutes against their electrochemical gradient is essential to achieving the appropriate intracellular

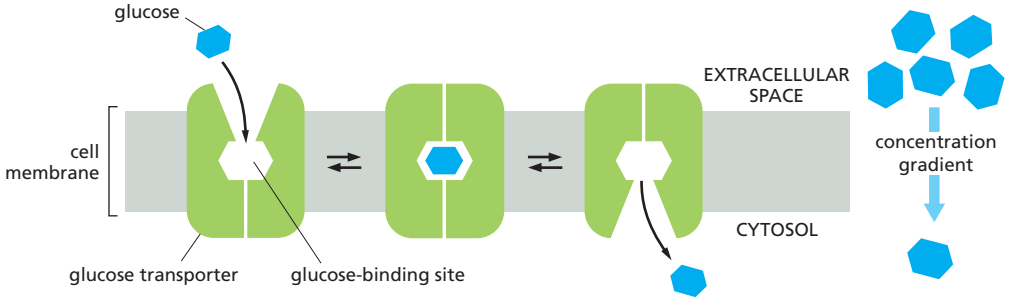


Figure 12–9 Conformational changes in a transporter mediate the passive transport of a solute such as glucose. The transporter is shown in three conformational states: in the outward-open state (*left*), the binding sites for solute are exposed on the outside; in the inward-open state (*right*), the sites are exposed on the inside of the bilayer; and in the occluded state (*center*), the sites are not accessible from either side. The transition between the states occurs randomly, is completely reversible, and—most importantly for the function of the transporter shown—does not depend on whether the solute-binding site is occupied. Therefore, if the solute concentration is higher on the outside of the bilayer, solute will bind more often to the transporter in the outward-open conformation than in the inward-open conformation, and there will be a net transport of glucose down its concentration gradient.

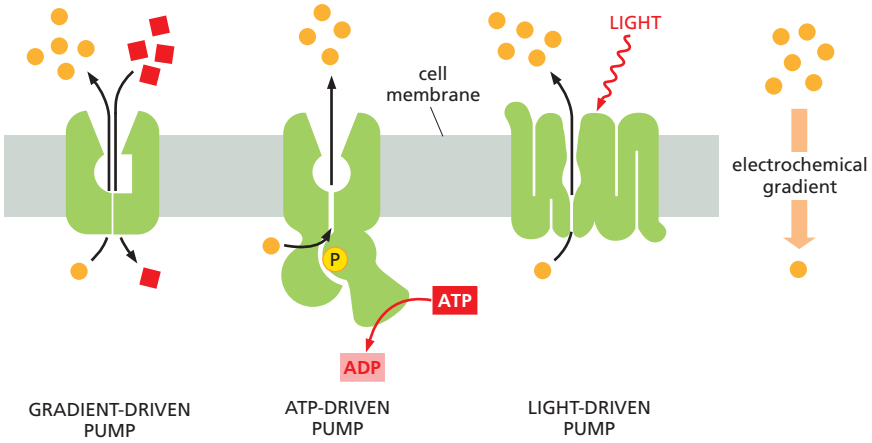


Figure 12–10 Pumps carry out active transport in three main ways. The actively transported solute is shown in *gold*, and the energy source is shown in *red*.

ionic composition and for importing solutes that are at a lower concentration outside the cell than inside. For these purposes, cells depend on transmembrane **pumps**, which can carry out active transport in three main ways (**Figure 12–10**): (i) *gradient-driven pumps* link the uphill transport of one solute across a membrane to the downhill transport of another; (ii) *ATP-driven pumps* use the energy released by the hydrolysis of ATP to drive uphill transport; and (iii) *light-driven pumps*, which are found mainly in bacterial cells, use energy derived from sunlight to drive uphill transport, as discussed in Chapter 11 for bacteriorhodopsin (see Figure 11–28).

These different forms of active transport are often linked. Thus, in the plasma membrane of an animal cell, an ATP-driven *Na^+ pump* transports Na^+ out of the cell against its electrochemical gradient; this Na^+ can then flow back into the cell, down its electrochemical gradient, through various Na^+ gradient-driven pumps. The influx of Na^+ through these gradient-driven pumps provides the energy for the active transport of many other substances into the cell against their electrochemical gradients. If the ATP-driven Na^+ pump ceased operating, the Na^+ gradient would soon run down, and transport through Na^+ gradient-driven pumps would come to a halt. For this reason, the ATP-driven Na^+ pump has a central role in the active transport of small molecules across the plasma membrane of animal cells. Plant cells, fungi, and many bacteria use ATP-driven H^+ pumps in an analogous way: in pumping H^+ out of the cell, these proteins create an electrochemical gradient of H^+ across the plasma membrane that is subsequently harnessed for solute transport, as we discuss later.

The Na^+ Pump in Animal Cells Uses Energy Supplied by ATP to Expel Na^+ and Bring in K^+

The ATP-driven **Na^+ pump** plays such a central part in the energy economy of animal cells that it typically accounts for 30% or more of their total ATP consumption. This pump uses the energy derived from ATP hydrolysis to transport Na^+ out of the cell as it carries K^+ in. The pump is therefore sometimes called the *Na^+-K^+ ATPase* or the *Na^+-K^+ pump*.

During the pumping process, the energy from ATP hydrolysis fuels a stepwise series of protein conformational changes that drives the exchange of Na^+ and K^+ ions. As part of the process, the phosphate group removed from ATP gets transferred to the pump itself (**Figure 12–11**).

The transport of Na^+ ions out, and K^+ ions in, takes place in a cycle in which each step depends on the one before (**Figure 12–12**). If any of the individual steps is prevented from occurring, the entire cycle halts. The toxin *ouabain*, for example, inhibits the Na^+ pump by preventing the binding of extracellular K^+, arresting the cycle.

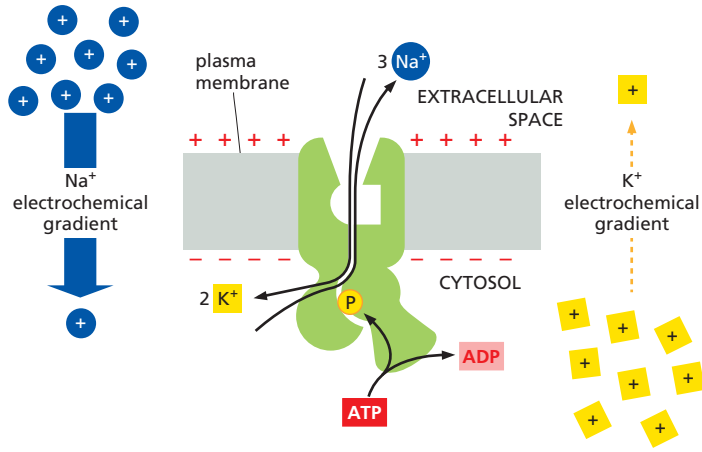


Figure 12–11 The Na^+ pump uses the energy of ATP hydrolysis to pump Na^+ out of animal cells and K^+ in. In this way, the pump helps keep the cytosolic concentrations of Na^+ low and K^+ high.

The Na^+ pump is very efficient: the whole pumping cycle takes only 10 milliseconds. Furthermore, the tight coupling between steps in the cycle ensures that the pump operates only when the appropriate ions—both Na^+ and K^+—are available to be transported, thereby avoiding a wasteful hydrolysis of ATP.

The Na^+ Pump Generates a Steep Concentration Gradient of Na^+ Across the Plasma Membrane

The Na^+ pump functions like a bilge pump in a leaky ship, ceaselessly expelling the Na^+ that is constantly slipping into the cell through other

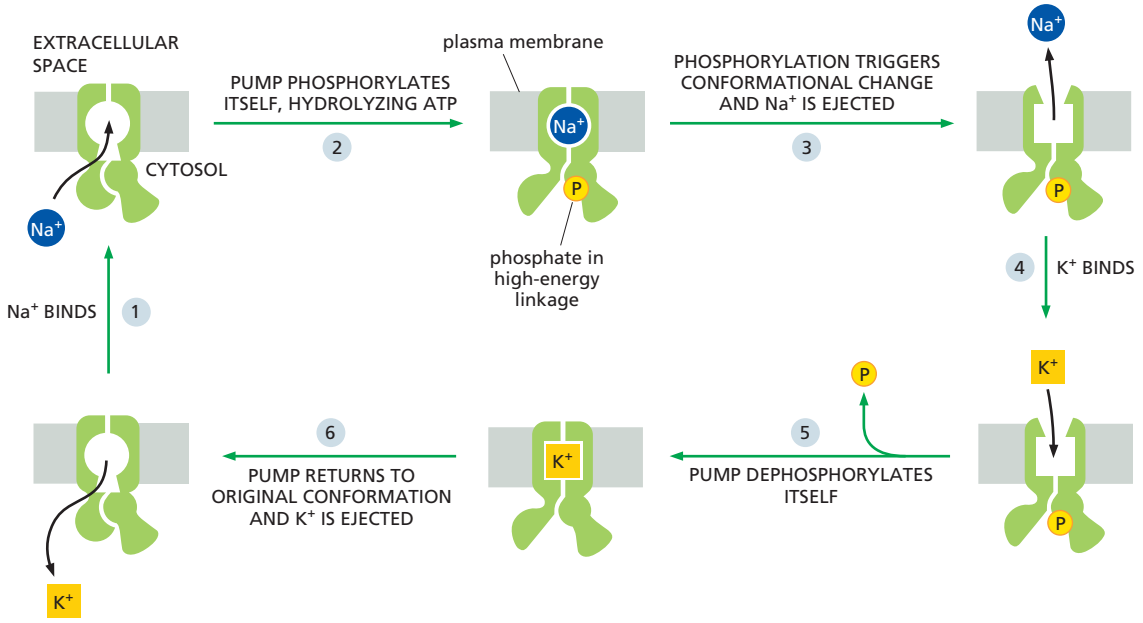


Figure 12–12 The Na^+ pump undergoes a series of conformational changes as it exchanges Na^+ ions for K^+. The binding of cytosolic Na^+ (1) and the subsequent phosphorylation by ATP of the cytosolic face of the pump (2) induce the protein to undergo conformational changes that transfer the Na^+ across the membrane and release it outside the cell (3). The high-energy linkage of the phosphate to the protein provides the energy to drive the conformational changes. The binding of K^+ from the extracellular space (4) and the subsequent dephosphorylation (5) allow the protein to return to its original conformation, which transfers the K^+ across the membrane and releases it into the cytosol (6).

The cycle is shown in **Movie 12.2**. The changes in conformation are analogous to those shown for the glucose transporter in Figure 12–9, except that here the Na^+-dependent phosphorylation and K^+-dependent dephosphorylation of the protein cause the conformational changes to occur in an orderly fashion, enabling the protein to do useful work. For simplicity, only one binding site is shown for each ion. The real pump in mammalian cells contains three binding sites for Na^+ and two for K^+. The net result of one cycle of the pump is therefore the transport of three Na^+ out and two K^+ in. Ouabain inhibits the pump by preventing K^+ binding (4).

Figure 12–13 The high concentration of Na^+ outside the cell is like water behind a high dam. The water behind the dam has potential energy, which can be used to drive energy-requiring processes. In the same way, an ion gradient across a membrane can be used to drive active processes in a cell, including the active transport of other molecules across the plasma membrane. Shown here is the Table Rock Dam in Branson, Missouri, USA. (Gary Saxe/Shutterstock.)

transporters and ion channels in the plasma membrane. In this way, the pump keeps the Na^+ concentration in the cytosol about 10–30 times lower than that in the extracellular fluid and the K^+ concentration about 10–30 times higher (see Table 12–1, p. 391).

This steep concentration gradient of Na^+ across the plasma membrane acts together with the membrane potential to create a large Na^+ electrochemical gradient (see Figure 12–5A). This high concentration of Na^+ outside the cell, on the uphill side of its electrochemical gradient, is like a large volume of water behind a high dam: it represents a very large store of energy (**Figure 12–13**). Even if one artificially halts the operation of the Na^+ pump with ouabain, this stored energy is sufficient to sustain for many minutes the various gradient-driven pumps in the plasma membrane that are fueled by the downhill flow of Na^+, which we discuss shortly.

Ca^{2+} Pumps Keep the Cytosolic Ca^{2+} Concentration Low

Ca^{2+}, like Na^+, is also kept at a low concentration in the cytosol compared with its concentration in the extracellular fluid. But Ca^{2+} is much less plentiful than Na^+, both inside and outside cells (see Table 12–1). The movement of this ion across cell membranes is nonetheless crucial, because Ca^{2+} can bind tightly to a variety of proteins in the cell, altering their activities. An influx of Ca^{2+} into the cytosol through Ca^{2+} channels, for example, is used by different cells as an intracellular signal to trigger various complex processes, such as muscle contraction (discussed in Chapter 17), fertilization (discussed in Chapters 16 and 19), and nerve cell communication, which is discussed later.

The lower the background concentration of free Ca^{2+} in the cytosol, the more sensitive the cell is to an increase in cytosolic Ca^{2+}. Thus eukaryotic cells in general maintain a very low concentration of free Ca^{2+} in their cytosol (about 10^{-4} mM) compared to the much higher concentration of Ca^{2+} outside of the cell (typically 1–2 mM). This huge concentration difference is achieved mainly by means of ATP-driven **Ca^{2+} pumps** in both the plasma membrane and the endoplasmic reticulum membrane, which actively remove Ca^{2+} from the cytosol.

Ca^{2+} pumps are ATPases that work in much the same way as the Na^+ pump depicted in Figure 12–12. The main difference is that Ca^{2+} pumps return to their original conformation without a requirement for binding and transporting a second ion (**Figure 12–14**). The Na^+ and Ca^{2+} pumps have similar amino acid sequences and structures, indicating that they share a common evolutionary origin.

Gradient-driven Pumps Exploit Solute Gradients to Mediate Active Transport

A gradient of any solute across a membrane, like the electrochemical Na^+ gradient generated by the Na^+ pump, can be used to drive the active transport of a second molecule. The downhill movement of the first solute down its gradient provides the energy to power the uphill transport of the second solute. The active transporters that work in this way are

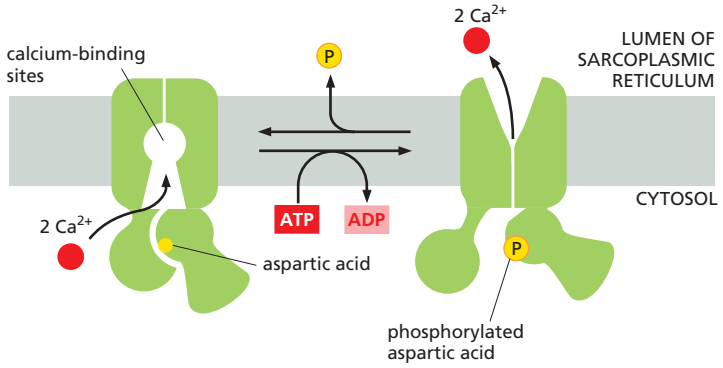


Figure 12–14 The Ca^{2+} pump in the sarcoplasmic reticulum was the first ATP-driven ion pump to have its three-dimensional structure determined by x-ray crystallography. When a muscle cell is stimulated, Ca^{2+} floods into the cytosol from the sarcoplasmic reticulum—a specialized form of endoplasmic reticulum. The influx of Ca^{2+} stimulates the cell to contract; to recover from the contraction, Ca^{2+} must be pumped back into the sarcoplasmic reticulum by this Ca^{2+} pump.

