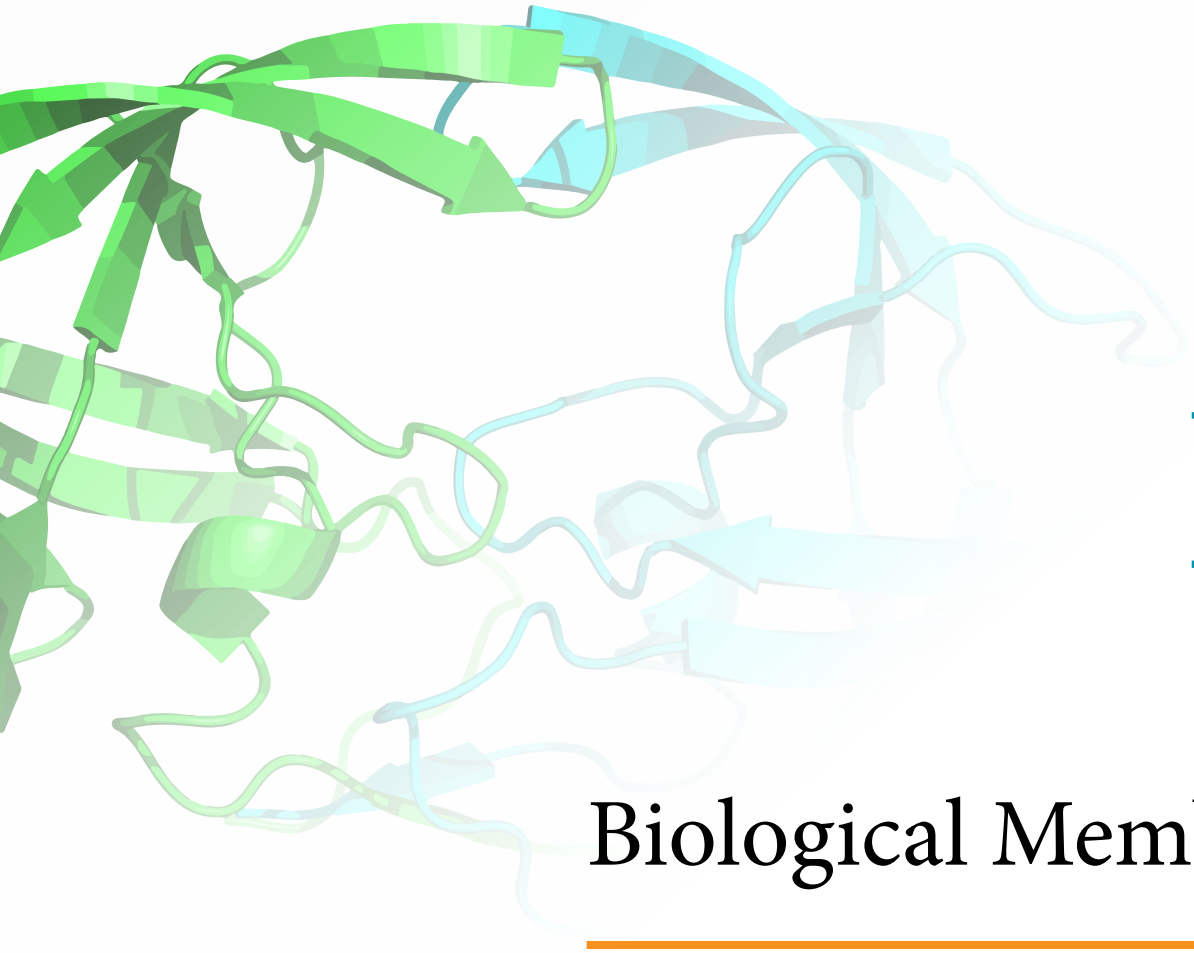

14

Biological Membranes

Goal

To understand the structure and composition of biological membranes.

Objectives

After this chapter, you should be able to

- distinguish between *cis* and *trans* unsaturated fatty acids.
- explain why phospholipids spontaneously form lipid bilayers and sealed compartments.
- describe membrane fluidity and how it is affected by membrane composition and temperature.
- explain the role of cholesterol in buffering membrane fluidity.
- explain how the polar backbone of a membrane protein can be accommodated in a bilayer.

The fundamental unit of life is the cell. All living things are composed of cells, be it a single cell in the case of many microorganisms or a highly organized ensemble of myriad cell types in the case of multicellular organisms. A defining feature of the cell is a membrane, the **cytoplasmic membrane**, that surrounds the cell and separates the inside of the cell, the cytoplasm, from other cells and the extracellular milieu. Membranes also surround specialized compartments inside of cells known as organelles. Whereas cells are typically several microns (μm) in diameter (although some cells can be much larger), the membrane is only about 10 nanometers (nm) thick. Yet, and as we will see in subsequent chapters, the membrane is not simply an ultra-thin, pliable sheet that encases the cytoplasm. Rather, membranes are dynamic structures that mediate many functions in the life of the cell. In this chapter we examine the composition of membranes, their assembly, the forces that stabilize them, and the chemical and physical properties that influence their function.

The preceding chapters have focused on two kinds of biological molecules, namely proteins and nucleic acids, that are important in the workings of living systems. Our discussion of membranes will introduce a third category of biological molecules—lipids. Lipids make up a broad group of naturally occurring molecules that are largely defined as being soluble in nonpolar organic solvents. They fall into two groups: hydrophobic lipids such as **triglycerides** (fats) and amphipathic lipids such as **fatty acids**, **phospholipids** and **cholesterol**, which have both hydrophobic and hydrophilic regions. All lipids are derived in whole or in part from carbon-

containing building blocks that are assembled enzymatically by protein complexes in the cell. Here we focus on the contributions of lipids to the properties of membranes.

Fatty acids consist of carboxyl groups with a carbon tail

Lipids in membranes often have complex structures that comprise several component parts (moieties). One common component of lipids, as we will see, is fatty acids. Fatty acids are composed of carboxyl groups with long hydrophobic tails (or chains) that have an even number of carbon atoms and consist only of carbon-carbon and carbon-hydrogen bonds. The chains found in membranes typically range in length from 16 to 20 carbon atoms. If the chain contains one or more double bonds, the fatty acid is said to be **unsaturated**. Conversely, if none of the carbons is linked with a double bond, the chain is **saturated**. The double bonds in unsaturated fatty acids usually have a *cis* configuration, meaning that the two hydrogens adjacent to the double bond project from the same side of the double bond. *Cis* double bonds cause the chain to kink.

So-called "trans fats", which are produced industrially, have ***trans*** double bonds in which the hydrogens are displayed on opposite sides of the double bond. *Trans* double bonds do not cause the chain to bend appreciably, and hence fatty acids containing *trans* bonds more closely resemble saturated fatty acids. Trans fats are notorious for their association with coronary heart disease when consumed in dietary products, such as margarine, that contain partially hydrogenated fatty acids.

