Chapter 10 Biological Membranes

The lipid bilayer introduced in Chapter 11 accounts for many of the fundamental properties of biological membranes. In particular, it provides a barrier to the movement of hydrophilic molecules and so not only enables cells to exclude components of the extracellular environment, but also permits compartmentation within the cell. An important principle of the organization and regulation of many cellular activities is that they take place in a spatially defined fashion. However, membranes are not merely inert permeability barriers but, under appropriate conditions, allow the passage of selected ions and molecules that contribute to the steady-state fluxes of energy and matter that are the essence of biological processes.
Biological membranes are composed of a great variety of proteins as well as lipids. These proteins are enzymes, facilitate transmembrane transport, and use information concerning the extracellular milieu to prompt the correct intracellular response. This chapter describes the features of membrane proteins that enable them to interact with lipids, and describes the overall arrangement of lipids and proteins in membranes. The structures and functions of lipoproteins are also covered. Lastly, the mechanisms involved in the transport of molecules across membranes are introduced.

Essential Concepts

Membrane Proteins

1. The types and relative amounts of membrane proteins and lipids vary among biological membranes. Membrane proteins, although extremely diverse, can be classified as integral, lipid-linked, or peripheral, depending on how they are associated with the membrane.

2. An integral protein is amphiphilic in that some regions of the molecule strongly associate with nonpolar membrane components by means of hydrophobic effects, whereas hydrophilic portions extend into the aqueous surroundings. Integral proteins can be solubilized and extracted from membranes only by using relatively harsh reagents, such as detergents and organic solvents, to disrupt the membrane structure.

3. Membrane proteins contribute to the asymmetry of a biological membrane because they either are present on one side of a membrane or, if they extend through it, are oriented in only one direction. The transmembrane domain of an integral protein may consist of one α helix (as in glycophorin A), a bundle of helices (as in bacteriorhodopsin), or a β barrel (as in porin). Both α helices and β barrels can form all their possible hydrogen bonds and are therefore stable in the interior of the lipid bilayer. In all cases, hydrophobic residues contact the hydrocarbon chains in the membrane core, whereas charged polar groups tend to predominate at the membrane surface.

4. Certain membrane proteins are covalently linked to a lipid moiety, which provides the protein with a hydrophobic anchor in the membrane, There are three main types of lipid-linked proteins:

(a) Prenylated proteins have an attached isoprenoid moiety, a polymer built of C_5 isoprene units. The most abundant of these are farnesyl (C_{15}) and geranylgeranyl (C_{20}) groups. As a rule, prenylation occurs at the C-terminus of a protein.

(b) Fatty acylated proteins have a covalently attached myristoyl or palmitoyl group. Myristoylation is a stable modification that occurs through an amide bond to the α amino group of an N-terminal glycine. Palmitoylation occurs via a thioester linkage to a cysteine and is reversible.

(c) Glycosylphosphatidylinositol (GPI)-linked proteins are located at the external surface of the cell and are anchored to the membrane by GPI, a glycolipid that is covalently attached to the C-terminus of the protein via an amide bond.

5. Peripheral membrane proteins associate with membrane surfaces via electrostatic and hydrogen bonding interactions. As a result, these proteins can be removed from membranes by relatively gentle methods, such as extraction with salt solutions or variations in pH, that do not greatly disturb native membrane structure. Once dissociated, they exhibit properties typical of water-soluble proteins.

Membrane Structure and Assembly

6. The widely accepted concept of biological membrane structure, for which there is much evidence, is the fluid mosaic model. The model envisions integral proteins that float in a sea of lipid and can diffuse laterally unless restrained by other cell constituents. The rate of membrane protein diffusion can be assessed by fluorescent photobleaching recovery, which measures the rate at which a fluorescent-labeled membrane component diffuses into an area of membrane that has been previously bleached by a laser beam.

7. The structure and function of the erythrocyte membrane have been extensively investigated, largely through the use of erythrocyte ghosts, which are cells from which hemoglobin and other soluble cell constituents have been removed by osmotic lysis. The erythrocyte's biconcave disklike shape, which is essential for its O_2-carrying properties, is maintained by the membrane skeleton, a network of proteins located just beneath the membrane. The chief element in the skeleton is spectrin, a fibrous heterotetrameric protein that is cross-linked to other skeletal and membrane proteins, including actin and ankyrin. The proteins of the erythrocyte membrane skeleton occur in many other cell types.

8. The lipid and protein components of biological membranes are unevenly distributed. The cytoskeleton apparently restricts the movements of many membrane proteins and hence contributes to this heterogeneity. Localized membrane domains that have different lipid compositions may result from the association of certain lipids with membrane proteins.

9. Membrane constituents are asymmetrically distributed with respect to the outer and inner leaflets of the bilayer in natural membranes. For example, the carbohydrate components of plasma membrane glycoproteins and glycolipids are always located on the extracellular membrane surface. The inner and outer leaflets also contain lipids in different proportions.