The Ca^{2+} pump uses ATP to phosphorylate itself, inducing a series of conformational changes (similar to the ones of the Na^+ pump shown in Figure 12–12); when the pump is open to the lumen of the sarcoplasmic reticulum, the Ca^{2+}-binding sites are eliminated, ejecting the two Ca^{2+} ions into the organelle (**Movie 12.3**).

called **gradient-driven pumps** (see Figure 12–10). They can couple the movement of one inorganic ion to that of another, the movement of an inorganic ion to that of a small organic molecule, or the movement of one small organic molecule to that of another. If the pump moves both solutes in the same direction across the membrane, it is called a *symport*. If it moves them in opposite directions, it is called an *antiport*. A transporter that ferries only one type of solute across the membrane down its concentration gradient (and is therefore not a pump) is called a *uniport* (**Figure 12–15**). The glucose transporter described earlier (see Figure 12–9) is an example of a uniport.

The Electrochemical Na^+ Gradient Drives the Transport of Glucose Across the Plasma Membrane of Animal Cells

Symports that make use of the inward flow of Na^+ down its steep electrochemical gradient have an especially important role in driving the import of solutes into animal cells. The epithelial cells that line the gut, for example, transport glucose from the gut lumen across the gut epithelium and, ultimately, into the blood. If these cells had only a passive glucose uniport (the transporter shown in Figure 12–9), they would release glucose into the gut lumen after fasting just as freely as they take it up from the gut after a feast. However, these epithelial cells also possess a *glucose–Na^+ symport*, which they can use to take up glucose from the gut lumen, even when the concentration of glucose is higher in the epithelial cell's cytosol than it is inside the gut. As the electrochemical gradient for Na^+ is so steep, when Na^+ moves into the cell down its gradient, glucose is, in a sense, "dragged" into the cell along with it. Because the binding of Na^+ and glucose is cooperative—the binding of one enhances the binding of the other—if one of the two solutes is missing, the other fails to bind; therefore both molecules must be present for this gradient-driven

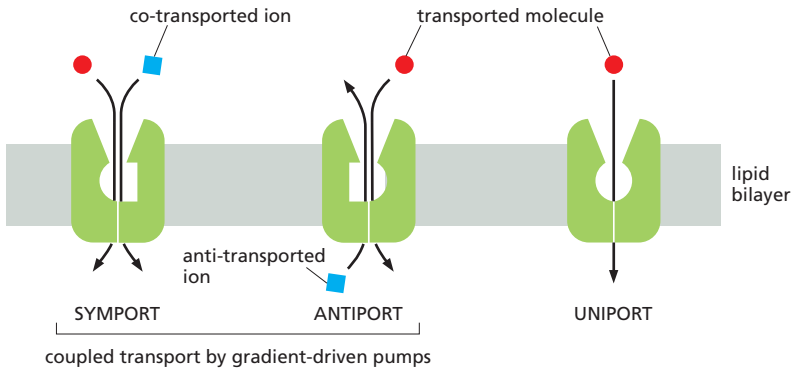


Figure 12–15 Gradient-driven pumps can act as symports or antiports. They transfer solutes either in the same direction, in which case they are called symports, or in opposite directions, which are antiports (**Movie 12.4**). Uniports, by contrast, only facilitate the movement of a solute down its concentration gradient. Because such movement does not require an additional energy source, uniports are not pumps.

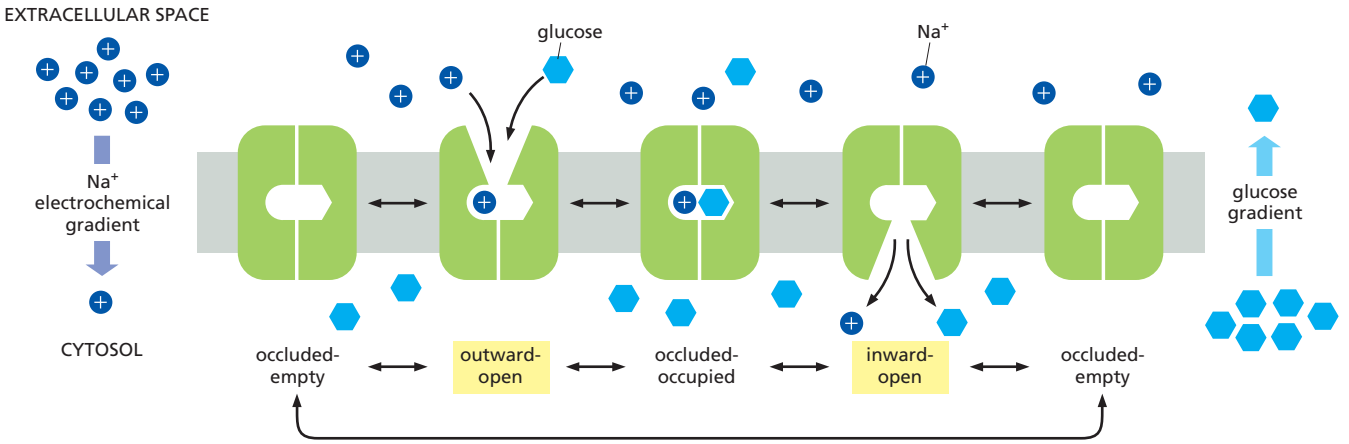


Figure 12–16 A glucose–Na^+ symport uses the electrochemical Na^+ gradient to drive the active import of glucose. The pump oscillates randomly between alternate states. In one state ("outward-open") the pump is open to the extracellular space; in another state ("inward-open") it is open to the cytosol. Although Na^+ and glucose can each bind to the pump in either of these "open" states, the pump can transition between them only through an "occluded" state in which both glucose and Na^+ are bound ("occluded-occupied") or neither is bound ("occluded-empty"). Because the Na^+ concentration is high in the extracellular space, the Na^+-binding site is readily occupied in the outward-open state, and the transporter must wait for a rare glucose molecule to bind. At that point, the pump flips to the occluded-occupied state, trapping both solutes.

Because conformational transitions are reversible, one of two things can happen to the pump in the occluded-occupied state. The transporter could flip back to the outward-open state; in this case, the solutes would dissociate, and nothing would be gained. Alternatively, it could flip into the inward-open state, exposing the solute-binding sites to the cytosol where the Na^+ concentration is very low. Thus sodium readily dissociates (and will be subsequently pumped back out of the cell by the Na^+ pump, shown in Figure 12–11, to maintain the steep Na^+ gradient). The transporter is now trapped with a partially occupied binding site until the glucose molecule also dissociates. At this point, with no solute bound, it can transition into the occluded-empty state and from there back to the outward-open state to repeat the transport cycle.

transport to occur and Na^+ will not leak into the cell without doing useful work (**Figure 12–16**).

If the gut epithelial cells had *only* this symport, however, they would take up glucose and never release it for use by the other cells of the body. These epithelial cells, therefore, have two types of glucose transporters located at opposite ends of the cell. In the apical domain of the plasma membrane, which faces the gut lumen, they have the glucose–Na^+ symports. These use the energy of the Na^+ gradient to actively import glucose, creating a high concentration of the sugar in the cytosol. In the basal and lateral domains of the plasma membrane, the cells have passive glucose uniports, which release the glucose down its concentration gradient for use by other tissues (**Figure 12–17**). As shown in Figure 12–17, the two types of glucose transporters are kept segregated in their proper domains of the plasma membrane by a diffusion barrier formed by a tight junction around the apex of the cell. This prevents mixing of membrane components between the two domains, as discussed in Chapter 11 (see Figure 11–32).

Cells in the lining of the gut and in many other organs, including the kidney, contain a variety of symports in their plasma membrane that are similarly driven by the electrochemical gradient of Na^+; each of these gradient-driven pumps specifically imports a small group of related sugars or amino acids into the cell. At the same time, Na^+-driven pumps that operate as **antiports** are also important for cells. For example, the *Na^+–H^+ exchanger* in the plasma membrane of many animal cells uses the downhill influx of Na^+ to pump H^+ out of the cell; it is one of the main devices that animal cells use to control the pH in their cytosol—preventing the cell interior from becoming too acidic.

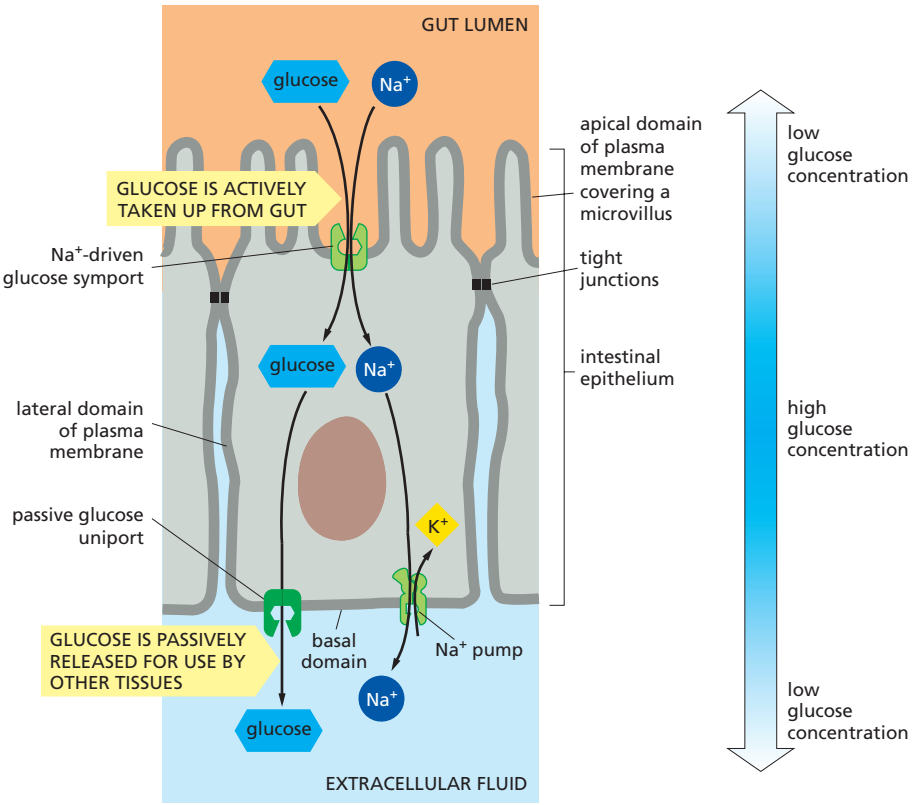


Figure 12–17 Two types of glucose transporters enable gut epithelial cells to transfer glucose across the epithelial lining of the gut. Na^+ that enters the cell via the Na^+-driven glucose symport is subsequently pumped out by Na^+ pumps in the basal and lateral plasma membranes, keeping the concentration of Na^+ in the cytosol low—and the Na^+ electrochemical gradient steep. The diet provides ample Na^+ in the gut lumen to drive the Na^+ gradient-driven glucose symport. The process is shown in Movie 12.5.

QUESTION 12–2

A rise in the intracellular Ca^{2+} concentration causes muscle cells to contract. In addition to an ATP-driven Ca^{2+} pump, muscle cells that contract quickly and regularly, such as those of the heart, have an additional type of Ca^{2+} pump—an antiport that exchanges Ca^{2+} for extracellular Na^+ across the plasma membrane. The majority of the Ca^{2+} ions that have entered the cell during contraction are rapidly pumped back out of the cell by this antiport, thus allowing the cell to relax. Ouabain and digitalis are used for treating patients with heart disease because they make heart muscle cells contract more strongly. Both drugs function by partially inhibiting the Na^+ pump in the plasma membrane of these cells. Can you propose an explanation for the effects of the drugs in the patients? What will happen if too much of either drug is taken?

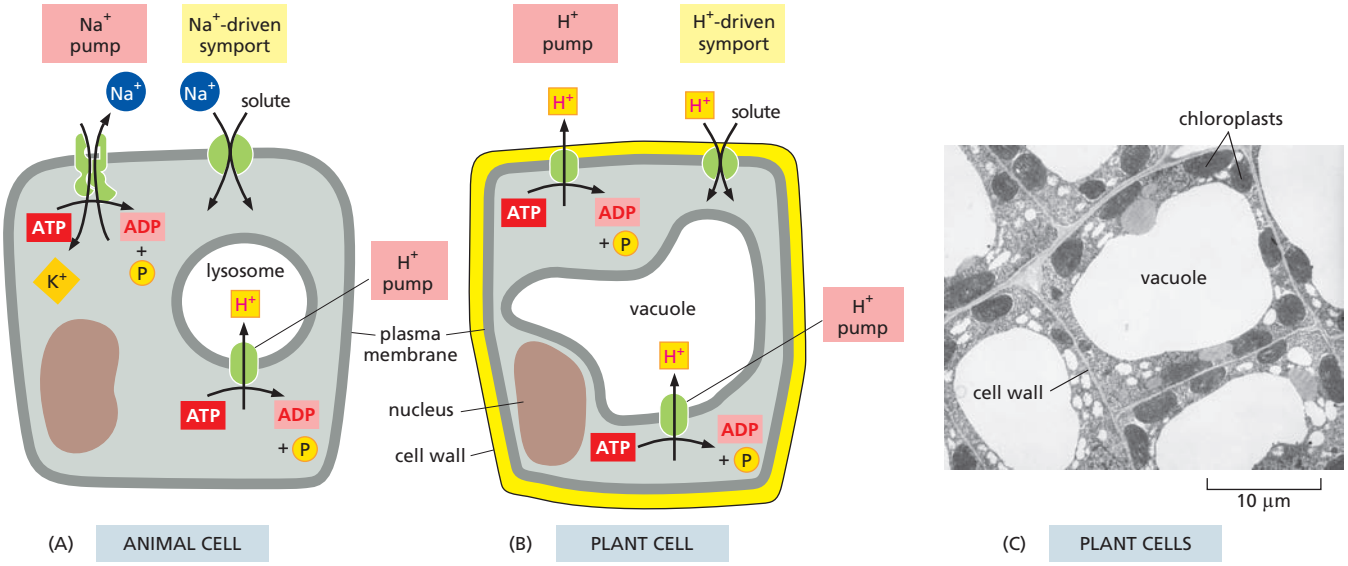
Electrochemical H^+ Gradients Drive the Transport of Solutes in Plants, Fungi, and Bacteria

Plant cells, bacteria, and fungi (including yeasts) do not have Na^+ pumps in their plasma membrane. Instead of an electrochemical Na^+ gradient, they rely mainly on an electrochemical gradient of H^+ to import solutes into the cell. The gradient is created by **H^+ pumps** in the plasma membrane that pump H^+ out of the cell, thus setting up an electrochemical proton gradient across this membrane and creating an acid pH in the medium surrounding the cell. The import of many sugars and amino acids into bacterial cells is then mediated by H^+ symports, which use the electrochemical H^+ gradient in much the same way that animal cells use the electrochemical Na^+ gradient to import these nutrients.

In some photosynthetic bacteria, the H^+ gradient is created by the activity of light-driven H^+ pumps such as bacteriorhodopsin (see Figure 11–28). In other bacteria, fungi, and plants, the H^+ gradient is generated by H^+ pumps in the plasma membrane that use the energy of ATP hydrolysis to pump H^+ out of the cell; these H^+ pumps resemble the Na^+ pumps and Ca^{2+} pumps of animal cells discussed earlier.

A different type of ATP-dependent H^+ pump is found in the membranes of some intracellular organelles, such as the lysosomes of animal cells and the central vacuole of plant and fungal cells. These pumps—which resemble the turbine-like enzyme that synthesizes ATP in mitochondria and chloroplasts (discussed in Chapter 14)—actively transport H^+ out of the cytosol into the organelle, thereby helping to keep the pH of the cytosol neutral and the pH of the interior of the organelle acidic. An acid environment is crucial to the function of many organelles, as we discuss in Chapter 15.

Some of the transmembrane pumps considered in this chapter are shown in Figure 12–18 and are listed in Table 12–2.



ION CHANNELS AND THE MEMBRANE POTENTIAL

In principle, the simplest way to allow a small, water-soluble substance to cross from one side of a membrane to the other is to create a hydrophilic channel through which the solute can pass. Channel proteins, or **channels**, perform this function in cell membranes, forming transmembrane pores that allow the passive movement of small, water-soluble molecules and ions into or out of the cell or organelle.