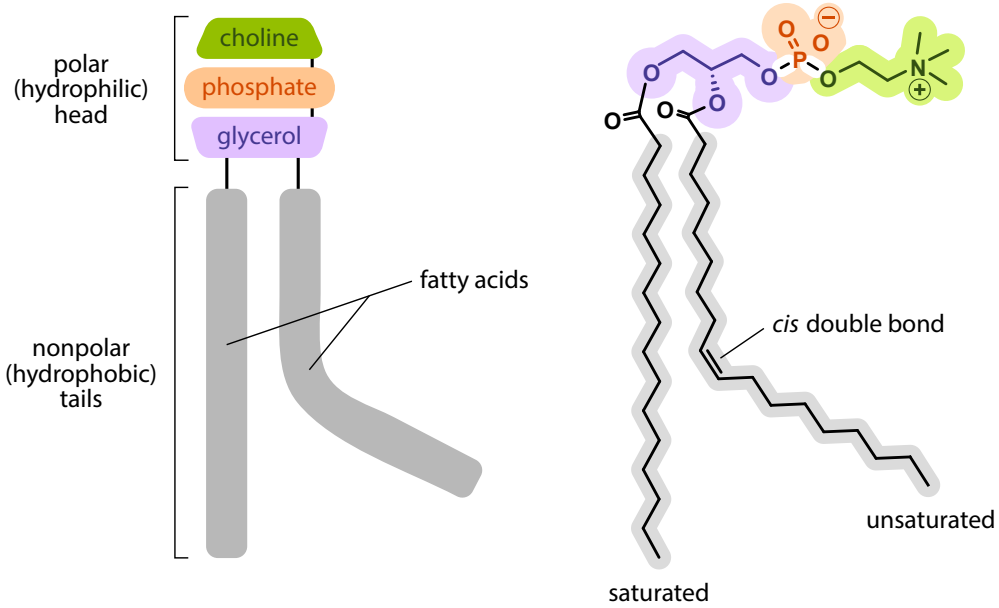

Figure 1 Phosphatidylcholine is an amphipathic molecule that contains a polar head group and nonpolar fatty acid tails

Phosphatidylcholine contains a polar head group that consists of a glycerol moiety (*purple*) that is linked to a phosphate (*red*). The head group of phosphatidylcholine also contains a choline moiety (*green*), which is unique to phosphatidylcholine. In addition to the polar head group, phosphatidylcholine and other phospholipids contain a pair of fatty acid tails (*gray*). The length of these tails varies from one phospholipid to the next. Some fatty acid tails are unsaturated, meaning that they contain at least one double bond. With few exceptions, this bond is *cis* in nature, giving the fatty acid tail a kinked shape.

Phospholipids consist of fatty acids linked to a phosphate-containing head group

One of the most abundant lipids in membranes is phospholipids. These amphipathic molecules generally have two hydrophobic tails composed of fatty acids and a polar head group containing a molecule of phosphate. The backbone of the head group is a molecule of glycerol to which the fatty acids and phosphate are attached by acyl linkages. One such phospholipid is **phosphatidylcholine** in which the phosphate is joined to a molecule of choline (Figure 1). The head groups of other phospholipids also contain glycerol and phosphate, but they contain other substituents in lieu of choline. The choline group of phosphatidylcholine is positively charged, whereas its phosphate group is negatively charged. Hence, and overall, phosphatidylcholine is electrostatically neutral. In contrast, some other phospholipids have negative net charges. Phospholipids also differ in the length and degree of saturation of their fatty acid tails. As we will see, the chemical composition of the phospholipids in a membrane can influence the physical properties of the membrane.

Phospholipids spontaneously form bilayers and sealed compartments in aqueous environments

Biological membranes are composed of two layers of phospholipids, collectively known as the **lipid bilayer** (Figure 2). The lipids in each layer are aligned side-by-side with their polar head groups oriented towards the aqueous external environment and their hydrophobic tails pointing towards the hydrophobic tails of the lipids in the other layer, thereby forming the interior of the bilayer. Each layer, or monolayer, is known as a **leaflet**. Thus, membranes consist of two leaflets with the hydrophobic portions of the lipids buried on the inside and the hydrophilic portions on the outside.

Phospholipids assemble into bilayers spontaneously when introduced into aqueous environments. Their assembly is driven by the hydrophobic effect such that the lipids spontaneously arrange themselves in a manner that minimizes the hydrophobic surface area that is exposed to water. Since phospholipids are amphipathic, an arrangement in which the polar head groups are in contact with water and the hydrophobic tails are buried is thermodynamically favored, maximizing the entropy of the system by preventing water molecules from forming ordered structures around exposed hydrophobic surfaces.

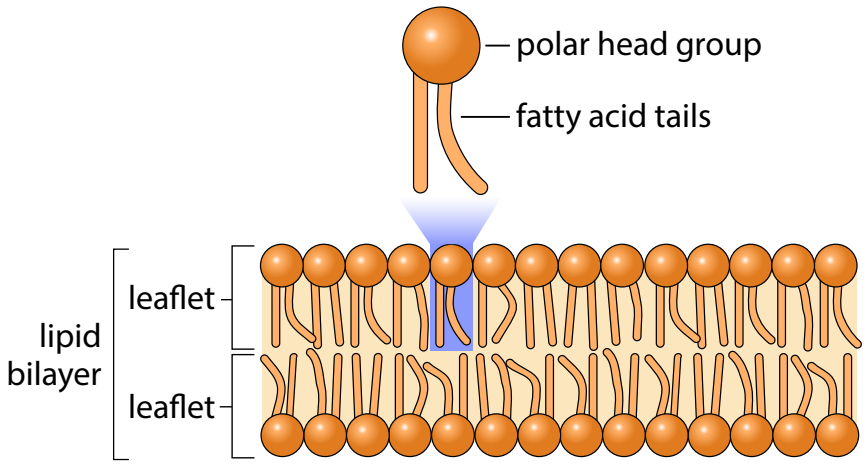

Figure 2 **Phospholipids spontaneously form membrane bilayers in aqueous environments**

In the bilayer the phospholipid head groups orient towards the aqueous environment, whereas the fatty acid tails orient towards the hydrophobic interior of the membrane.