10. Lipid asymmetry, which can be assessed through the use of specific phospholipases, results in part from the asymmetric synthesis of lipids on the cytoplasmic face of the plasma membrane (in prokaryotes) or the endoplasmic reticulum (in eukaryotes). Lipids are subsequently redistributed by two mechanisms: (a) Enzymes called flipases facilitate the flip-flop of specific phospholipids from one leaflet to the other; and (b) ATP-dependent translocases establish a nonequilibrium distribution of lipids. In eukaryotes, lipids are transported as components of vesicles that bud off the endoplasmic reticulum and ultimately fuse with other cellular membranes.

11. Protein synthesis starts at the N-terminus and proceeds to the C-terminus of a polypeptide chain. Synthesis takes place either on free ribosomes (for soluble proteins) or on ribosomes bound to the endoplasmic reticulum (for integral membrane and secretory proteins). The signal hypothesis explains how polypeptides come to traverse the endoplasmic reticulum membrane:
    (a) Proteins synthesized on ER-bound ribosomes have an N-terminal amino acid sequence known as a signal peptide.
    (b) After an ~80-residue polypeptide chain has been synthesized, the signal peptide binds to the signal recognition particle (SRP). This event stops further polypeptide synthesis.
    (c) The SRP–ribosome complex, bearing the nascent polypeptide chain, binds to a docking protein (the SRP receptor) on the ER surface. Protein synthesis resumes, and the N-terminal sequence is translocated to the lumen of the ER via a transmembrane channel. At this point, the SRP dissociates from the complex.
    (d) Once in the ER lumen, the signal peptide is removed from the polypeptide by hydrolysis catalyzed by a signal peptidase.
    (e) The growing polypeptide undergoes folding and posttranslational modification, such as addition of core *N*-linked oligosaccharides.
    (f) When synthesis is complete, secretory proteins have passed entirely through the ER membrane into the lumen, whereas transmembrane proteins remain embedded in the membrane with their C-terminus on the cytoplasmic face.

12. Transmembrane and secretory proteins move from the endoplasmic reticulum to the Golgi apparatus as part of COPI- and COPII-coated vesicles (COP = coat protein), to be further processed, primarily by modification of their oligosaccharide chains. Clathrin-coated vesicles, which have a polyhedral structure, transport proteins from the Golgi apparatus to the plasma membrane and other destinations. After vesicle fusion, integral membrane proteins that faced the ER lumen face the extracellular space.

Lipoproteins and Receptor-Mediated Endocytosis

13. Many lipids are transported between tissues through the circulation as components of lipoproteins. Lipoproteins are globular structures that are composed of a hydrophobic interior containing triacylglycerols and cholesteryl esters encased in an amphiphilic outer layer of protein, phospholipid, and cholesterol. There are five classes of lipoproteins: (a) chylomicrons; (b) very low density lipoproteins (VLDL); (c) intermediate density lipoproteins (IDL); (d) low density lipoproteins (LDL); and (e) high density lipoproteins

(HDL). The densities of lipoprotein classes increase as the quantity of lipid contained in the core decreases.

14. At least nine apolipoproteins comprise the protein components of human lipoproteins. Most of these have a high content of α helices, whose nonpolar and polar residues are on opposite sides of the α-helical cylinders. The nonpolar faces of the apolipoproteins interact with the phospholipid hydrophobic tails, and the polar faces interact with the phospholipid polar head groups.

15. Cholesterol can enter cells via receptor-mediated endocytosis of LDL. In this process, LDL binds to cell-surface receptors that recognize the apolipoprotein B-100 component of LDL. The LDL–receptor complex enters clathrin-coated pits, which pinch off from the plasma membrane to form clathrin-coated vesicles that are transported to endosomes. There, LDL dissociates from its receptor, which moves back to the plasma membrane. LDL is degraded in lysosomes, releasing cholesteryl esters that are then hydrolyzed to yield cholesterol.

16. Receptor-mediated endocytosis is a widely employed mechanism for the uptake of macromolecules.

Transport across Membranes

17. The hydrophobic interior of biological membranes renders them impermeable to ions and small polar compounds, such as amino acids, carbohydrates, and nucleotides, although water can traverse the bilayer with surprising ease. Polar substances cross membranes through the mediation of transport proteins.

18. The thermodynamics of diffusion across a membrane can be expressed in terms of a chemical equilibrium. The difference in concentration of substance A on two sides of a membrane generates a chemical potential difference, $\Delta\overline{G}_A$. When A is ionic, an electrical potential difference ($\Delta\Psi$) may also develop. Thus, the equation for the electrochemical potential difference contains terms for the concentration and the charge of substance A:

$$\Delta\overline{G}_A = RT \ln \frac{[A]_{in}}{[A]_{out}} + Z_A \mathcal{F} \Delta\Psi$$

where Z_A is the ionic charge of A and $\mathcal{F}$ is the Faraday constant (96,485 coulomb·mol^{-1}). A negative value for $\Delta\overline{G}_A$ indicates spontaneous transport of substance A from the outside to the inside.