A few channels form relatively large, aqueous pores; examples are the proteins that form *gap junctions* between two adjacent cells (see Figure 20–28) and the *porins* that form pores in the outer membrane of mitochondria and some bacteria (see Figure 11–25). But such large, permissive channels would lead to disastrous leaks if they directly connected the cytosol of a cell to the extracellular space. Thus most of the channels in the plasma membrane form narrow, highly selective pores.

Figure 12–18 Animal and plant cells use a variety of transmembrane pumps to drive the active transport of solutes. (A) In animal cells, an electrochemical Na^+ gradient across the plasma membrane, generated by the Na^+ pump, is used by symports to import various solutes. (B) In plant cells, an electrochemical gradient of H^+, set up by an H^+ pump, is often used for this purpose; a similar strategy is used by bacteria and fungi (not shown). The lysosomes in animal cells and the vacuoles in plant and fungal cells contain a similar H^+ pump in their membranes that pumps in H^+, helping to keep the internal environment of these organelles acidic. (C) An electron micrograph shows the vacuole in plant cells in a young tobacco leaf. (C, courtesy of J. Burgess.)

TABLE 12–2 SOME EXAMPLES OF TRANSMEMBRANE PUMPS

Pump	Location	Energy Source	Function
Na^+-driven glucose pump (glucose–Na^+ symport)	apical plasma membrane of kidney and intestinal cells	Na^+ gradient	active import of glucose
Na^+–H^+ exchanger	plasma membrane of animal cells	Na^+ gradient	active export of H^+ ions, pH regulation
Na^+ pump (Na^+-K^+ ATPase)	plasma membrane of most animal cells	ATP hydrolysis	active export of Na^+ and import of K^+
Ca^{2+} pump (Ca^{2+} ATPase)	plasma membrane of eukaryotic cells	ATP hydrolysis	active export of Ca^{2+}
Ca^{2+} pump (Ca^{2+} ATPase)	sarcoplasmic reticulum membrane of muscle cells and endoplasmic reticulum membrane of most animal cells	ATP hydrolysis	active import of Ca^{2+} into sarcoplasmic reticulum or endoplasmic reticulum
H^+ pump (H^+ ATPase)	plasma membrane of plant cells, fungi, and some bacteria	ATP hydrolysis	active export of H^+
H^+ pump (H^+ ATPase)	membranes of lysosomes in animal cells and of vacuoles in plant and fungal cells	ATP hydrolysis	active export of H^+ from cytosol into lysosome or vacuole
Bacteriorhodopsin	plasma membrane of some bacteria	light	active export of H^+

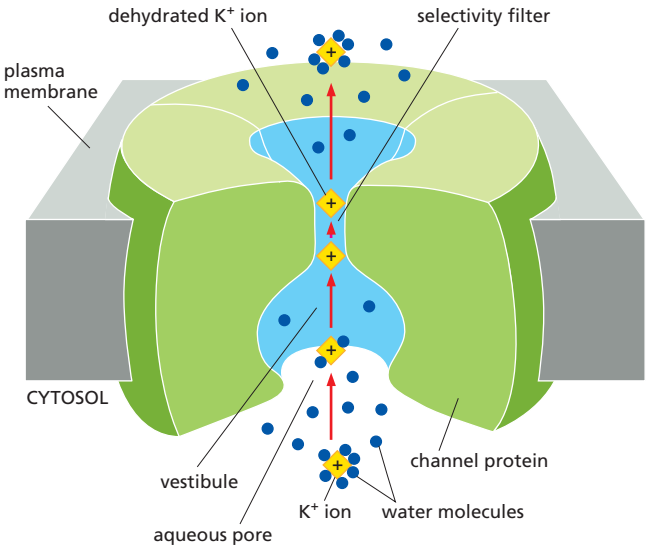


Figure 12–19 An ion channel has a selectivity filter that controls which inorganic ions it will allow to cross the membrane. Shown here is a portion of a bacterial K^+ channel. One of the four protein subunits has been omitted from the drawing to expose the interior structure of the pore (*blue*). From the cytosolic side, the pore opens into a vestibule that sits in the middle of the membrane. K^+ ions in the vestibule are still partially cloaked with associated water molecules. The narrow selectivity filter, which connects the vestibule with the outside of the cell, is lined with polar groups (not shown) that form transient binding sites for the K^+ ions once the ions have shed their water shell. To observe this selectivity in action. (Adapted from D.A. Doyle et al., *Science* 280:69–77, 1998.)

The aquaporins discussed earlier, for example, facilitate the flow of water across the plasma membrane of some prokaryotic and eukaryotic cells. These pores are structured in such a way that they allow the passive diffusion of uncharged water molecules, while prohibiting the movement of ions, including even the smallest ion, H^+.

The bulk of a cell's channels facilitate the passage of select inorganic ions. It is these ion channels that we discuss in this section.

Ion Channels Are Ion-selective and Gated

Two important properties distinguish **ion channels** from simple holes in the membrane. First, they show *ion selectivity*, permitting some inorganic ions to pass but not others. Ion selectivity depends on the diameter and shape of the ion channel and on the distribution of the charged amino acids that line it. Each ion in aqueous solution is surrounded by a small shell of water molecules, most of which have to be shed for the ions to pass, in single file, through the selectivity filter in the narrowest part of the ion channel (**Figure 12–19**). An ion channel is narrow enough in places to force ions into contact with the channel wall, so that only those ions of appropriate size and charge are able to pass (**Movie 12.6**).

The second important distinction between ion channels and simple holes in the membrane is that ion channels are not continuously open. Ion transport would be of no value to the cell if the many thousands of ion channels in a cell membrane were open all the time and there were no means of controlling the flow of ions through them. Instead, ion channels open only briefly and then close again (**Figure 12–20**). As we discuss later, most ion channels are *gated*: a specific stimulus triggers them to switch between a closed and an open state by inducing a change in their conformation.

Unlike a transporter, an ion channel does not need to undergo conformational changes for each ion it passes, and so it has a large advantage over a transporter with respect to its maximum rate of transport. More than a million ions can pass through an open channel each second, which is 1000 times greater than the fastest rate of transfer known for any transporter. On the other hand, channels cannot couple the ion flow to an energy source to carry out active transport; they simply make the membrane transiently permeable to selected inorganic ions, mainly Na^+, K^+, Ca^{2+}, or Cl^-.

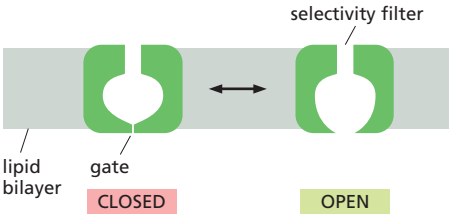


Figure 12–20 A typical ion channel fluctuates between closed and open conformations. The channel shown here in cross section forms a hydrophilic pore across the lipid bilayer only in the "open" conformation. As illustrated in Figure 12–19, the pore narrows to atomic dimensions in the selectivity filter, where the ion selectivity of the channel is largely determined.

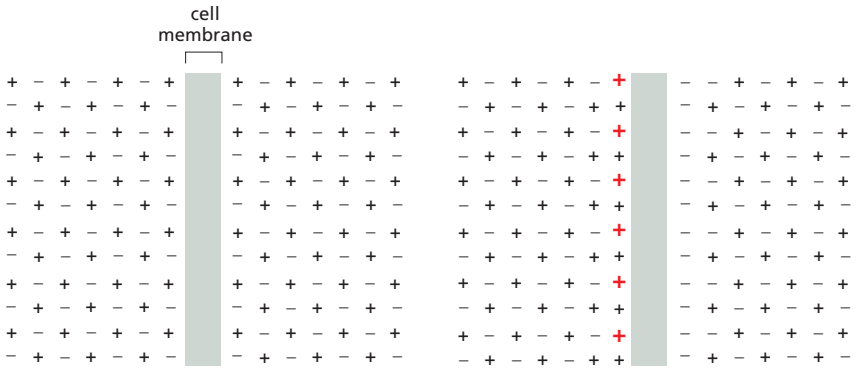
Figure 12–21 A Venus flytrap uses electrical signaling to capture its prey. The leaves snap shut in less than half a second when an insect moves across them. The response is triggered by touching any two of the three trigger hairs in succession in the center of each leaf. This mechanical stimulation opens ion channels in the plasma membrane and thereby sets off an electrical signal, which, by an unknown mechanism, leads to a rapid change in turgor pressure that closes the leaf. (Gabor Izso/Getty Images.)

Thanks to active transport by pumps, the concentrations of many ions are far from equilibrium across a cell membrane. When an ion channel opens, therefore, ions usually flow through it, moving rapidly down their electrochemical gradients. This rapid shift of ions changes the membrane potential, as we discuss next.

Membrane Potential Is Governed by the Permeability of a Membrane to Specific Ions

Changes in membrane potential are the basis of electrical signaling in many types of cells, whether they are the nerve or muscle cells in animals, or the touch-sensitive cells of a carnivorous plant (**Figure 12–21**). Such electrical changes are mediated by alterations in the permeability of membranes to ions. As we saw earlier, in an animal cell that is in an unstimulated, or "resting," state, the negative charges on the many types of organic molecules found inside the cell are largely balanced by K^+, the predominant intracellular ion (see Table 12–1). K^+ is continuously imported into the cell by the Na^+ pump, which generates a K^+ gradient across the plasma membrane as it pumps Na^+ out and K^+ in (see Figure 12–11).

The plasma membrane, however, also contains a set of K^+ channels, known as **K^+ leak channels**, that allow K^+ to move freely across the membrane. In a resting cell, these are the main ion channels open in the plasma membrane, rendering the membrane much more permeable to K^+ than to other ions. When K^+ flows out of the cell—down the concentration gradient generated by the ceaseless operation of the Na^+ pump—the loss of positive charge inside the cell creates a voltage difference, or membrane potential (**Figure 12–22**). Because this charge imbalance will oppose any further movement of K^+ out of the cell, an equilibrium condition is established in which the membrane potential keeping K^+ inside the cell is just strong enough to counteract the tendency of K^+ to move down its concentration gradient and out of the cell. In this state of equilibrium,



(A) exact balance of charges on each side of the membrane: membrane potential = 0

(B) a few positive ions (*red*) cross the membrane from right to left, setting up a nonzero membrane potential

Figure 12–22 The distribution of ions on either side of a cell membrane gives rise to its membrane potential. The membrane potential results from a thin (<1 nm) layer of ions close to the membrane, held in place by their electrical attraction to oppositely charged ions on the other side of the membrane. (A) When there is an exact balance of charges on either side of the membrane, there is no membrane potential. (B) When ions of one type cross the membrane, they establish a charge difference across the two sides of the membrane that creates a membrane potential. The number of ions that must move across the membrane to set up a membrane potential is a tiny fraction of all those present on either side. In the case of the plasma membrane in animal cells, for example, 6000 K^+ ions crossing 1 μm^2 of membrane are enough to shift the membrane potential by about 100 mV; the number of K^+ ions in 1 μm^3 of cytosol is 70,000 times larger than this.

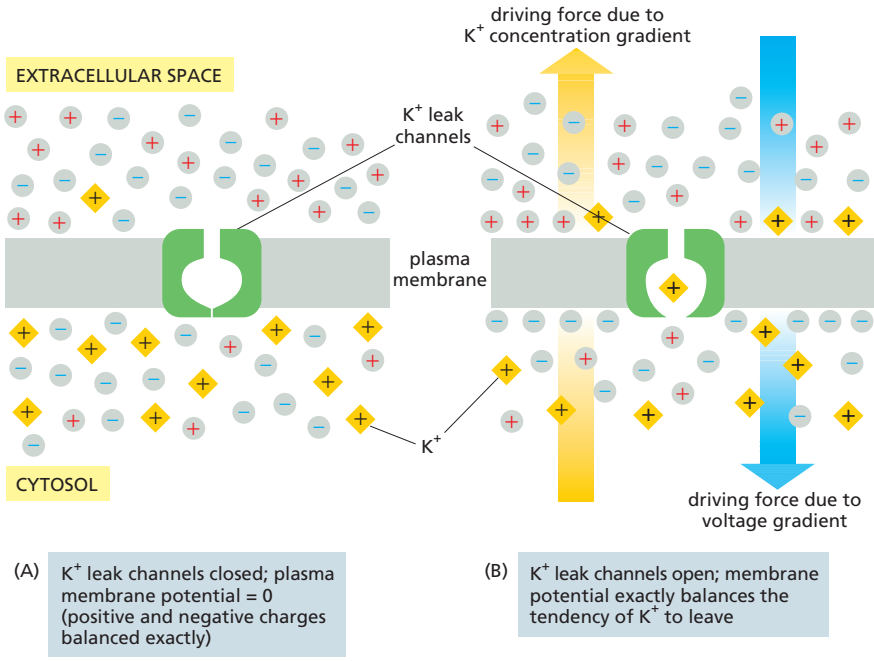


Figure 12–23 The K^+ concentration gradient and K^+ leak channels play major parts in generating the resting membrane potential across the plasma membrane in animal cells. (A) A hypothetical situation in which the K^+ leak channels are closed and the membrane potential is zero. (B) As soon as the channels open, K^+ will tend to leave the cell, moving down its concentration gradient. Assuming the membrane contains no open channels permeable to other ions, K^+ will cross the membrane but negative ions will be unable to follow. The resulting charge imbalance gives rise to a membrane potential that tends to drive K^+ back into the cell. At equilibrium, the effect of the K^+ concentration gradient is exactly balanced by the effect of the membrane potential, and there is no net movement of K^+ across the membrane.

The Na^+ pump (not shown here) also contributes to the resting potential—both by helping to establish the K^+ gradient and by pumping 3 Na^+ ions out of the cell for every 2 K^+ ions it pumps in (see Figure 12–11). Moving one more positively charged ion out of the cell with each pumping cycle helps to keep the inside of the cell more negative than the outside.

the electrochemical gradient for K^+ is zero, even though there is still a much higher concentration of K^+ inside the cell than out (**Figure 12–23**).

The membrane potential in such steady-state conditions—in which the flow of positive and negative ions across the plasma membrane is precisely balanced, so that no further difference in charge accumulates across the membrane—is called the **resting membrane potential**. A simple formula called the **Nernst equation** expresses this equilibrium quantitatively and makes it possible to calculate the theoretical resting membrane potential if the ion concentrations on either side of the membrane are known (**Figure 12–24**). In animal cells, the resting membrane potential—which varies between –20 and –200 mV—is chiefly a reflection of the electrochemical K^+ gradient across the plasma membrane, because, at rest, the plasma membrane is chiefly permeable to K^+, and K^+ is the main positive ion inside the cell.

When a cell is stimulated, other ion channels in the plasma membrane open, changing the membrane's permeability to those ions. Whether the ions enter or leave the cell depends on the direction of their electrochemical gradients. Thus the membrane potential at any time depends on both the state of the membrane's ion channels and the ion concentrations on either side of the plasma membrane. Bulk changes in ion concentrations cannot occur quickly enough to drive the rapid changes in membrane potential that are associated with electrical signaling. Instead, it is the rapid opening and closing of ion channels, which occurs within milliseconds, that matters most for this type of cell signaling.

The force tending to drive an ion across a membrane is made up of two components: one due to the electrical membrane potential and one due to the concentration gradient of the ion. At equilibrium, the two forces are balanced and satisfy a simple mathematical relationship given by the

Nernst equation

$$V = 62 \log_{10} (C_o / C_i)$$

where V is the membrane potential in millivolts, and C_o and C_i are the outside and inside concentrations of the ion, respectively. This form of the equation assumes that the ion carries a single positive charge and that the temperature is 37°C.

Figure 12–24 The Nernst equation can be used to calculate the contribution of each ion to the resting potential of the membrane. The relevant ion concentrations are those on either side of the membrane. From this equation, we see that each tenfold change in the ion concentration ratio (C_o/C_i) across the membrane alters the membrane potential by 62 millivolts. The resting potential can then be calculated by combining the individual ion gradient contributions and adjusting for the relative permeability for each ion.