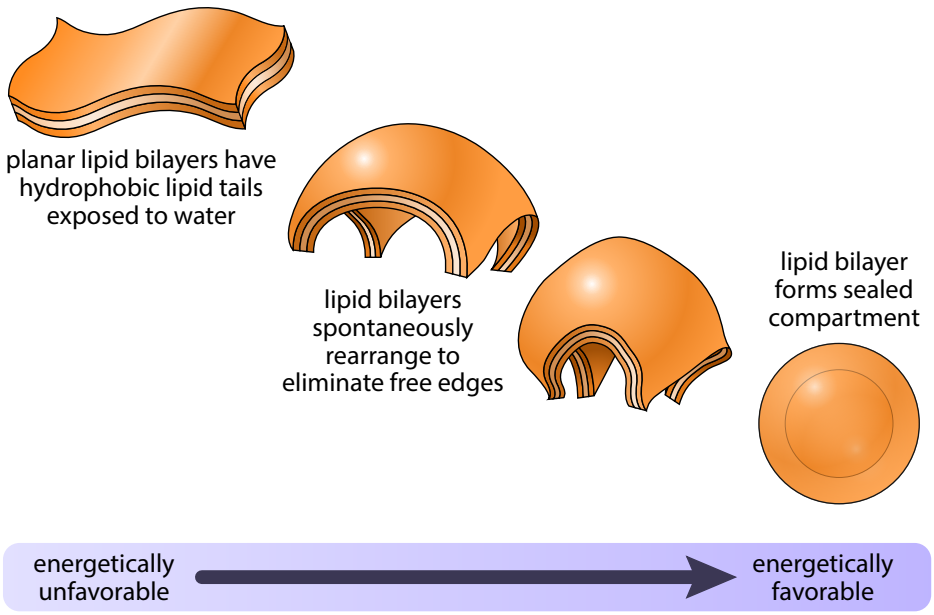

Figure 3 Bilayers spontaneously close to form sealed compartments

Consider a flat, planar bilayer in an aqueous solution in a test tube. The large majority of the fatty acid tails are buried within the two leaflets, but those at the edges of the bilayer are not. Instead, the hydrophobic tails of the lipids at the edges of the bilayer are in direct contact with the aqueous environment. Because the exposure of hydrophobic surfaces to water is energetically (i.e., entropically) unfavorable, bilayers organize themselves to eliminate free edges (Figure 3). In fact, synthetic bilayers eliminate these free edges by spontaneously forming sealed compartments known as **vesicles**. The ability of lipid bilayers to spontaneously form sealed compartments is fundamental to living cells; it arises because of the amphipathic nature of phospholipids, their shapes, and the fact that these molecules exist in an aqueous environment. The hydrophobic effect is the major driving force for both bilayer formation and the formation of sealed compartments. A sealed compartment represents the configuration in which the exposed hydrophobic surface area and the Gibbs free energy are minimized.

Remarkably, then, the most fundamental unit of life, the cell, is in essence an intrinsic feature of the behavior of lipids in solution. If we think back to Chapter 13 on the Origin of Life, we can imagine how the RNA World could have acted in concert with lipids to form primitive, membrane-bound progenitors of living systems.

Lipids with one fatty acid tail form micelles in solution

The shapes of lipids and the way they pack together with each other have a large effect on the structures they form in aqueous solutions. Lipids with two fatty acid tails, like the phospholipids that we introduced earlier, have a cylindrical shape. As a consequence, the packing arrangement that minimizes contact between their fatty acid tails and water is a bilayer as we discussed earlier. Lipids with only one fatty acid and a large head group, however, have an overall shape that is more conical. When these lipids pack together, they form a structure called a **micelle** in which the lipid tails are on the inside and the polar head groups are in contact with water (Figure 4).

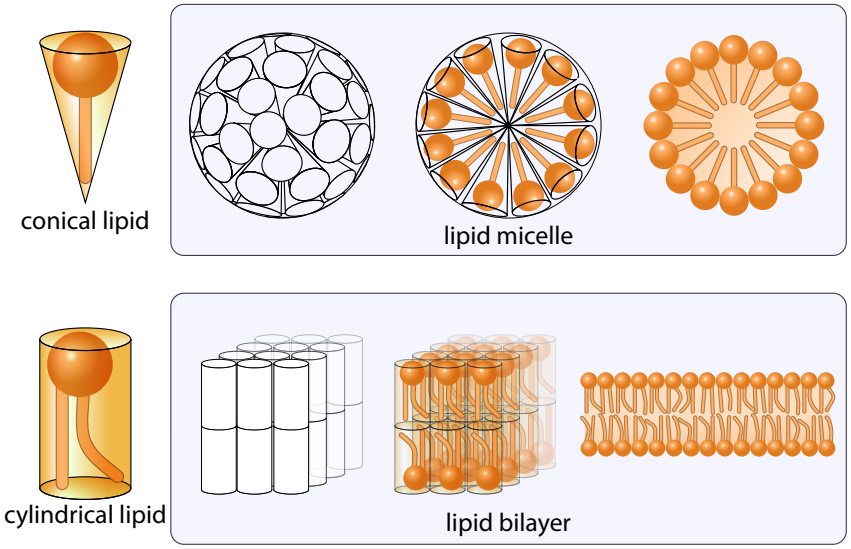

Figure 4 Lipid composition determines how lipids pack together

Lipids with just one hydrophobic tail are conical in shape, whereas lipids with two hydrophobic tails are more cylindrical. These shape differences influence the structure that is formed when each lipid is exposed to water. Conical lipids coalesce to form spherical micelles, whereas cylindrical lipids pack together to form bilayers.

Most detergents, like the dish and laundry detergents we use, have a charged head group and a single hydrocarbon chain, causing them to form micelles in aqueous solutions. When detergents are added to a solution containing nonpolar molecules (e.g., grease or oil), the nonpolar molecules partition into the interior of the micelle, where they are shielded from water and interact with the hydrocarbon tails of the detergent.

Membranes are dynamic

Although we often represent them using static diagrams, biological membranes are dynamic, and individual phospholipids are highly mobile. Phospholipids are free to rotate along their long axis, their fatty acid tails can flex, and they can diffuse laterally within a single membrane leaflet. Because of this mobility, membranes are highly fluid (Figure 5). In fact, lateral diffusion is so fast that a phospholipid in a bacterial cell can laterally diffuse within the membrane from one end of the cell to the other, about two microns, in just one second. In contrast, the exchange of phospholipids between the two leaflets, sometimes called flipping, is rare. Like any other slow chemical process, flipping has a high activation barrier. In order for a phospholipid to flip to the opposite leaflet, its head group must enter the