19. Transport may be classified as either mediated or nonmediated. Diffusion accounts for nonmediated transport. The chemical potential difference determines the direction of nonmediated transport, and the substance moves in the direction that will eliminate the concentration difference and at a rate proportional to the size of the gradient.

20. Mediated transport makes use of carrier molecules, called permeases, transporters, or translocases. In passive-mediated transport (also called facilitated diffusion), a molecule is transported from a high to a low concentration. In active transport, an energy-yielding process must be coupled to the movement of a substance from a lower to a higher concentration.

21. Ionophores are model carrier molecules that greatly increase the permeability of a membrane to certain ions. Some of these organic compounds (such as valinomycin) bind the ion on one side of the membrane and carry it to the other side; others (such as gramicidin A) form transmembrane channels through which the ion can pass. Porins are channel-forming membrane proteins that display some solute selectivity.

22. Integral membrane proteins that mediate passive transport cycle between conformational states in which binding sites for the molecule to be transported are alternately accessible on one side of the membrane and then on the other. For example, the glucose transporter binds glucose at the external cell surface, then changes conformation so as to release glucose at the cytoplasmic surface. It then reverts to its previous conformation.

23. Passive transport proteins can operate in either direction, depending on the concentrations of the transported substance on both sides of the membrane. A uniport mechanism transfers a single atom or molecule across the membrane; symport involves two substances moving in the same direction; and antiport is the movement of two substances in opposite directions.

24. Active transport is coupled to the hydrolysis of ATP. A well-studied example is the plasma membrane (Na^+–K^+)–ATPase, which pumps 3 Na^+ out and 2 K^+ into the cell, thereby moving both ions against their concentration gradients, with every ATP hydrolyzed. The resulting electrochemical gradient of Na^+ and K^+ is the basis for the electrical excitability of nerve cells.

25. The mechanism of the (Na^+–K^+)–ATPase involves a series of reaction steps that normally operate in only one direction because ATP breakdown and ion movement are coupled vectorial processes. The Ca^{2+}–ATPase, which pumps cytosolic Ca^{2+} to the cell exterior against a large concentration gradient, functions in a similar manner to the (Na^+–K^+) pump.

26. In secondary active transport, the energy generated by an electrochemical gradient is utilized to drive another, endergonic transport process. One example is the uptake and concentration of glucose by the intestinal epithelial cells. This task is accomplished by using the Na^+ gradient produced by the (Na^+–K^+) pump. Another example is the bacterial lactose permease, which utilizes a proton gradient across the bacterial membrane to power the transport of lactose.

Key Equation

$$\Delta\overline{G}_A = RT \ln\left(\frac{[A]_{in}}{[A]_{out}}\right) + Z_A\mathcal{F}\Delta\Psi$$

Guide to Study Exercises (text p. 277)

1. Integral membrane proteins contain hydrophobic segments that span the width of the lipid bilayer and thereby anchor the protein in the hydrophobic core of a membrane. An integral membrane protein can be separated from the membrane only by disrupting the membrane.

   Peripheral membrane proteins, in contrast, are not anchored in the lipid bilayer but are associated with the outer surface of the membrane. A peripheral protein can therefore be separated from the membrane under relatively mild conditions. (Sections 10-1A and C)

2. Lipids can be covalently attached to proteins in three ways:
   (1) A prenyl group, such as a farnesyl or geranylgeranyl group, can be attached to a Cys residue at the protein's C-terminus;
   (2) A fatty acyl group, such as myristoyl or palmitoyl group, can be attached to a protein via an amide linkage to an α-amino group or via thioester linkage to a Cys side chain.
   (3) Glycosylphosphatidylinositol can be linked to a protein's C-terminus. (Section 10-1B)

3. According to the fluid mosaic model, integral membrane proteins are free to diffuse laterally within a lipid bilayer due to its lateral fluidity. The movements of a membrane protein may be limited by their interactions with other membrane proteins or with cytoskeletal proteins underlying the membrane. (Sections 10-2A and B)

4. The cytoskeleton is an organized network of proteins associated with the cytosolic side of the plasma membrane. Some of its components are integral membrane proteins, others are peripheral proteins. The distributions of other membrane proteins are influenced by interactions between these proteins and elements of the cytoskeleton. The cytoskeleton may also limit the diffusion of membrane proteins to an extent determined by the degree to which they interact with cytoskeletal proteins. (Section 10-2B)

5. Lysosomal proteins, as are all proteins, are ribosomally synthesized, beginning with the N-terminus. An RNA–protein complex, the signal recognition particle (SRP), recognizes the N-terminal region of the nascent polypeptide, which contains a stretch of hydrophobic residues (the signal peptide). The SRP binds to the ribosome, thereby arresting polypeptide synthesis, and eventually becomes attached to the SRP receptor on the rough endoplasmic reticulum. This causes polypeptide synthesis to resume. The N-terminus of the polypeptide is extruded through a transmembrane protein channel into the lumen of the ER. A membrane-bound signal peptidase removes the signal peptide, and the remainder of the lysosomal protein is synthesized and enters the lumen of the ER, where it folds and undergoes posttranslational modification. It is then transported to the Golgi apparatus,