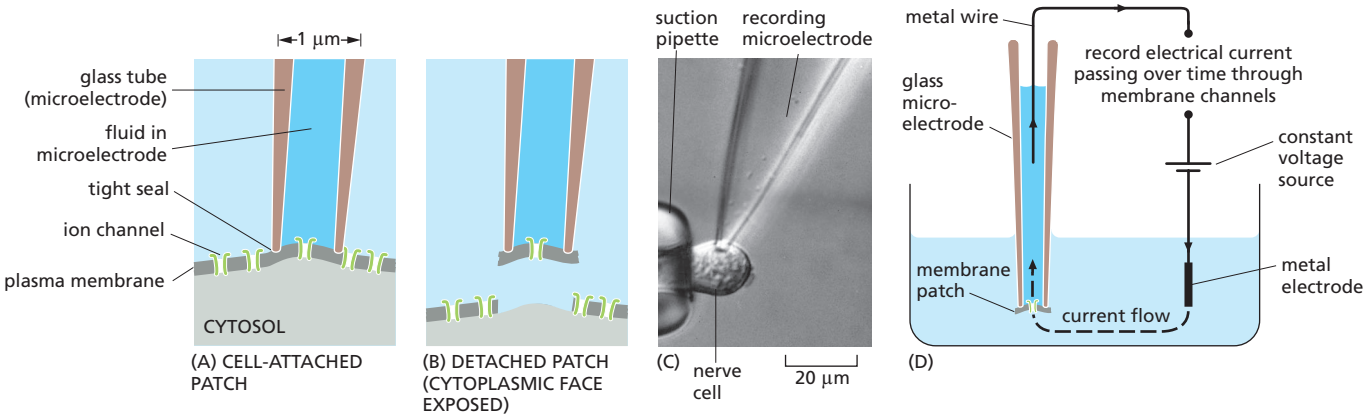


Figure 12–25 Patch-clamp recording is used to monitor ion channel activity. First, a microelectrode is filled with an aqueous conducting solution, and its tip is pressed against the surface of the cell. (A) With gentle suction, a tight seal is formed where the cell membrane contacts the mouth of the microelectrode. Because of the extremely tight seal, current can enter or leave the microelectrode only by passing through the ion channel or channels in the patch of membrane covering its tip. (B) To expose the cytosolic face of the membrane, the patch of membrane held in the microelectrode can be torn from the cell. This technique makes it easy to alter the composition of the solution on either side of the membrane to test the effect of various solutes on channel activity. (C) A micrograph showing an isolated nerve cell held in a suction pipette (the tip of which is shown on the left), while a microelectrode is being used for patch-clamp recording. (D) The circuitry for patch-clamp recording. At the open end of the microelectrode, a metal wire is inserted. Current that enters the microelectrode through ion channels in the small patch of membrane covering its tip passes via the wire, through measuring instruments, back into the bath of medium surrounding the cell or the detached patch. (C, from T.D. Lamb, H.R. Matthews, and V. Torre, *J. Physiol.* 372:315–349, 1986. With permission from Blackwell Publishing.)

Ion Channels Randomly Snap Between Open and Closed States

Measuring changes in electrical current is the main method used to study ion movements and ion channels in living cells. Amazingly, electrical recording techniques can detect and measure the current flowing through a single channel molecule. The procedure developed for doing this is known as **patch-clamp recording**, and it provides a direct and surprising picture of how individual ion channels behave.

In patch-clamp recording, a fine glass tube is used as a *microelectrode* to isolate and make electrical contact with a small area of the membrane at the surface of the cell (**Figure 12–25**). When a sufficiently small area of membrane is trapped in the patch, sometimes only a single ion channel will be present. Modern electrical instruments are sensitive enough to monitor the ion flow through this single channel, detected as a minute electric current (of the order of 10^{-12} ampere or 1 picoampere).

Monitoring individual ion channels in this way revealed something surprising about the way they behave: even when conditions are held constant, the currents abruptly appear and disappear, as though an on/off switch were being jiggled randomly (**Figure 12–26**). This behavior

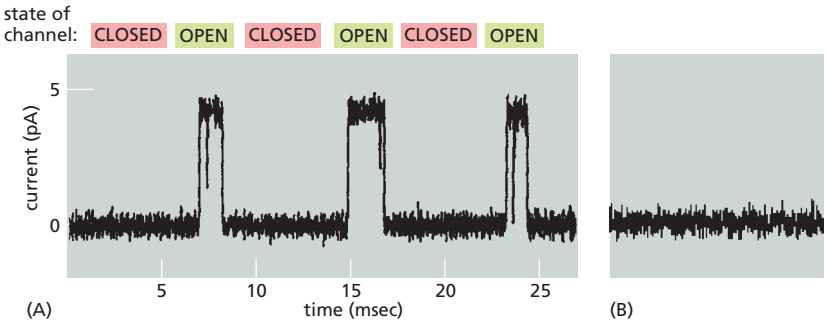


Figure 12–26 The behavior of a single ion channel can be observed using the patch-clamp technique. The voltage (the membrane potential) across the isolated patch of membrane is held constant during the recording. (A) In this example, the neurotransmitter acetylcholine is present, and the membrane patch from a muscle cell contains a single channel protein that is responsive to acetylcholine (discussed later, see Figure 12–42). This ion channel opens to allow passage of positive ions when acetylcholine binds to the exterior face of the channel. But even when acetylcholine is bound to the channel, as is the case during the three channel openings shown here, the channel does not remain open all the time. Instead, it flickers between open and closed states. Note that how long the channel remains open is variable. (B) When acetylcholine is not present, the channel opens very rarely. (Courtesy of David Colquhoun.)

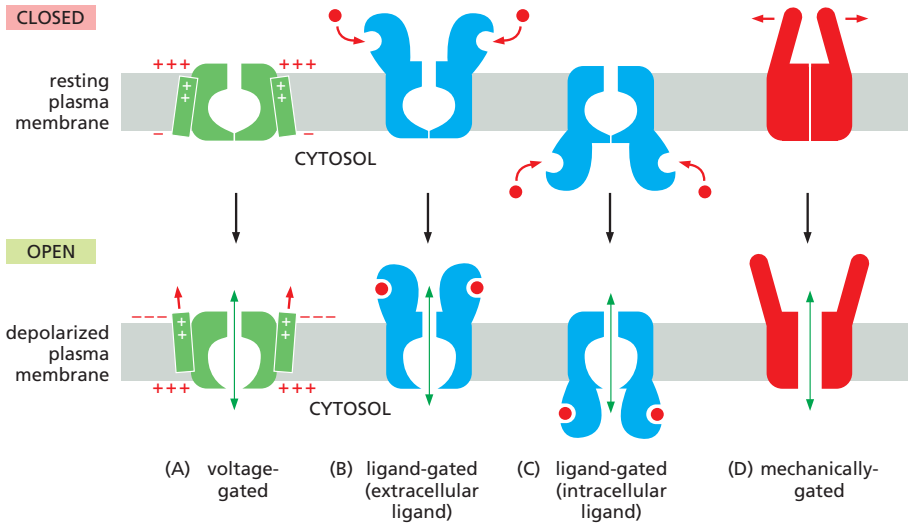


Figure 12–27 Different types of gated ion channels respond to different types of stimuli. Depending on the type of channel, the probability of gate opening is controlled by (A) a change in the voltage difference across the membrane, (B) the binding of a chemical ligand to the extracellular face of a channel, (C) ligand binding to the intracellular face of a channel, or (D) mechanical stress. In the case of the voltage-gated channels, positively charged amino acids (*white* plus signs) in the channel's voltage sensor domains become attracted to negative charges on the extracellular surface of the depolarized plasma membrane, pulling the channel into its open conformation.

indicates that the channel has moving parts and is snapping back and forth between open and closed conformations as the channel is knocked from one conformation to the other by the random thermal movements of the molecules in its environment. Patch-clamp recording was the first technique that could detect such conformational changes, and the picture it paints—of a jerky piece of machinery subjected to constant external buffeting—is now known to apply also to other proteins with moving parts.

The activity of each ion channel is very much "all-or-none": when an ion channel is open, it is fully open; when it is closed, it is fully closed. That raises a fundamental question: If ion channels randomly snap between open and closed conformations even when conditions on each side of the membrane are held constant, how can their state be regulated by conditions inside or outside the cell? The answer is that when the appropriate conditions change, the random behavior continues but with a greatly changed bias: if the altered conditions tend to open the channel, for example, the channel will now spend a much greater proportion of its time in the open conformation, although it will not remain open continuously (see Figure 12–26).

Different Types of Stimuli Influence the Opening and Closing of Ion Channels

There are more than a hundred types of ion channels, and even simple organisms can possess many different types. The human genome contains 80 genes that encode different but related K^+ channels alone. Ion channels differ from one another primarily with respect to their *ion selectivity*—the type of ions they allow to pass—and their *gating*—the conditions that influence their opening and closing. For a **voltage-gated channel**, the probability of being open is controlled by the membrane potential (**Figure 12–27A**). For a **ligand-gated channel**, opening is controlled by the binding of some molecule (a ligand) to the channel (**Figure 12–27B and C**). For a **mechanically-gated channel**, opening is controlled by a mechanical force applied to the channel (**Figure 12–27D**).

The *auditory hair* cells in the ear are an important example of cells that depend on mechanically-gated channels. Sound vibrations pull the channels open, causing ions to flow into the hair cells; this ion flow sets up an electrical signal that is transmitted from the hair cell to the auditory nerve, which then conveys the signal to the brain (**Figure 12–28**).

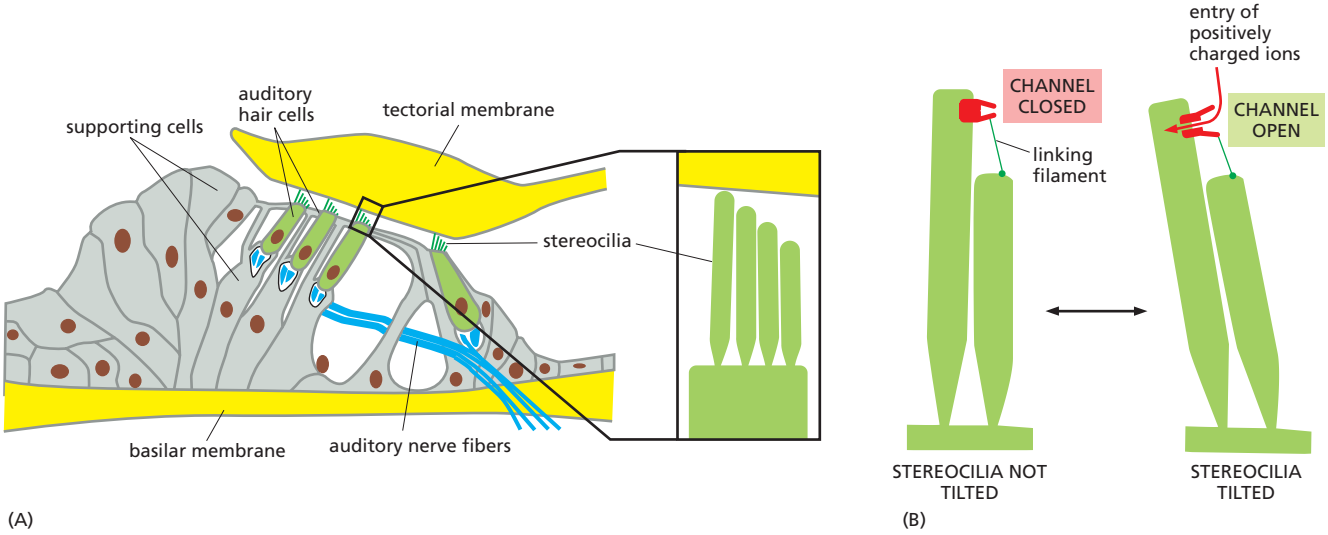


Figure 12–28 Mechanically-gated ion channels allow us to hear. (A) A section through the organ of Corti, which runs the length of the cochlea, the auditory portion of the inner ear. Each auditory hair cell has a tuft of spiky extensions called stereocilia projecting from its upper surface. The hair cells are embedded in an epithelial sheet of supporting cells, which is sandwiched between the *basilar membrane* below and the *tectorial membrane* above. (These are not lipid bilayer membranes but sheets of extracellular matrix.) (B) Sound vibrations cause the basilar membrane to vibrate up and down, causing the stereocilia to tilt. Each stereocilium in the staggered array of stereocilia on a hair cell is attached to the next, shorter stereocilium by a fine filament. The tilting stretches the filaments, which pull open mechanically-gated ion channels in the stereocilium plasma membrane, allowing positively charged ions to enter from the surrounding fluid (**Movie 12.7**). The influx of ions activates the hair cells, which stimulate underlying nerve endings of the auditory nerve fibers that relay the auditory signal to the brain.

The hair-cell mechanism is astonishingly sensitive: the faintest sounds we can hear have been estimated to stretch the filaments by an average of about 0.04 nm, which is less than the diameter of a hydrogen ion (**Movie 12.8**).

Voltage-gated Ion Channels Respond to the Membrane Potential

Voltage-gated ion channels play a major role in propagating electrical signals along all nerve cell extensions, such as those that relay signals from our brain to our toe muscles. But voltage-gated ion channels are present in many other cell types, too, including muscle cells, egg cells, protozoans, and even plant cells, where they enable electrical signals to travel from one part of the plant to another, as in the leaf-closing response of a *Mimosa pudica* plant (**Figure 12–29**).

Voltage-gated ion channels have domains called *voltage sensors* that are extremely sensitive to changes in the membrane potential: changes

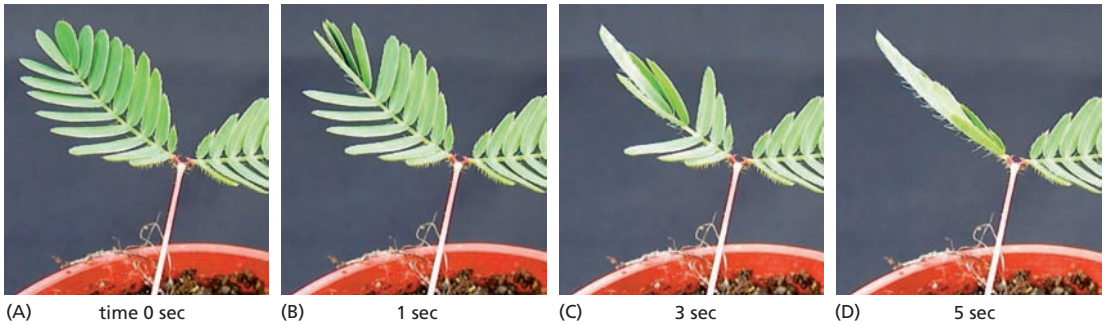
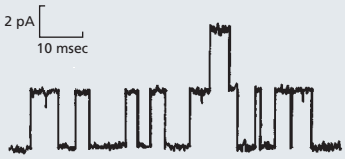


Figure 12–29 Both mechanically-gated and voltage-gated ion channels underlie the leaf-closing response in the touch-sensitive plant *Mimosa pudica*. (A) Resting leaf. (B–D) Successive leaflet closures in response to touch. A few seconds after the leaf on the left is touched, its leaflets snap shut. The response involves the opening of mechanically-gated ion channels in touch-sensitive sensory cells, which then pass a signal to cells containing voltage-gated ion channels, generating an electric impulse. When the impulse reaches specialized hinge cells at the base of each leaflet, a rapid loss of water by these cells occurs, causing the leaflets to fold into a closed conformation suddenly and progressively down the leaf stalk (**Movie 12.9**).