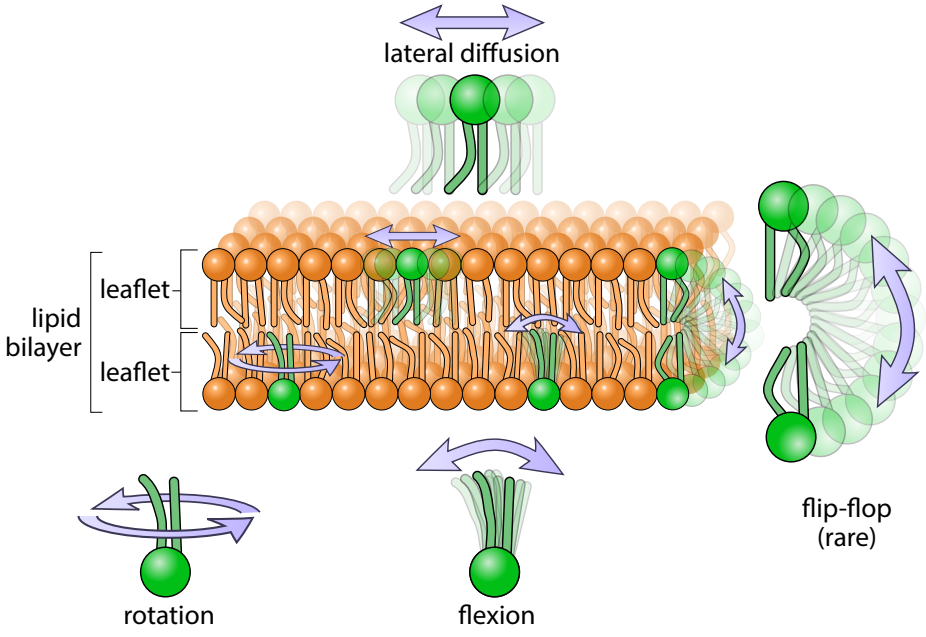

Figure 5 Lipids in the membrane are free to move in a variety of ways

Lipids in membranes have several modes of motion available to them. Lipids can rotate in place, and their fatty acid tails are free to bend and flex. Lipids are also free to laterally diffuse within a single leaflet of the membrane. Flip-flopping to the opposite leaflet is possible as well; however, this exchange is kinetically slow and happens only rarely.

hydrophobic interior of the membrane. For this to occur, however, the polar head group must break the favorable ion-dipole and dipole-dipole interactions it forms with water, which requires an input of energy. The energy required to break these interactions is the reason for the high activation energy and slow rate of phospholipid flipping. Because they are so dynamic, you can think of membrane lipids as constituting a two-dimensional solvent for proteins in the membrane, a topic we will come to shortly, just as water constitutes a three-dimensional solvent for proteins in an aqueous solution.

Membrane fluidity is determined by membrane composition and temperature

The **fluidity** of a membrane, or the extent to which the membrane components are free to move, is determined by both membrane composition and temperature. The types of fatty acids that compose the lipids in a membrane have a significant effect on fluidity. Generally speaking, membranes containing lipids with longer fatty acid tails are less fluid than membranes containing lipids with shorter fatty acid tails. This is because lipids with longer fatty acid tails have more surface area with which they can interact with one another. The nonpolar fatty acid tails interact with one another via van der Waals interactions, and for a lipid to move around within a fluid membrane, these van der Waals interactions must be easily broken. Therefore, the more interactions that form between the lipids in a membrane, the less fluid the membrane. Because the strength of van der Waals interactions scales with the surface area over which they occur, lipids with longer fatty acid chains form stronger interactions with one another than lipids with shorter fatty acid chains.

The degree to which fatty acid chains are saturated also affects membrane fluidity. The *cis* double bonds found in unsaturated fatty acids create kinks in the fatty acid chain that prevent lipids in the membrane from packing together tightly, thus decreasing the surface area over which lipids interact. As a consequence, membranes that are rich in unsaturated fatty acids are more fluid than membranes that contain fewer unsaturated fatty acids. Olive oil and candle wax are familiar examples of this phenomenon. Olive oil is rich in unsaturated fatty acids, such as oleic acid, whereas wax is rich in saturated fatty acids, principally stearic acid (Figure 6). As you know from experience, olive oil is a liquid at room temperature, whereas wax is a solid. The reason for their difference in melting temperature, even though both lipids have the same chain length (Figure 6), is the different strengths of the intermolecular interactions that form in wax and olive oil. The saturated fatty acids found in wax allow lipids to pack together more tightly, enabling strong intermolecular interactions. In contrast, the unsaturated fatty acids found in olive oil interfere with the close packing of lipids, reducing the strength of the intermolecular interactions that form between lipids and giving rise to a liquid instead of a solid. Double bonds in the *cis* configuration have a much greater effect on fluidity than do double bonds in the *trans* conformation. *Trans* double bonds do not create a kink in the fatty acid tail, and as such they do not interfere with lipid packing as much as *cis* double bonds do.

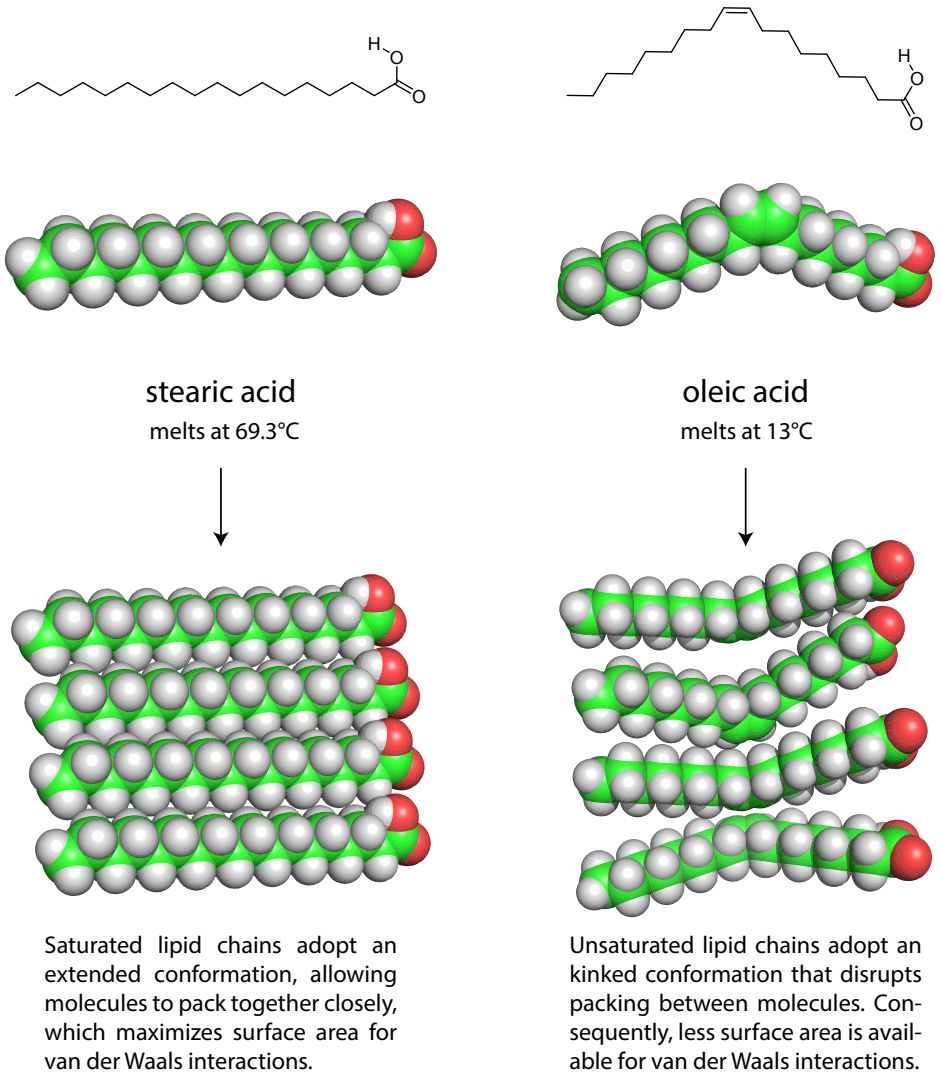

Figure 6 Membranes rich in *cis* unsaturated fatty acids are more fluid than membranes rich in saturated fatty acids

Shown are the structures of two related fatty acids, stearic acid and oleic acid. Both fatty acids contain 18 carbon atoms, making their tails the same length. Stearic acid is saturated, whereas oleic acid is unsaturated and contains one *cis* double bond. This single difference dramatically influences the physical properties of these molecules. The melting point of pure stearic acid is 69.3°C, much higher than the 13°C melting point of oleic acid.

Generally speaking, the fluidity of a membrane increases as temperature increases. Membranes can even undergo a phase transition in response to temperature changes. Above a critical **melting temperature** (the transition midpoint or $\mathbf{T_m}$), the physical properties of the membrane abruptly change, and the membrane transitions from a solid-like state to a fluid-like state. At low temperatures the lipids within the bilayer are well-ordered and packed into a crystal-like arrangement in which the immobilized lipids form strong interactions with one another. As the temperature increases, these interactions are broken, and the lipids become less ordered and more liquid-like. Membranes with different compositions tend to have different T_m values. Membranes consisting of long-chain lipids have higher melting temperatures than membranes consisting of short-chain lipids. Similarly, membranes that are rich in unsaturated fatty acids have lower melting temperatures than membranes that have a smaller proportion of unsaturated fatty acids.

Cholesterol modulates membrane fluidity

In addition to phospholipids, the membranes of animal cells contain the lipid **cholesterol** as a major component. Cholesterol belongs to a class of

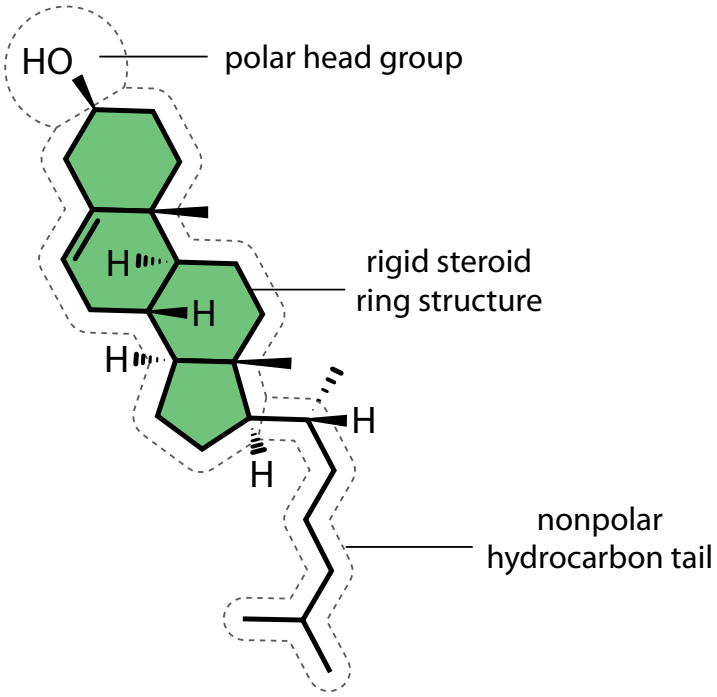

Figure 7 Cholesterol contains a polar head group, a rigid fused ring structure, and a flexible nonpolar tail

Like phospholipids, cholesterol is an amphipathic molecule. It contains a small polar head group that consists only of a single hydroxyl group. The remainder of the cholesterol molecule is nonpolar.

lipids called steroids, which are characterized by four fused carbon rings (Figure 7). Not only is cholesterol a major component of membranes, it is also the precursor to other steroids that function as hormones, such as the sex hormones testosterone and estrogen and corticosteroids, which regulate inflammation and other physiological processes. Like phospholipids, cholesterol is amphipathic. It has a polar head that contains a hydroxyl group, whereas the rest of the molecule is hydrophobic, consisting of the fused ring structure and a hydrocarbon tail. The four fused rings make most of the cholesterol molecule rigid, as the geometric constraints of being in a ring prevent the bonds connecting the atoms in the rings from rotating freely. Like phospholipids, cholesterol inserts into the membrane bilayer with its polar head group exposed to the aqueous solution and its nonpolar component concealed within the hydrophobic interior of the bilayer.

Cholesterol influences the fluidity of the membrane, and it does so in a bidirectional manner; at high temperatures it decreases fluidity and at low temperatures it increases fluidity. At high temperatures, cholesterol's flat, rigid structure limits phospholipid movement. The steroid ring interacts with, and partly immobilizes, the regions of the phospholipid fatty acid chains that are closest to the polar head groups (Figure 8). By decreasing the mobility of the first few CH_2 groups in the fatty acid chains, cholesterol makes that region of the lipid bilayer less deformable.

Cholesterol interacts differentially with each type of membrane lipid, and it forms particularly strong associations with saturated, high-melting point phospholipids and with sphingolipids. Conversely, cholesterol interacts weakly with unsaturated lipids. As an example, Figure 9 shows a diagram of the interaction that forms between cholesterol and a specific sphingolipid called sphingomyelin. The hydroxyl group of cholesterol forms a specific hydrogen bond with the amide group in the head of sphingomyelin. This interaction orients cholesterol with respect to sphingomyelin and creates a large, rigid structure that interferes with lipid movement and decreases membrane fluidity. Note that cholesterol is an asymmetric molecule and