QUESTION 12–3



The figure above shows a recording from a patch-clamp experiment in which the electrical current passing across a patch of membrane is measured as a function of time. The membrane patch was plucked from the plasma membrane of a muscle cell by the technique shown in Figure 12–25 and contains molecules of the acetylcholine receptor, which is a ligand-gated cation channel that is opened by the binding of acetylcholine to the extracellular face of the channel. To obtain a recording, acetylcholine was added to the solution inside the microelectrode. (A) Describe what you can learn about the channels from this recording. (B) How would the recording differ if acetylcholine were (i) omitted or (ii) added to the solution outside the microelectrode only?

above a certain threshold value exert sufficient electrical force on these domains to encourage the channel to switch from its closed to its open conformation (see Figure 12–27A). As discussed earlier, a change in the membrane potential does not affect how wide the channel is open, but instead alters the probability that it will open. Thus, in a large patch of membrane containing many molecules of the channel protein, one might find that on average 10% of them are open at any instant when the membrane is at one potential, whereas 90% are open after this potential changes.

When one type of voltage-gated ion channel opens, the membrane potential of the cell can change. This in turn can activate or inactivate other voltage-gated ion channels. Such circuits, which couple the opening of ion channels to changes in membrane potential to the opening of additional ion channels, are fundamental to all electrical signaling in cells. In the next section, we consider the special case of nerve cells: they—more than any other cell type—have made a profession of electrical signaling, and they employ ion channels in very sophisticated ways.

ION CHANNELS AND NERVE CELL SIGNALING

The fundamental task of a nerve cell, or **neuron**, is to receive, integrate, and transmit signals. Neurons carry signals from sense organs, such as eyes and ears, to the *central nervous system*—the brain and spinal cord. In the central nervous system, neurons signal from one to another through networks of enormous complexity, allowing the brain and spinal cord to analyze, interpret, and respond to the signals coming in from the sense organs.

Every neuron consists of a *cell body*, which contains the nucleus and has a number of long, thin extensions radiating outward from it. Usually, a neuron has one long extension called an **axon**, which conducts electrical signals away from the cell body toward distant target cells; it also usually has several shorter, branching extensions called **dendrites**, which radiate from the cell body like antennae and provide an enlarged surface area to receive signals from the axons of other neurons (**Figure 12–30**). The axon commonly divides at its far end into many branches, each of which ends in a **nerve terminal**, so that the neuron's message can be passed simultaneously to many target cells—muscle or gland cells or other neurons. Likewise, the branching of the dendrites can be extensive, in some cases sufficient to receive as many as 100,000 inputs on a single neuron (see Figure 12–43A).

No matter what the meaning of the signal a neuron carries—whether it is visual information from the eye, a motor command to a muscle, or one

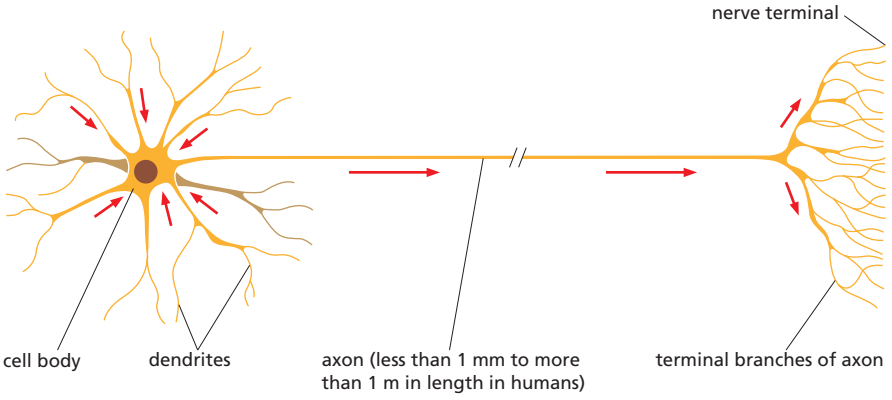


Figure 12–30 A typical neuron has a cell body, a single axon, and multiple dendrites. The axon conducts electrical signals away from the cell body toward its target cells, while the multiple dendrites receive signals from the axons of other neurons. The *red* arrows indicate the direction in which signals travel. During brain development, neurons probe their environment for guidance clues to extend axons and dendrites in appropriate directions to make useful connections (**Movies 12.10 and 12.11**).

Figure 12–31 The squid *Loligo* has a nervous system that is adept at responding rapidly to threats in the animal's environment. Among the nerve cells that make up this escape system is one that possesses a "giant axon," with a very large diameter. Long before patch clamping allowed recordings from single ion channels in small cells (see Figure 12–25), the squid giant axon was routinely used to record and study action potentials. (NOAA.)

step in a complex network of neural processing in the brain—the form of the signal is always the same: it consists of changes in the electrical potential across the neuron's plasma membrane.

Action Potentials Allow Rapid Long-Distance Communication Along Axons

A neuron is stimulated by a signal—typically from another neuron—delivered to a localized site on its surface. This signal initiates a change in the membrane potential at that site. To transmit the signal onward, this local change in membrane potential has to spread from this initial site, which is usually on a dendrite or the cell body, to the axon terminals. There, the signal is relayed to the next cells in the pathway—forming a *neural circuit*. The distances covered by such circuits can be substantial: a signal that leaves a motor neuron in your spinal cord may have to travel a meter or more before it reaches a muscle in your foot.

The local change in membrane potential generated by a signal will spread passively along an axon or a dendrite to adjacent regions of the plasma membrane. Over long distances, such *passive spread* is inadequate, as the signal rapidly becomes weaker with increasing distance from the source. Neurons solve this long-distance communication problem by employing an active signaling mechanism. In this case, a local electrical stimulus of sufficient strength triggers a burst of electrical activity in the plasma membrane that propagates rapidly along the membrane of the axon, continuously renewing itself all along the way. This traveling wave of electrical excitation, known as an **action potential**, or a *nerve impulse*, can carry a message, without weakening, all the way from one end of a neuron to the other, at speeds of up to 100 meters per second.

The early research that established this mechanism of electrical signaling along axons was done on the giant axon of the squid (**Figure 12–31**). This axon has such a large diameter that it is possible to record its electrical activity from an electrode inserted directly into it (**How We Know**, pp. 412–413). From such studies, it was deduced how action potentials are the direct consequence of the properties of voltage-gated ion channels in the axonal plasma membrane, as we now explain.

Action Potentials Are Mediated by Voltage-gated Cation Channels

When a neuron is stimulated, the membrane potential of the plasma membrane shifts to a less negative value (that is, toward zero). If this **depolarization** is sufficiently large, it will cause **voltage-gated Na^+ channels** in the membrane to open transiently at the site. As these channels flicker open, they allow a small amount of Na^+ to enter the cell down its steep electrochemical gradient. The influx of positive charge depolarizes the membrane further (that is, it makes the membrane potential even less negative), thereby opening additional voltage-gated Na^+ channels and causing still further depolarization. This process continues in an explosive, self-amplifying fashion until, within about a millisecond, the membrane potential in the local region of the neuron's plasma

QUESTION 12–4

Using the Nernst equation and the ion concentrations given in Table 12–1 (p. 391), calculate the equilibrium membrane potential of K^+ and Na^+—that is, the membrane potential where there would be no net movement of the ion across the plasma membrane (assume that the concentration of intracellular Na^+ is 10 mM). What membrane potential would you predict in a resting animal cell? Explain your answer. What would happen if a large number of Na^+ channels suddenly opened, making the membrane much more permeable to Na^+ than to K^+? (Note that because few ions need to move across the membrane to change drastically the charge distribution across that membrane, you can safely assume that the ion concentrations on either side of the membrane do not change significantly.) What would you predict would happen next if the Na^+ channels closed again?

HOW WE KNOW

SQUID REVEAL SECRETS OF MEMBRANE EXCITABILITY

Each spring, *Loligo pealei* migrate to the shallow waters off Cape Cod on the eastern coast of the United States. There they spawn, launching the next generation of squid. But more than just meeting and breeding, these animals provide neuroscientists summering at the Marine Biological Laboratory in Woods Hole, Massachusetts, with a golden opportunity to study the mechanism of electrical signaling along nerve axons.

Like most animals, squid survive by catching prey and escaping predators. Fast reflexes and an ability to accelerate rapidly and make sudden changes in swimming direction help them avoid danger while chasing down a decent meal. Squid derive their speed and agility from a specialized biological jet propulsion system: they draw water into their mantle cavity and then contract their muscular body wall to expel the collected water rapidly through a tubular siphon, thus propelling themselves through the water.

Controlling such quick and coordinated muscle contraction requires a nervous system that can convey signals with great speed down the length of the animal's body. Indeed, *Loligo pealei* possesses some of the largest nerve cell axons found in nature. Squid giant axons can reach 10 cm in length and are over 100 times the diameter of a mammalian axon—about the width of a pencil lead. Generally speaking, the larger the diameter of an axon, the more rapidly signals can travel along its length.

In the 1930s, scientists first started to take advantage of the squid giant axon for studying the electrophysiology of the nerve cell. Because of its relatively large size, an investigator can isolate an individual axon and insert an electrode into it to measure the axon's membrane potential and monitor its electrical activity. This experimental system allowed researchers to address a variety of questions, including which ions are important for establishing the resting membrane potential and for initiating and propagating an action potential, and how changes in the membrane potential control ion permeability.

Set-up for action

Because the squid axon is so long and wide, an electrode made from a glass capillary tube containing a conducting solution can be thrust down the axis of the isolated axon so that its tip lies deep in the cytoplasm. This set-up allowed investigators to measure the voltage difference between the inside and the outside of the axon—that is, the membrane potential—as an action potential sweeps past the tip of the electrode (**Figure 12–32**). The action potential itself would be triggered by applying a brief electrical stimulus to one end of the axon. It didn't matter which end was stimulated, as the action potential could travel in either direction; it also didn't matter how big the stimulus was, as long as it exceeded a certain threshold (see Figure 12–35), indicating that an action potential is an "all or nothing" response.

Once researchers could reliably generate and measure an action potential, they could use the preparation to answer other questions about membrane excitability. For example, which ions are critical for an action potential? The three most plentiful ions, both inside and outside an axon, are Na^+, K^+, and Cl^-. Do they have

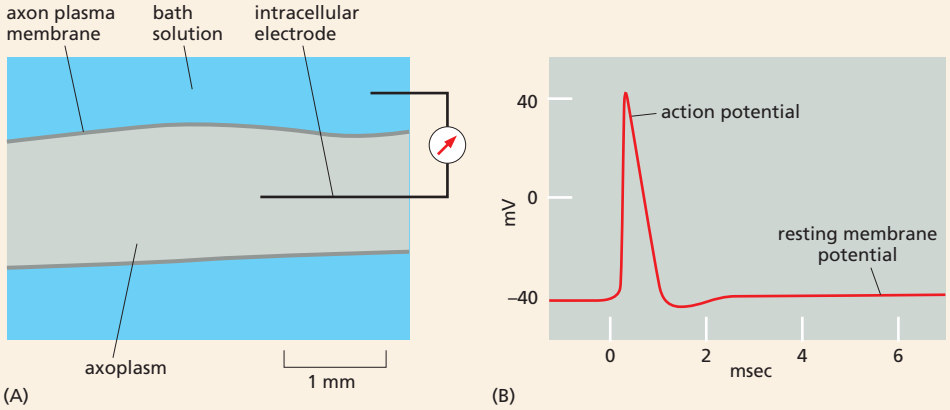


Figure 12–32 Scientists can study nerve cell excitability using an isolated axon from squid. (A) An electrode can be inserted into the cytoplasm (axoplasm) of a squid giant axon to (B) measure the resting membrane potential and monitor action potentials induced when the axon is electrically stimulated.

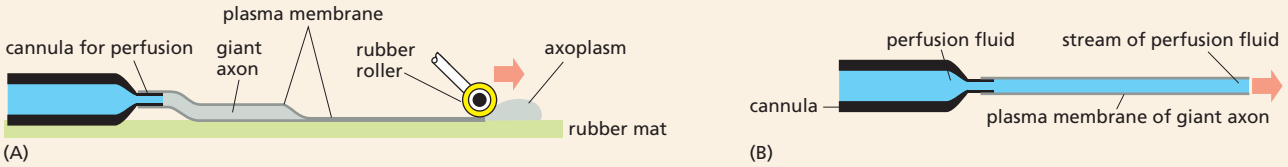


Figure 12–33 The cytoplasm in a squid axon can be removed and replaced with an artificial solution of pure ions. (A) The axon cytoplasm (axoplasm) is extruded using a rubber roller. (B) A perfusion fluid containing the desired concentration of ions is pumped gently through the emptied-out axon.

equal importance when it comes to the action potential? Because the squid axon is so large and strong, investigators could extrude the cytoplasm from the axon like toothpaste from a tube (**Figure 12–33A**). The emptied-out axon could then be reinflated by filling it with a pure solution of Na^+, K^+, or Cl^- (**Figure 12–33B**). Thus, the ions inside the axon and in the bath solution could be varied independently (see Figure 12–32A). Using this set-up, the researchers discovered that the axon would generate a normal action potential if, and only if, the concentrations of Na^+ and K^+ approximated the natural concentrations found inside and outside the cell. Thus, they concluded that the cell components crucial to the action potential are the plasma membrane, Na^+ and K^+ ions, and the energy provided by the concentration gradients of these ions across the membrane; all other components, including other sources of metabolic energy, were presumably removed when the axon was emptied and refilled.

Channel traffic

Once Na^+ and K^+ had been singled out as critical for an action potential, the questions then became: What does each of these ions contribute to the action potential? How permeable is the membrane to each, and how does the membrane permeability change as an action potential sweeps by? Again, the squid giant axon provided some answers. The concentrations of Na^+ and K^+ inside and outside the axon could be altered, and the effects of these changes on the membrane potential could be measured directly. From such studies, it was determined that, at rest, the membrane potential of an axon is close to the equilibrium potential for K^+: when the external concentration of K^+ was varied, the resting potential of the axon changed roughly in accordance with the Nernst equation (see Figure 12–24). The results suggested that at rest, the membrane is chiefly permeable to K^+; we now know that K^+ leak channels provide the main pathway that these ions can take through the resting plasma membrane.

The situation for Na^+ is very different. When the external concentration of Na^+ was varied, there was no effect on the resting potential of the axon. However, the height of the peak of the action potential varied with the concentration of Na^+ outside the axon (**Figure 12–34**). During the action potential, therefore, the membrane appeared to be chiefly permeable to Na^+, presumably as the result of the opening of Na^+ channels. In the aftermath of the action potential, the Na^+ permeability decreased and the membrane potential reverted to a negative value, which depended on the external concentration of K^+. As the membrane lost its permeability to Na^+, it became even more permeable to K^+ than before, presumably because additional K^+ channels opened, accelerating the resetting of the membrane potential to the resting state, and readying the membrane for the next action potential.

These studies on the squid giant axon made an enormous contribution to our understanding of nerve cell excitability, and the researchers who made these discoveries in the 1940s and 1950s—Alan Hodgkin and Andrew Huxley—received a Nobel Prize in 1963. However, it was years before the various ion channel proteins that they had hypothesized to exist would be biochemically identified. We now know the three-dimensional structures of many of these channel proteins, allowing us to marvel at the fundamental beauty of these molecular machines.

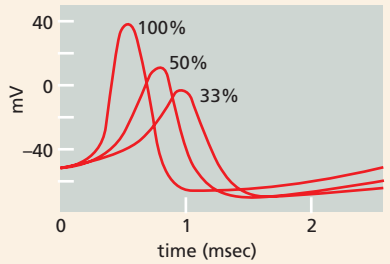


Figure 12–34 The shape of the action potential depends on the concentration of Na^+ outside the squid axon. Shown here are action potentials recorded when the external medium contains 100%, 50%, or 33% of the normal extracellular concentration of Na^+.