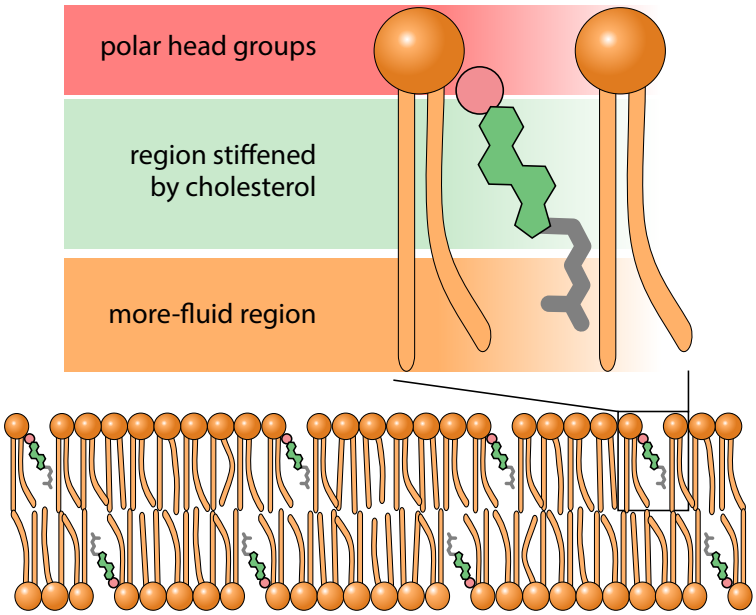

Figure 8 Cholesterol influences membrane fluidity by forming strong interactions with phospholipids

Like phospholipids, cholesterol in the bilayer orients with its polar head group facing the aqueous environment and its nonpolar region facing the membrane interior. One of cholesterol's functions is to reduce the fluidity of the membrane. To do so, it forms strong interactions with phospholipids using its rigid steroid ring structure. At high temperatures, these interactions stiffen the membrane and interfere with phospholipid mobility. At low temperatures, the flexible, nonpolar tail of cholesterol interferes with the tight packing of adjacent phospholipid chains.

only one specific face interacts with sphingomyelin, leaving a second face that can interact with proteins in the membrane.

In addition to decreasing membrane fluidity at high temperatures, cholesterol increases membrane fluidity at low temperatures. At the high concentrations found in most animal-cell cytoplasmic membranes, cholesterol prevents the hydrocarbon chains of lipids from packing together and forming ordered, crystal-like structures. This interference with packing broadens the phase transition that occurs during freezing, eliminating the rapid change in membrane fluidity that would otherwise occur near the T_m (Figure 9). The effects of cholesterol at low temperatures can be attributed to cholesterol's kinked tail, which disrupts lipid packing. Thus, cholesterol can be thought of as a buffering molecule in the membranes of animal cells that prevents abrupt changes in membrane fluidity over a range of temperatures.

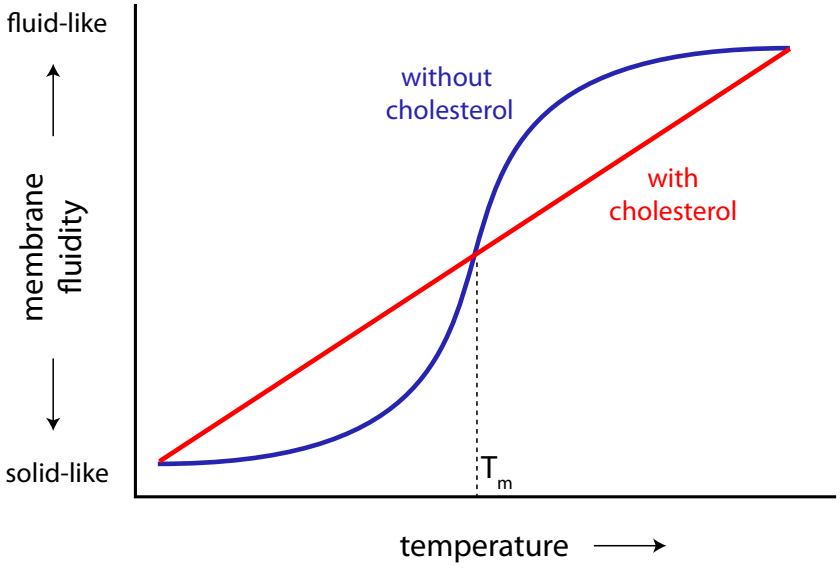

Figure 9 Cholesterol acts as a buffer to lessen the impact of temperature on membrane fluidity

Generally speaking, membrane fluidity increases as temperature increases. Membranes that contain only phospholipids exhibit an abrupt change in membrane fluidity as the temperature approaches the membrane's T_m (*blue*). This phase transition is broadened for membranes that contain significant quantities of cholesterol, and the corresponding change in membrane fluidity occurs more gradually and over a wider temperature range (*red*). Overall, cholesterol lowers membrane fluidity at high temperatures and increases membrane fluidity at low temperatures.

Box 1 Membrane fluidity can be measured using FRAP

How do we measure the fluidity of membranes in living cells? By what methods can we determine the effects of temperature and membrane composition on the ability of molecules in the membrane to diffuse laterally? A powerful laboratory technique for making these measurements is **Fluorescence Recovery After Photobleaching** (**FRAP**). To measure membrane fluidity using FRAP, we incorporate into the membrane of a cell we wish to study a particular membrane protein that has been fused to the **green fluorescent protein** (**GFP**). (We will discuss membrane proteins and their integration into the membrane in the next section.) GFP is a natural protein (from a jellyfish) that emits (fluoresces) green light when stimulated with blue or ultraviolet light. A selected area of the membrane is then "bleached" with a powerful laser beam. This method is known as "photobleaching" because a light source ("photo") is used to damage the fluorophore (i.e., fluorescent component) of GFP such that it can no longer fluoresce. Once a region of the membrane has been bleached, the observation is that it recovers fluorescence again over time (Figure 10). The explanation is that unbleached molecules of the GFP fusion protein from elsewhere in the cell membrane diffuse in the plane of the membrane and exchange places with the bleached proteins. The rate at which the bleached region becomes fluorescent again corresponds with the speed of such protein exchange and hence is a measure of the membrane fluidity. More-fluid membranes recover fluorescence faster than less-fluid membranes.

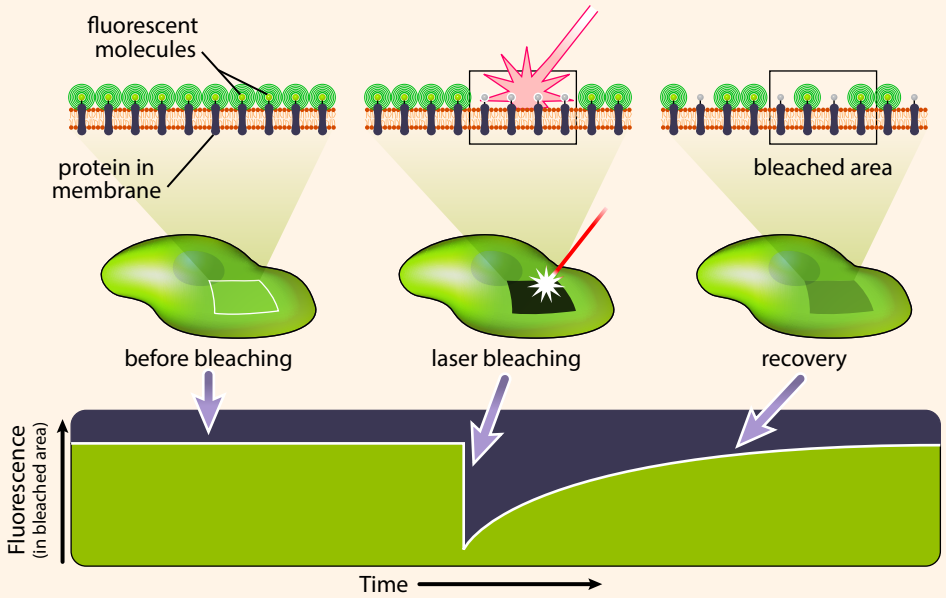

Figure 10 **FRAP experiments are used to measure membrane fluidity**

Biological membranes contain proteins as well as lipids

Central to the function of biological membranes are proteins (Figure 11). As we will come to in later chapters, these proteins carry out critical roles in the physiology of the cell, including transporting small molecules across the membrane and receiving and transducing signals from the outside to the inside of the cell.

How are membrane proteins embedded in the bilayer? Many membrane proteins fully span the bilayer, extending across it from one side to the other and displaying additional regions of protein on either side. These are known as transmembrane proteins. How is it possible for a stretch of polypeptide to cross the membrane given the highly polar nature of the peptide backbone, which would be expected to form favorable interactions

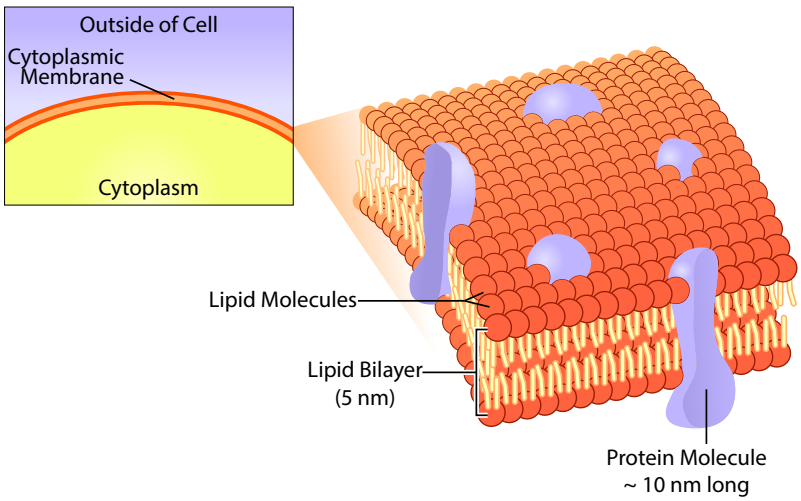

Figure 11 Membranes contain an abundance of proteins

with water in aqueous environments? These favorable interactions would have to be broken in order for the protein to be inserted in the membrane, and this would require an input of energy ($\Delta H_{rxn} > 0$). Transmembrane proteins commonly use α-helices to avoid this problem. In an α-helix, the polar atoms in the peptide backbone do not form hydrogen bonds to water; instead, these atoms form hydrogen bonds to other atoms in the backbone. This is true whether the helix is in water or in a membrane. In fact, the hydrogen bonds between backbone atoms in an α-helix are more favorable than are hydrogen bonds formed with water. As a consequence, the enthalpic penalty for "desolvating" the backbone atoms of an α-helix and inserting it into a membrane is minimized.

Not just any α-helix will favorably insert into a membrane, as we must also consider the side chains of the amino acids that compose the helix. If you recall from Chapter 6, the side chains of an α-helix point outward, away from the helical axis, thus placing them in contact with the hydrophobic interior of the membrane (Figure 12). If these side chains were polar or charged, energy would be required to desolvate them (i.e., break the interactions they form with water) upon insertion into the membrane. As a consequence, it is rare to find polar/charged residues in transmembrane α-helices unless those residues are used to mediate interactions with other transmembrane α-helices. Instead, most side chains in transmembrane α-helices are nonpolar. The insertion of nonpolar α-helices into the membrane is favorable because of the hydrophobic effect. Water forms ordered structures around the nonpolar side chains in aqueous solutions, and when these side chains are buried in the membrane, the ordered water molecules are released, increasing the entropy of the system. In sum, most proteins that span the membrane do so using α-helices composed of nonpolar amino acids. Transmembrane proteins often contain more than one transmembrane α-helix. In such cases, the transmembrane helices are linked to each other via stretches of amino acids that are displayed on one or both sides of the membrane.

Although most transmembrane proteins are α-helical, some are instead composed of β-strands. More specifically, the transmembrane portion of such proteins is composed of a single β-sheet that curves back around on itself to form a cylinder or barrel such that the edges of the sheet contact each other (Figure 13). As in all β-sheets, all of the polar backbone atoms

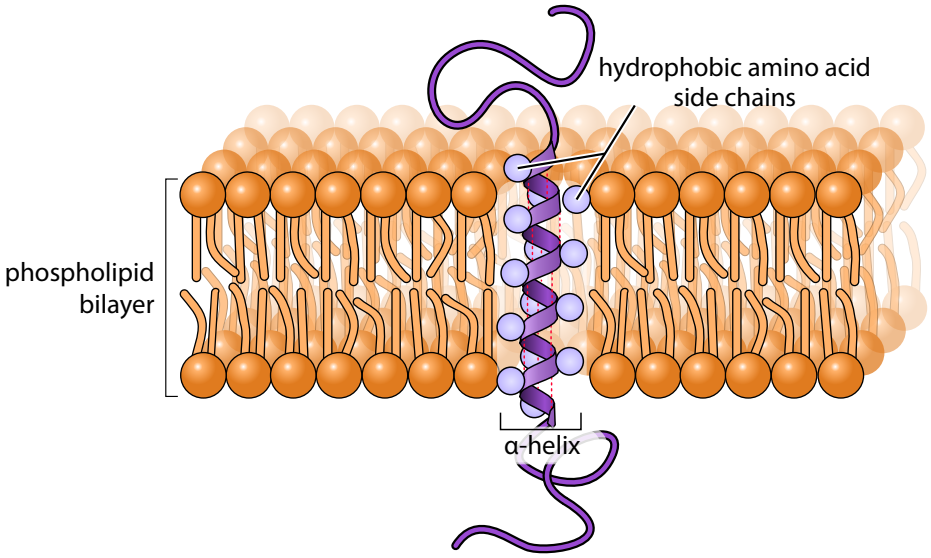

Figure 12 **Most transmembrane proteins are embedded in the membrane using α-helices composed of nonpolar amino acids**

form hydrogen bonds to backbone atoms in adjacent strands (see Chapter 6). As with transmembrane α-helices, the side chains exposed on the surface of these β-barrel proteins are almost always nonpolar.