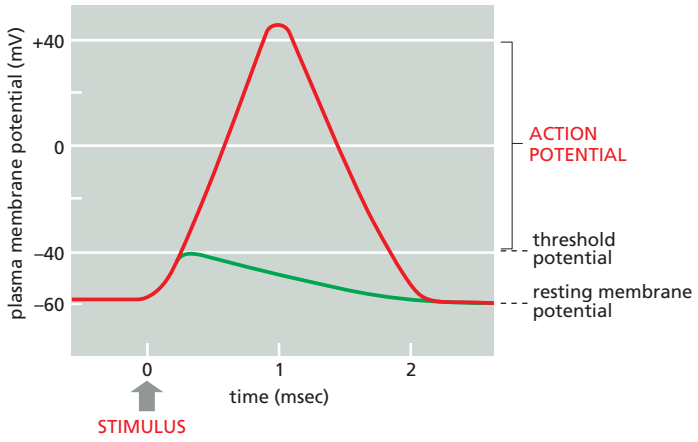


Figure 12–35 An action potential is triggered by a depolarization of a neuron's plasma membrane. The resting membrane potential in this neuron is –60 mV, and a stimulus that depolarizes the plasma membrane to about –40 mV (the threshold potential) is applied. This depolarizing stimulus is sufficient to open voltage-gated Na^+ channels in the membrane and thereby trigger an action potential. As the membrane rapidly depolarizes further, the membrane potential (*red* curve) swings past zero, reaching +40 mV before it returns to its resting negative value as the action potential terminates. The *green* curve shows how the membrane potential would simply have relaxed back to the resting value after the initial depolarizing stimulus if there had been no amplification by voltage-gated ion channels in the plasma membrane.

membrane has shifted from its resting value of about –60 mV to about +40 mV (**Figure 12–35**).

The voltage of +40 mV is close to the membrane potential at which the electrochemical driving force for movement of Na^+ across the membrane is zero—that is, the effects of the membrane potential and the concentration gradient for Na^+ are equal and opposite; therefore Na^+ has no further tendency to enter or leave the cell.

If these voltage-gated channels continued to respond to the depolarized membrane potential, the cell would get stuck with most of its Na^+ channels open. The cell is saved from this fate because voltage-gated Na^+ channels have an automatic inactivating mechanism—a kind of "timer" that causes them to rapidly adopt (within a millisecond or so) a special inactivated conformation in which the channel is closed, even though the membrane is still depolarized. The Na^+ channels remain in this *inactivated state* until the membrane potential has returned to its resting, negative value. A schematic illustration of these three distinct states of the voltage-gated Na^+ channel—*closed*, *open*, and *inactivated*—is shown in **Figure 12–36**. How they contribute to the rise and fall of an action potential is shown in **Figure 12–37**.

During an action potential, voltage-gated Na^+ channels do not act alone. The depolarized axonal membrane is helped to return to its resting potential by the opening of *voltage-gated K^+ channels*. These also open in response to depolarization, but not as promptly as the Na^+ channels, and they stay open as long as the membrane remains depolarized. As the local depolarization reaches its peak, K^+ ions (carrying positive charge) therefore start to flow out of the cell, down their electrochemical gradient,

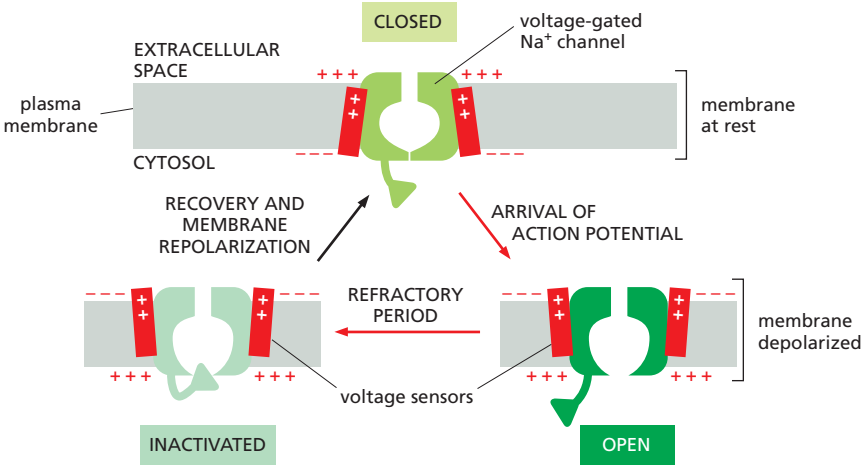


Figure 12–36 A voltage-gated Na^+ channel can flip from one conformation to another, depending on the membrane potential. When the membrane is at rest and highly polarized, positively charged amino acids in the voltage sensors of the channel (*red* bars) are oriented by the membrane potential in a way that keeps the channel in its closed conformation. When the membrane is depolarized, the voltage sensors shift, changing the channel's conformation so the channel has a high probability of opening. But in the depolarized membrane, the inactivated conformation is even more stable than the open conformation, and so, after a brief period spent in the open conformation, the channel becomes temporarily inactivated and cannot open. The *red* arrows indicate the sequence that follows a sudden depolarization, and the *black* arrow indicates the return to the original conformation after the membrane has repolarized.

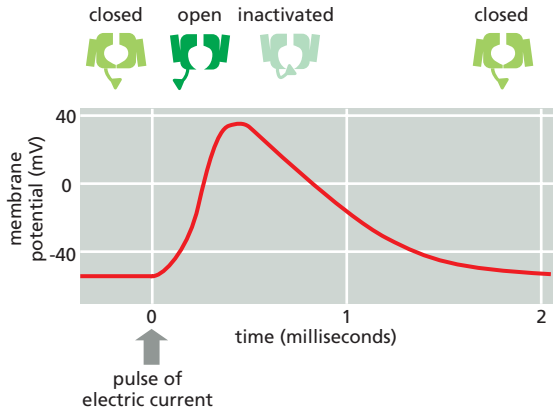


Figure 12–37 Voltage-gated Na^+ channels change their conformation during an action potential. In this example, the action potential is triggered by a brief pulse of electric current (arrow), which partially depolarizes the membrane, as shown in the plot of membrane potential versus time. The course of the action potential reflects the opening and subsequent inactivation of voltage-gated Na^+ channels, as shown (*top*). Even if restimulated, the plasma membrane cannot produce a second action potential until the Na^+ channels have returned from the inactivated to the closed conformation (see Figure 12–36). Until then, the membrane is resistant, or refractory, to stimulation.

through these newly opened K^+ channels—temporarily unhindered by the negative membrane potential that normally restrains them in the resting cell. The rapid outflow of K^+ through the voltage-gated K^+ channels brings the membrane back to its resting state much more quickly than could be achieved by K^+ outflow through the K^+ leak channels alone.

Once it begins, the self-amplifying depolarization of a small patch of plasma membrane quickly spreads outward: the Na^+ flowing in through open Na^+ channels begins to depolarize the neighboring region of the membrane, which then goes through the same self-amplifying cycle. In this way, an action potential spreads outward as a traveling wave from the initial site of depolarization, eventually reaching the axon terminals (**Figure 12–38**).

QUESTION 12–5

Explain as precisely as you can, but in no more than 100 words, the ionic basis of an action potential and how it is passed along an axon.

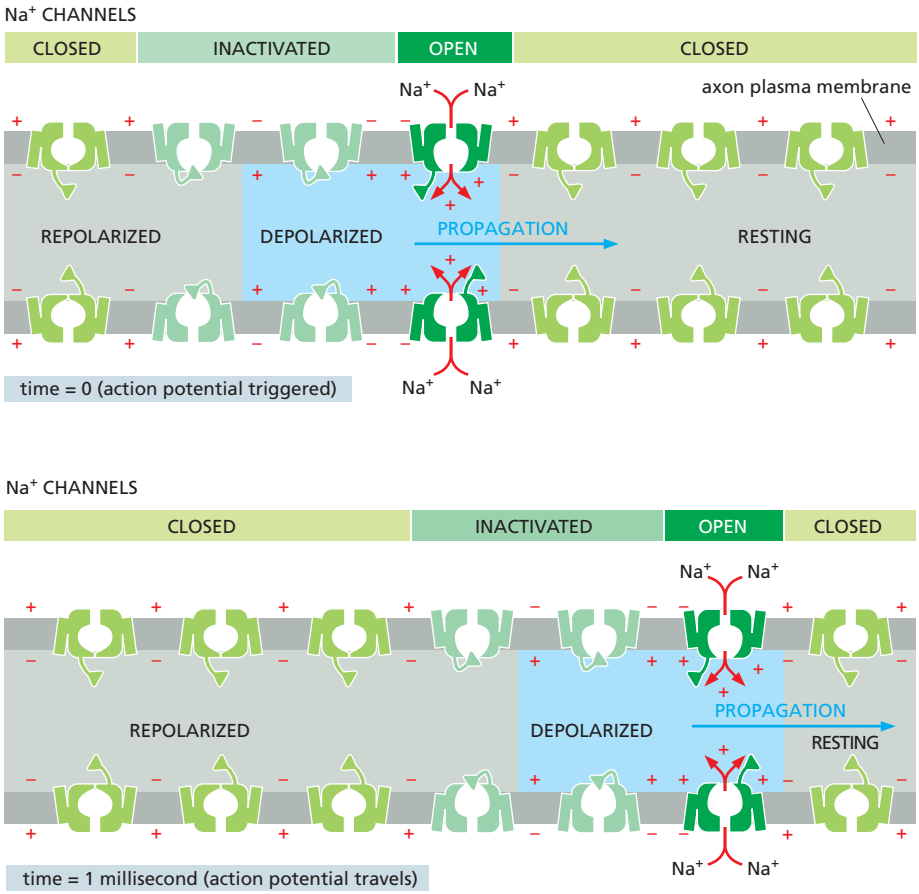


Figure 12–38 An action potential propagates along the length of an axon. The changes in the Na^+ channels and the consequent flow of Na^+ across the membrane (*red* arrows) alters the membrane potential and gives rise to the traveling action potential, as shown here and in **Movie 12.12**. The region of the axon with a depolarized membrane is shaded in *blue*. Note that an action potential can only travel forward; that is, away from the site of depolarization. This is because Na^+ channel inactivation in the aftermath of an action potential prevents the advancing front of depolarization from spreading backward (see also Figure 12–37).

Once an action potential has passed, Na^+ pumps in the axon plasma membrane labor to restore the Na^+ and K^+ ion gradients to their levels in the resting cell. The human brain consumes 20% of the total energy generated from the metabolism of food, mostly to power these pumps.

Voltage-gated Ca^{2+} Channels in Nerve Terminals Convert an Electrical Signal into a Chemical Signal

When an action potential reaches the *nerve terminals* at the end of an axon, the signal must somehow be relayed to the *target cells* that the terminals contact—usually neurons or muscle cells. The signal is transmitted to the target cells at specialized junctions known as **synapses**. At most synapses, the plasma membranes of the cells transmitting and receiving the message—the *presynaptic* and the *postsynaptic* cells, respectively—are separated from each other by a narrow *synaptic cleft* (typically 20 nm across), which the electrical signal cannot cross. To transmit the message across this gap, the electrical signal is converted into a chemical signal, in the form of a small, secreted signal molecule called a **neurotransmitter**. Neurotransmitters are stored in the nerve terminals within membrane-enclosed **synaptic vesicles** (Figure 12–39).

When an action potential reaches the nerve terminal, some of the synaptic vesicles fuse with the plasma membrane, releasing their neurotransmitter into the synaptic cleft. This link between the arrival of an action potential and the secretion of neurotransmitter involves the activation of yet another type of voltage-gated cation channel: *voltage-gated Ca^{2+} channels* located in the plasma membrane of the presynaptic nerve terminal. Because the Ca^{2+} concentration outside the nerve terminal is more than 1000 times greater than the free Ca^{2+} concentration in its cytosol (see Table 12–1), Ca^{2+} rushes into the nerve terminal through the open channels. The resulting increase in Ca^{2+} concentration in the cytosol of the terminal immediately triggers the fusion of synaptic vesicles with the plasma membrane, which releases the neurotransmitter into the synaptic cleft. Thanks to these voltage-gated Ca^{2+} channels, the electrical signal has now been converted into a chemical signal (Figure 12–40).

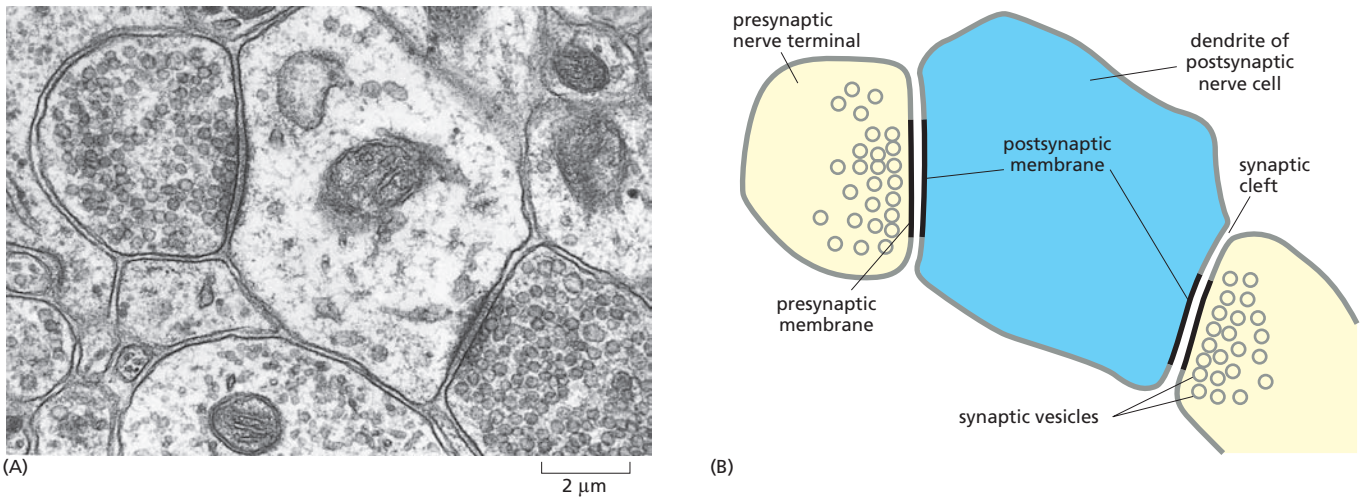


Figure 12–39 Neurons connect to their target cells at synapses. (A) An electron micrograph and (B) a drawing of a cross section of two nerve terminals (*yellow*) forming synapses on a single nerve cell dendrite (*blue*) in the mammalian brain. Neurotransmitters carry the signal across the synaptic cleft that separates the presynaptic and postsynaptic cells. The neurotransmitter in the presynaptic terminal is contained within synaptic vesicles, which release neurotransmitter into the synaptic cleft. Note that both the presynaptic and postsynaptic membranes are thickened and highly specialized at the synapse. (A, courtesy of Cedric Raine.)

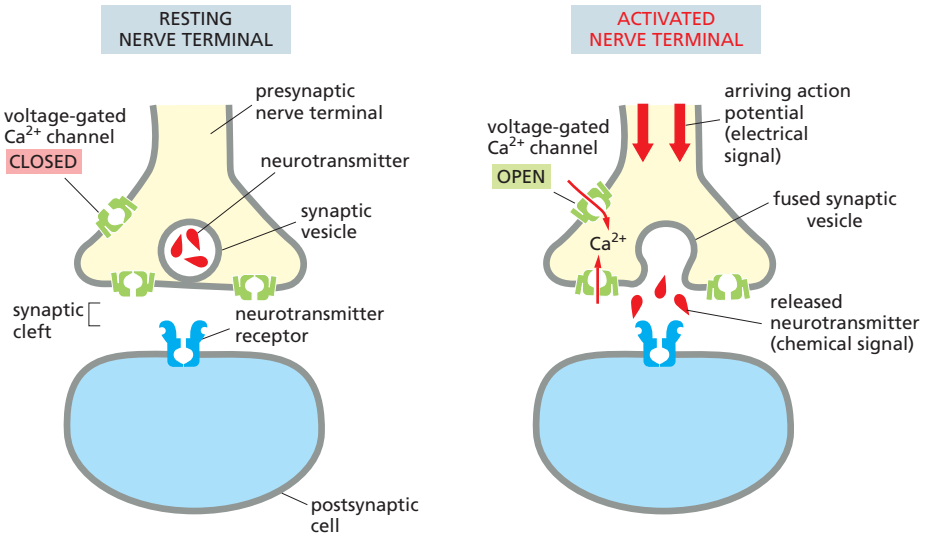


Figure 12–40 An electrical signal is converted into a secreted chemical signal at a nerve terminal. When an action potential reaches a nerve terminal, it opens voltage-gated Ca^{2+} channels in the plasma membrane, allowing Ca^{2+} to flow into the terminal. The increased Ca^{2+} in the nerve terminal stimulates the synaptic vesicles to fuse with the plasma membrane, releasing their neurotransmitter into the synaptic cleft—a process called exocytosis (discussed in Chapter 15).

Transmitter-gated Ion Channels in the Postsynaptic Membrane Convert the Chemical Signal Back into an Electrical Signal

The released neurotransmitter rapidly diffuses across the synaptic cleft and binds to *neurotransmitter receptors* concentrated in the plasma membrane of the postsynaptic target cell. Once released, neurotransmitters are rapidly removed from the synaptic cleft—either by enzymes that destroy them or by pumps that return them to the nerve terminal or that transport them into neighboring non-neuronal cells. This rapid removal of the neurotransmitter limits the duration and spread of the signal and ensures that when the presynaptic cell falls quiet, the postsynaptic cell will do the same.

Neurotransmitter receptors can be of various types; some mediate relatively slow effects in the target cell, whereas others trigger more rapid responses. Rapid responses—on a time scale of milliseconds—depend on receptors that are **transmitter-gated ion channels** (also called ion-channel-coupled receptors). These constitute a subclass of ligand-gated ion channels (see Figure 12–27B), and their function is to convert the chemical signal carried by a neurotransmitter back into an electrical signal. The channels open transiently in response to the binding of the neurotransmitter, thus changing the ion permeability of the postsynaptic membrane. This in turn causes a change in the membrane potential (**Figure 12–41**).

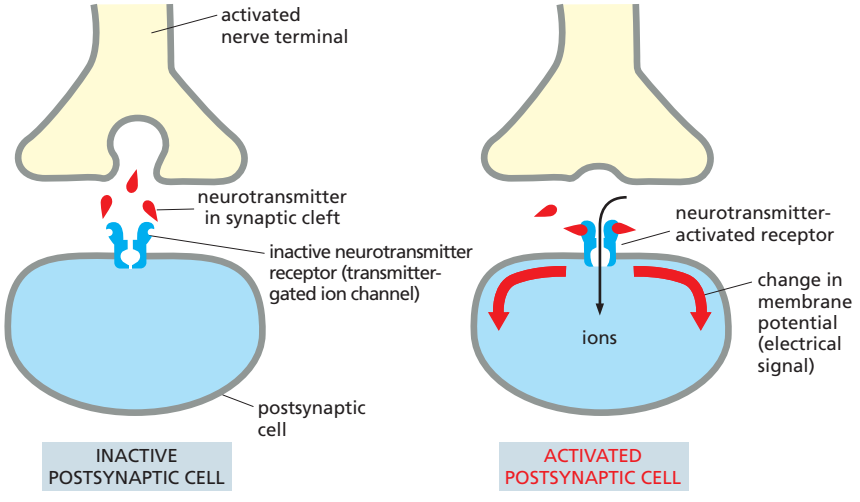


Figure 12–41 A chemical signal is converted into an electrical signal by postsynaptic transmitter-gated ion channels at a synapse. The released neurotransmitter binds to and opens the transmitter-gated ion channels in the plasma membrane of the postsynaptic cell. The resulting ion flows alter the membrane potential of the postsynaptic cell, thereby converting the chemical signal back into an electrical one (**Movie 12.13**).

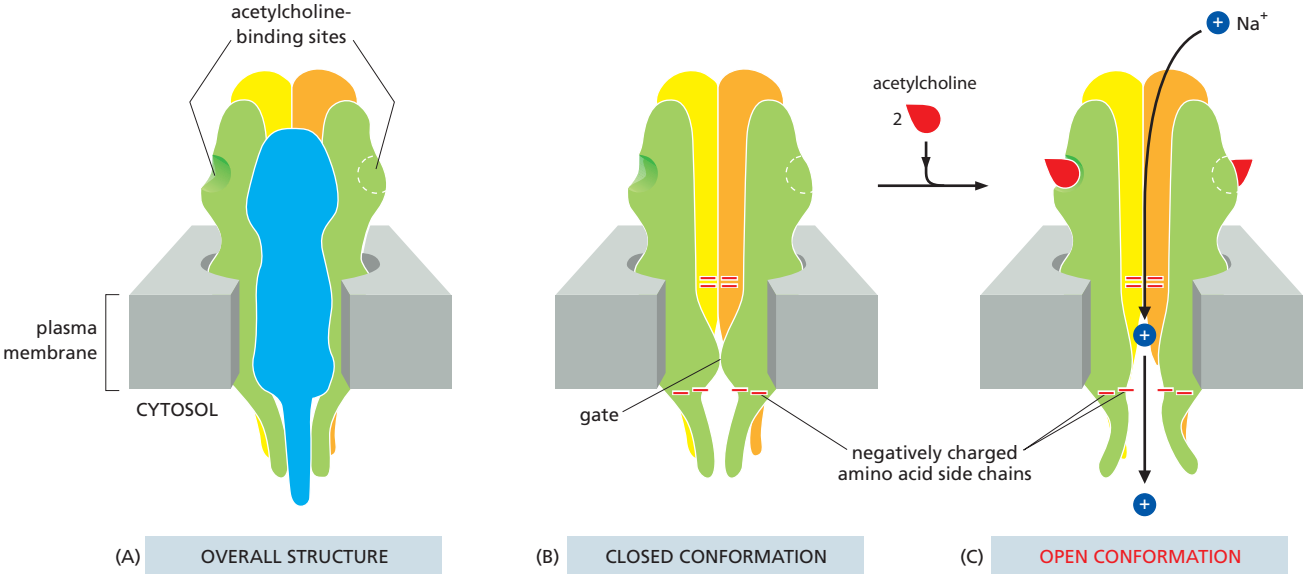


Figure 12–42 The acetylcholine receptor in the plasma membrane of vertebrate skeletal muscle cells opens when it binds the neurotransmitter acetylcholine. (A) This transmitter-gated ion channel is composed of five transmembrane protein subunits, two of which (*green*) are identical. The subunits combine to form a transmitter-gated aqueous pore across the lipid bilayer. There are two acetylcholine-binding sites, one formed by parts of a *green* and *blue* subunit, the other by parts of a *green* and *orange* subunit, as shown. (B) The closed conformation. The *blue* subunit has been removed here and in (C) to show the interior of the pore. Negatively charged amino acid side chains at either end of the pore (indicated here by *red* minus signs) ensure that only positively charged ions, mainly Na^+ and K^+, can pass. But when acetylcholine is not bound and the channel is in its closed conformation, the pore is occluded (blocked) by hydrophobic amino acid side chains in the region called the gate. (C) The open conformation. When acetylcholine, released by a motor neuron, binds to both binding sites, the channel undergoes a conformational change; the hydrophobic side chains move apart and the gate opens, allowing Na^+ to flow across the membrane down its electrochemical gradient, depolarizing the membrane. Even with acetylcholine bound, the channel flickers randomly between the open and closed states (see Figure 12–26); without acetylcholine bound, the channel rarely opens.

QUESTION 12–6

In the disease myasthenia gravis, the human body makes—by mistake—antibodies to its own acetylcholine receptor molecules. These antibodies bind to and inactivate acetylcholine receptors on the plasma membrane of muscle cells. The disease leads to a devastating progressive weakening of the muscles of people affected. Early on, they may have difficulty opening their eyelids, for example, and, in an animal model of the disease, rabbits have difficulty holding their ears up. As the disease progresses, most muscles weaken, and people with myasthenia gravis have difficulty speaking and swallowing. Eventually, impaired breathing can cause death. Explain which step of muscle function is affected.

If the change is large enough, the postsynaptic membrane will depolarize and trigger an action potential in the postsynaptic cell.

A well-studied example of a neurotransmitter in action is found at the *neuromuscular junction*—the specialized synapse formed between a motor neuron and a skeletal muscle cell. In vertebrates, the neurotransmitter *acetylcholine* stimulates muscle contraction by binding to the *acetylcholine receptor*, a transmitter-gated ion channel in the muscle cell's membrane (**Figure 12–42**). However, not all neurotransmitters excite the postsynaptic cell, as we consider next.

Neurotransmitters Can Be Excitatory or Inhibitory

Neurotransmitters can either excite or inhibit a postsynaptic cell, and it is the character of the receptor that recognizes the neurotransmitter that determines how the postsynaptic cell will respond. The chief receptors for excitatory neurotransmitters, such as *acetylcholine* and *glutamate*, are ligand-gated cation channels. When a neurotransmitter binds, these channels open to allow an influx of Na^+, which depolarizes the plasma membrane and thus tends to activate the postsynaptic cell, encouraging it to fire an action potential. By contrast, the main receptors for inhibitory neurotransmitters, such as *γ-aminobutyric acid* (*GABA*) and *glycine*, are ligand-gated Cl^- channels. When neurotransmitters bind, these channels open, allowing Cl^- to enter the cell; this influx of Cl^- inhibits the postsynaptic cell by making its plasma membrane harder to depolarize.

TABLE 12–3 SOME EXAMPLES OF ION CHANNELS

Ion Channel	Typical Location	Function
K^+ leak channel	plasma membrane of most animal cells	maintenance of resting membrane potential
Voltage-gated Na^+ channel	plasma membrane of nerve cell axon	generation of action potentials
Voltage-gated K^+ channel	plasma membrane of nerve cell axon	return of membrane to resting potential after initiation of an action potential
Voltage-gated Ca^{2+} channel	plasma membrane of nerve terminal	stimulation of neurotransmitter release
Acetylcholine receptor (acetylcholine-gated cation channel)	plasma membrane of muscle cell (at neuromuscular junction)	excitatory synaptic signaling
Glutamate receptor (glutamate-gated cation channel)	plasma membrane of many neurons (at synapses)	excitatory synaptic signaling
GABA receptor (GABA-gated Cl^- channel)	plasma membrane of many neurons (at synapses)	inhibitory synaptic signaling
Glycine receptor (glycine-gated Cl^- channel)	plasma membrane of many neurons (at synapses)	inhibitory synaptic signaling
Mechanically-gated cation channel	auditory hair cell in inner ear	detection of sound vibrations

Toxins that bind to any of these excitatory or inhibitory neurotransmitter receptors can have dramatic effects on an animal—or a human. *Curare*, for example, causes muscle paralysis by blocking excitatory acetylcholine receptors at the neuromuscular junction. This drug was used by South American Indians to make poison arrows and is still used by surgeons to relax muscles during an operation. By contrast, *strychnine*—a common ingredient in rat poisons—causes muscle spasms, convulsions, and death by blocking inhibitory glycine receptors on neurons in the brain and spinal cord.

The locations and functions of the ion channels discussed in this chapter are summarized in **Table 12–3**.

Most Psychoactive Drugs Affect Synaptic Signaling by Binding to Neurotransmitter Receptors

Many drugs used in the treatment of insomnia, anxiety, depression, and schizophrenia act by binding to transmitter-gated ion channels in the brain. Sedatives and tranquilizers such as barbiturates, Valium, Ambien, and Restoril, for example, bind to GABA-gated Cl^- channels. Their binding makes the channels easier to open by GABA, rendering the neuron more sensitive to GABA's inhibitory action. By contrast, the antidepressant Prozac blocks the Na^+-driven symport responsible for the reuptake of the excitatory neurotransmitter *serotonin*, increasing the amount of serotonin available in the synapses that use it. This drug has changed the lives of many people who suffer from depression—although why boosting serotonin can elevate mood is still unknown.

QUESTION 12–7

When an inhibitory neurotransmitter such as GABA opens Cl^- channels in the plasma membrane of a postsynaptic neuron, why does this make it harder for an excitatory neurotransmitter to excite the neuron?

The number of distinct types of neurotransmitter receptors is very large, although they fall into a small number of families. There are, for example, many subtypes of acetylcholine, glutamate, GABA, glycine, and serotonin receptors; they are usually located on different neurons and often differ only subtly in their electrophysiological properties. With such a large variety of receptors, it may be possible to design a new generation of psychoactive drugs that will act more selectively on specific sets of neurons to mitigate the mental illnesses that devastate so many people's lives. One percent of the human population, for example, have schizophrenia, another 1% have bipolar disorder, about 1% have an autistic disorder, and many more suffer from anxiety or depressive disorders. The fact that these disorders are so prevalent suggests that the complexity of synaptic signaling may make the brain especially vulnerable to genetic alterations. But complexity also provides some distinct advantages, as we discuss next.

The Complexity of Synaptic Signaling Enables Us to Think, Act, Learn, and Remember

For a process so critical for animal survival, the mechanism that governs synaptic signaling seems unnecessarily cumbersome, as well as error-prone. For a signal to pass from one neuron to the next, the nerve terminal of the presynaptic cell must convert an electrical signal into a secreted chemical. This chemical signal must then diffuse across the synaptic cleft so that a postsynaptic cell can convert it back into an electric one. Why would evolution have favored such an apparently inefficient and vulnerable method for passing a signal between two cells? It would seem more efficient and robust to have a direct electrical connection between them—or to do away with the synapse altogether and use a single continuous cell.

The value of synapses that rely on secreted chemical signals becomes clear when we consider how they function in the context of the nervous system—an elaborate network of neurons, interconnected by many branching circuits, performing complex computations, storing memories, and generating plans for action. To carry out these functions, neurons have to do more than merely generate and relay signals: they must also combine them, interpret them, and record them. Chemical synapses make these activities possible. A motor neuron in the spinal cord, for example, receives inputs from hundreds or thousands of other neurons that make synapses on it (**Figure 12–43**). Some of these signals tend to

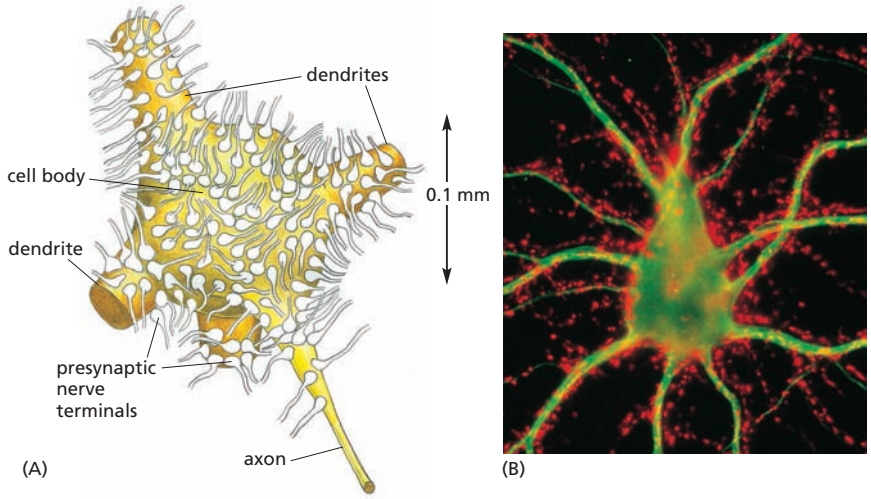


Figure 12–43 Thousands of synapses form on the cell body and dendrites of a motor neuron in the spinal cord. (A) Many thousands of nerve terminals synapse on this neuron, delivering signals from other parts of the animal to control the firing of action potentials along the neuron's axon. (B) A rat nerve cell in culture. Its cell body and dendrites (*green*) are stained with a fluorescent antibody that recognizes a cytoskeletal protein. Thousands of axon terminals (*red*) from other nerve cells (not visible) make synapses on the cell's surface; they are stained with a fluorescent antibody that recognizes a protein in synaptic vesicles, which are located in the nerve terminals (see Figure 12–39). (B, courtesy of Olaf Mundigl and Pietro de Camilli.)

stimulate the neuron, while others inhibit it. The motor neuron has to combine all of the information it receives and react, either by stimulating a muscle to contract or by remaining quiet.