Finally, some proteins that are associated with the membrane don't actually cross the lipid bilayer. Rather, these membrane-associated proteins use an α-helix that is oriented along the long axis of the membrane. In such cases, the α-helix is amphipathic, displaying nonpolar sidechains on one face of the helix that are buried in the hydrophobic interior of the membrane and hydrophilic side chains on the other face that project into the aqueous environment.

Summary

Biological membranes are composed of lipids and proteins. Most of the lipids are phospholipids. Phospholipids consist of a molecule of glycerol to which the phosphate is attached together with two fatty acids. The phosphate in turn is linked to a polar moiety, such as choline. Fatty acids have long hydrophobic tails. The lengths of the fatty acid tails and their degree of saturation (i.e., whether they contain double bonds) varies from

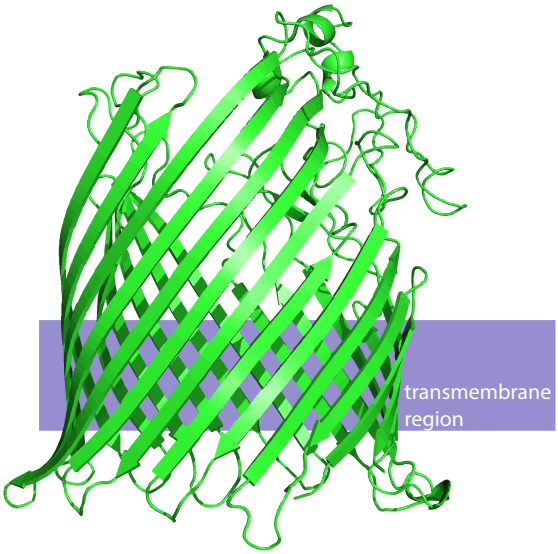

Figure 13 **Some transmembrane proteins are β-barrels**

Box 2 Lipid microdomains allow proteins to distribute heterogeneously within a membrane bilayer

Not all membranes contain the same proteins. Different proteins are targeted to different membranes in the cell, such as the cytoplasmic membrane and the membranes that encapsulate various organelles. Interestingly, and surprisingly, even proteins within a single membrane bilayer can be distributed in a non-uniform manner. Such heterogeneity results from **lipid microdomains**, which are clusters of lipids with longer fatty acid chains than most of the other lipids in the membrane (Figure 14). Such lipids favorably associate with each other to maximize the van der Waals interactions among the fatty acid chains and to prevent the extra-long fatty acid chains from being exposed to water. Proteins with longer-than-usual transmembrane regions are similarly sequestered into such microdomains. This kind of partitioning can promote protein-protein interactions by raising the local concentrations of specific proteins relative to one another. Because of interactions between lipids and cholesterol and between proteins and cholesterol, cholesterol is also enriched in some regions of the membrane over others.

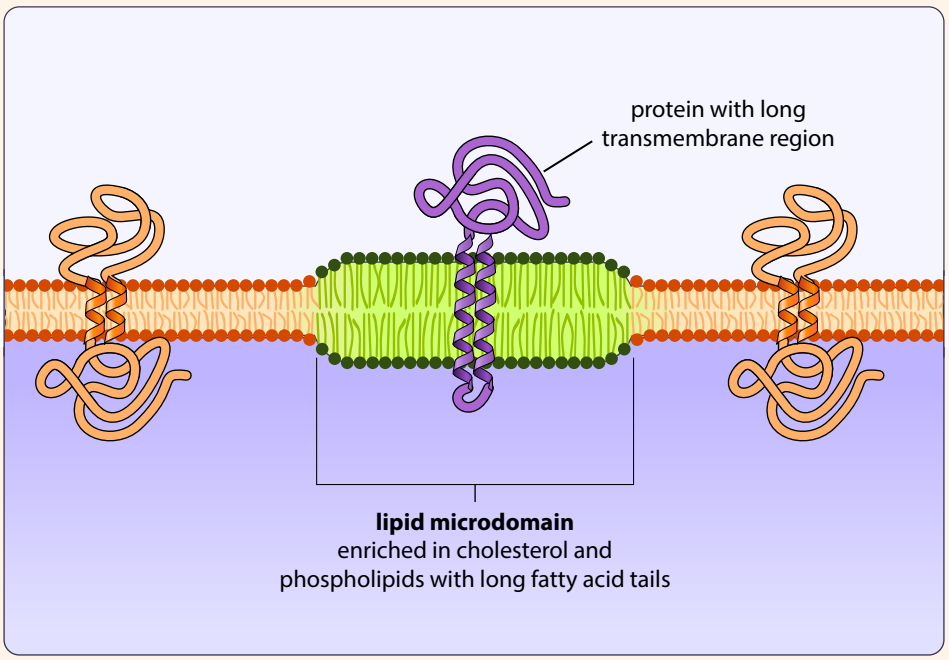

Figure 14 **Membranes can be heterogeneously organized**

one phospholipid to the next. *Cis* double bonds, in which the hydrogen atoms are on the same side of the double bond, cause the chain to kink.

Phospholipids spontaneously form bilayers in which the hydrophobic tails of the fatty acids of each leaflet are oriented toward the inside of the bilayer and the polar head groups point out toward the aqueous environment. In addition to phospholipids, animal membranes contain cholesterol. Cholesterol contains a polar hydroxyl group that orients in the bilayer to face the aqueous environment. The remainder of cholesterol is nonpolar, and as such, it orients towards the interior of the bilayer to avoid water.

Membranes are not static, and the individual lipids and proteins in membranes are generally free to move within the two-dimensional plane of each leaflet of the bilayer. The speed with which these molecules move is determined by the membrane's fluidity. Membrane fluidity is influenced by lipid composition and by temperature. Lipids with long and/or saturated

fatty acids tend to produce less-fluid membranes than lipids with short and/or unsaturated fatty acids. Membrane fluidity increases with increasing temperature. Cholesterol counters temperature's effect on membrane fluidity. At higher temperatures cholesterol decreases membrane fluidity, and at lower temperatures it increases membrane fluidity.

Proteins that span the membrane, known as transmembrane proteins, typically do so by means of α-helices composed of nonpolar amino acid residues. The α-helical secondary structure allows all of the polar backbone atoms to participate in hydrogen bonds with one another in the hydrophobic membrane interior. This self-bonding removes the energetic cost associated with breaking the interactions that these molecules would otherwise be forming with water prior to membrane insertion. The insertion of nonpolar α-helices into the membrane is favorable because of the hydrophobic effect. Some transmembrane proteins are composed of β-barrels instead of α-helices. The β-barrel structure similarly allows all polar backbone atoms to form hydrogen bonds to other backbone atoms in the hydrophobic interior of the membrane.