This task of computing an appropriate output from a babble of inputs is achieved by a complicated interplay between different types of ion channels in the neuron's plasma membrane. Each of the hundreds of types of neurons in the brain has its own characteristic set of receptors and ion channels that enables the cell to respond in a particular way to a certain set of inputs and thus to perform its specialized task.

Ion channels are thus critical components of the machinery that enables us to act, think, feel, speak, learn, and remember. Given that these channels operate within neuronal circuits that are dauntingly complex, will we ever be able to deeply understand the molecular mechanisms that direct the complex behaviors of organisms such as ourselves? Although cracking this problem in humans is still far in the future, we now have increasingly powerful ways to study the neural circuits—and molecules—that underlie behavior in experimental animals. One of the most promising techniques makes use of a different type of ion channel, a light-gated ion channel borrowed from unicellular algae, as we now discuss.

Light-gated Ion Channels Can Be Used to Transiently Activate or Inactivate Neurons in Living Animals

Photosynthetic green algae use light-gated channels to sense and navigate toward sunlight. In response to blue light, one of these channels—called *channelrhodopsin*—allows Na^+ to flow into the cell. This depolarizes the plasma membrane and, ultimately, modulates the beating of the flagella that the organism uses to swim. Although these channels are peculiar to unicellular green algae, they function perfectly well when they are artificially transferred into other cell types, thereby rendering the recipient cells responsive to light.

Because nerve cells are also activated by a depolarizing influx of Na^+, as we have discussed (see Figure 12–38), channelrhodopsin can be used to manipulate the activity of neurons and neural circuits—including those in living animals. In one particularly stunning experiment, the channelrhodopsin gene was introduced into a select subpopulation of neurons in the mouse hypothalamus—a brain region involved in many functions, including aggression. The activity of these neurons could then be controlled by light that was provided by a thin, optic fiber implanted in the animal's brain. When the channels were illuminated, the mouse would launch an attack on any object in its path—including other mice or, in one comical instance, an inflated rubber glove. When the light was switched off, the neurons once again fell silent, and the mouse's behavior would immediately return to normal (**Figure 12–44** and **Movie 12.14**).

Because the approach relies on a light-gated channel that is introduced into cells by genetic engineering techniques (discussed in Chapter 10), the method has been dubbed **optogenetics**. This tool is revolutionizing neurobiology, allowing investigators to dissect the neural circuits that govern even the most complex behaviors in a variety of experimental animals, from fruit flies to monkeys. But its implications extend beyond the laboratory. As genetic studies continue to identify genes associated with various human neurological and psychiatric disorders, the ability to exploit light-gated ion channels to study where and how these genes function in model organisms promises to greatly advance our understanding of the molecular and cellular basis of our own behavior.

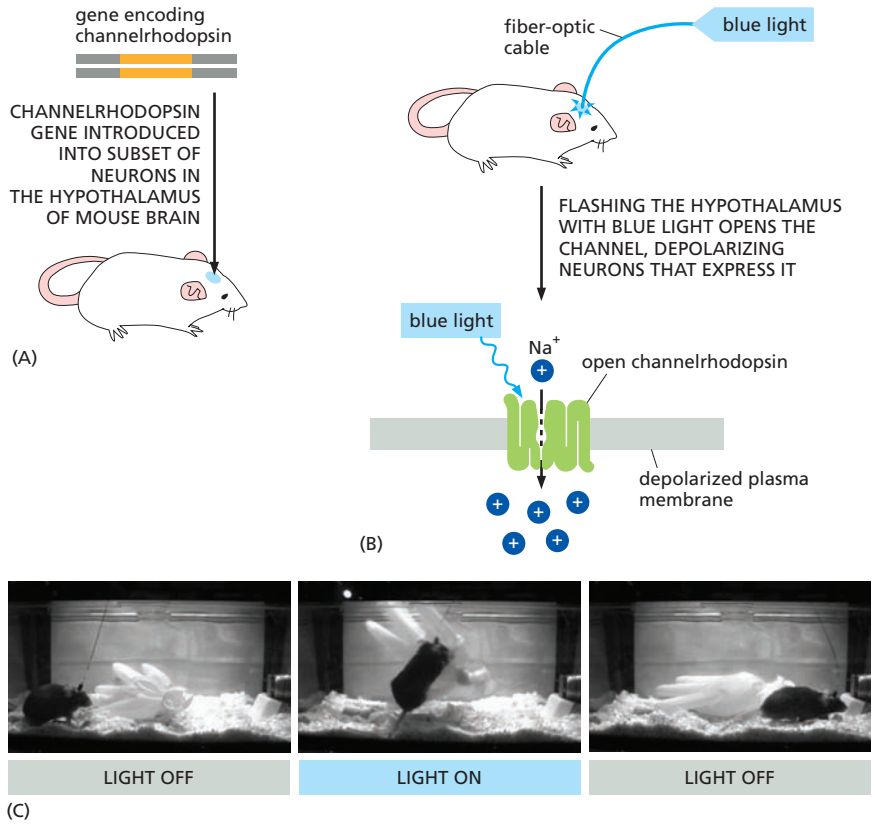


Figure 12–44 Light-gated ion channels can control the activity of specific neurons in a living animal. (A) In this experiment, the gene encoding channelrhodopsin was introduced into a subset of neurons in the mouse hypothalamus. (B) When the neurons are exposed to blue light using a tiny fiber-optic cable implanted into the animal's brain, channelrhodopsin opens, depolarizing and stimulating the channel-containing neurons. (C) When the light is switched on, the mouse immediately becomes aggressive; when the light is switched off, its behavior immediately returns to normal. (C, from D. Lin et al., *Nature* 470:221–226, 2011.)

ESSENTIAL CONCEPTS

- The lipid bilayer of cell membranes is highly permeable to small, non-polar molecules such as oxygen and carbon dioxide and, to a lesser extent, to very small, polar molecules such as water. It is highly impermeable to most large, water-soluble molecules and to all ions.
- Transfer of nutrients, metabolites, and inorganic ions across cell membranes depends on membrane transport proteins.
- Cell membranes contain a variety of transport proteins that function either as transporters or channels, each responsible for the transfer of a particular type of solute.
- Channel proteins form pores across the lipid bilayer through which solutes can passively diffuse.
- Both transporters and channels can mediate passive transport, in which an uncharged solute moves spontaneously down its concentration gradient.
- For the passive transport of a charged solute, its electrochemical gradient determines its direction of movement, rather than its concentration gradient alone.
- Transporters can act as pumps to mediate active transport, in which solutes are moved uphill against their concentration or electrochemical gradients; this process requires energy that is provided by ATP hydrolysis, a downhill flow of Na^+ or H^+ ions, or sunlight.
- Transporters transfer specific solutes across a membrane by undergoing conformational changes that expose the solute-binding site first on one side of the membrane and then on the other.
- The Na^+ pump in the plasma membrane of animal cells is an ATPase; it actively transports Na^+ out of the cell and K^+ in, maintaining a steep Na^+ gradient across the plasma membrane that is used to drive other active transport processes and to convey electrical signals.

- Ion channels allow inorganic ions of appropriate size and charge to cross the membrane. Most are gated and open transiently in response to a specific stimulus.
- Even when activated by a specific stimulus, ion channels do not remain continuously open: they flicker randomly between open and closed conformations. An activating stimulus increases the proportion of time that the channel spends in the open state.
- The membrane potential is determined by the unequal distribution of charged ions on the two sides of a cell membrane; it is altered when these ions flow through open ion channels in the membrane.
- In most animal cells, the negative value of the resting membrane potential across the plasma membrane depends mainly on the K^+ gradient and the operation of K^+-selective leak channels; at this resting potential, the driving force for the movement of K^+ across the membrane is almost zero.
- Neurons produce electrical impulses in the form of action potentials, which can travel long distances along an axon without weakening. Action potentials are propagated by voltage-gated Na^+ and K^+ channels that open sequentially in response to depolarization of the plasma membrane.
- Voltage-gated Ca^{2+} channels in a nerve terminal couple the arrival of an action potential to neurotransmitter release at a synapse. Transmitter-gated ion channels convert this chemical signal back into an electrical one in the postsynaptic target cell.
- Excitatory neurotransmitters open transmitter-gated cation channels that allow the influx of Na^+, which depolarizes the postsynaptic cell's plasma membrane and encourages the cell to fire an action potential. Inhibitory neurotransmitters open transmitter-gated Cl^- channels in the postsynaptic cell's plasma membrane, making it harder for the membrane to depolarize and fire an action potential.
- Complex sets of nerve cells in the human brain exploit all of the above mechanisms to make human behaviors possible.

KEY TERMS

action potential
active transport
antiport
axon
Ca^{2+} pump (or Ca^{2+} ATPase)
channel
dendrite
depolarization
electrochemical gradient
gradient-driven pump
H^+ pump (or H^+ ATPase)
ion channel
K^+ leak channel
ligand-gated channel
mechanically-gated channel
membrane potential
membrane transport protein
Na^+ pump (or Na^+-K^+ ATPase)
Nernst equation
nerve terminal
neuron
neurotransmitter
optogenetics
osmosis
passive transport
patch-clamp recording
pump
resting membrane potential
symport
synapse
synaptic vesicle
transmitter-gated ion channel
transporter
voltage-gated channel
voltage-gated Na^+ channel

QUESTIONS

QUESTION 12–8

The diagram in Figure 12–9 shows a transporter that mediates the passive transfer of a solute down its concentration gradient across the membrane. How would you need to change the diagram to convert the transporter into a pump that moves the solute up its concentration gradient by hydrolyzing ATP? Explain the need for each of the steps in your new illustration.

QUESTION 12–9

Which of the following statements are correct? Explain your answers.

A. The plasma membrane is highly impermeable to all charged molecules.

B. Channels have specific binding pockets for the solute molecules they allow to pass.

C. Transporters allow solutes to cross a membrane at much faster rates than do channels.

D. Certain H^+ pumps are fueled by light energy.

E. The plasma membrane of many animal cells contains open K^+ channels, yet the K^+ concentration in the cytosol is much higher than outside the cell.

F. A symport would function as an antiport if its orientation in the membrane were reversed (i.e., if the portion of the molecule normally exposed to the cytosol faced the outside of the cell instead).

G. The membrane potential of an axon temporarily becomes more negative when an action potential excites it.

QUESTION 12–10

List the following compounds in order of decreasing lipid-bilayer permeability: RNA, Ca^{2+}, glucose, ethanol, N_2, water.

QUESTION 12–11

Name at least one similarity and at least one difference between the following (it may help to review the definitions of the terms using the Glossary):

A. Symport and antiport

B. Active transport and passive transport

C. Membrane potential and electrochemical gradient

D. Pump and transporter

E. Axon and telephone wire

F. Solute and ion

QUESTION 12–12

Discuss the following statement: "The differences between a channel and a transporter are like the differences between a bridge and a ferry."

QUESTION 12–13

The neurotransmitter acetylcholine is made in the cytosol and then transported into synaptic vesicles, where its concentration is more than 100-fold higher than in the cytosol. When synaptic vesicles are isolated from neurons, they can take up additional acetylcholine added to the solution in which they are suspended, but only when ATP is present. Na^+ ions are not required for the uptake, but, curiously, raising the pH of the solution in which the synaptic vesicles are suspended increases the rate of uptake. Furthermore, transport is inhibited when drugs are added that make the membrane permeable to H^+ ions. Suggest a mechanism that is consistent with all of these observations.

QUESTION 12–14

The resting membrane potential of a typical animal cell is about –70 mV, and the thickness of a lipid bilayer is about 4.5 nm. What is the strength of the electric field across the membrane in V/cm? What do you suppose would happen if you applied this field strength to two metal electrodes separated by a 1-cm air gap?

QUESTION 12–15

Phospholipid bilayers form sealed, spherical vesicles in water (discussed in Chapter 11). Assume you have constructed lipid vesicles that contain Na^+ pumps as the sole membrane protein, and assume for the sake of simplicity that each pump transports one Na^+ one way and one K^+ the other way in each pumping cycle. All the Na^+ pumps have the portion of the molecule that normally faces the cytosol oriented toward the outside of the vesicles. With the help of Figures 12–11 and 12–12, determine what would happen in each of the following cases.

A. Your vesicles were suspended in a solution containing both Na^+ and K^+ ions and had a solution with the same ionic composition inside them.

B. You add ATP to the suspension described in (A).

C. You add ATP, but the solution—outside as well as inside the vesicles—contains only Na^+ ions and no K^+ ions.

D. The concentrations of Na^+ and K^+ were as in (A), but half of the pump molecules embedded in the membrane of each vesicle were oriented the other way around, so that the normally cytosolic portions of these molecules faced the inside of the vesicles. You then add ATP to the suspension.

E. You add ATP to the suspension described in (A), but in addition to Na^+ pumps, the membrane of your vesicles also contains K^+ leak channels.

QUESTION 12–16

Name the three ways in which an ion channel can be gated.

QUESTION 12–17

One thousand Ca^{2+} channels open in the plasma membrane of a cell that is 1000 μm^3 in size and has a cytosolic Ca^{2+} concentration of 100 nM. For how long would the channels need to stay open in order for the cytosolic Ca^{2+} concentration to rise to 5 μM? There is virtually unlimited Ca^{2+} available in the outside medium (the extracellular Ca^{2+} concentration in which most animal cells live is a few millimolar), and each channel passes 10^6 Ca^{2+} ions per second.

QUESTION 12–18

Amino acids are taken up by animal cells using a symport in the plasma membrane. What is the most likely ion whose electrochemical gradient drives the import? Is ATP consumed in the process? If so, how?

QUESTION 12–19

We will see in Chapter 15 that endosomes, which are membrane-enclosed intracellular organelles, need an acidic lumen in order to function. Acidification is achieved by an H^+ pump in the endosomal membrane, which also contains Cl^- channels. If the channels do not function properly (e.g., because of a mutation in the genes encoding the channel proteins), acidification is also impaired.

A. Can you explain how Cl^- channels might help acidification?

B. According to your explanation, would the Cl^- channels be absolutely required to lower the pH inside the endosome?

QUESTION 12–20

Some bacterial cells can grow on either ethanol (CH_3CH_2OH) or acetate (CH_3COO^-) as their only carbon source. Dr. Schwips measured the rate at which the two compounds traverse the bacterial plasma membrane but, due to excessive inhalation of one of the compounds (which one?), failed to label his data accurately.

A. Plot the data from the table below.

Concentration of Carbon Source (mM)	Rate of Transport (μmol/min)	
	Compound A	Compound B
0.1	2.0	18
0.3	6.0	46
1.0	20	100
3.0	60	150
10.0	200	182

B. Determine from your graph whether the data describing compound A correspond to the uptake of ethanol or acetate.

Explain your answers.

QUESTION 12–21

Acetylcholine-gated cation channels do not discriminate between Na^+, K^+, and Ca^{2+} ions, allowing all to pass through them freely. So why is it that when acetylcholine binds to this protein in the plasma membrane of muscle cells, the channel opens and there is a large net influx of primarily Na^+ ions?

QUESTION 12–22

The ion channels that are regulated by binding of neurotransmitters, such as acetylcholine, glutamate, GABA, or glycine, have a similar overall structure. Yet each class of these channels consists of a very diverse set of subtypes with different transmitter affinities, different channel conductances, and different rates of opening and closing. Do you suppose that such extreme diversity is a good or a bad thing from the standpoint of the pharmaceutical industry?