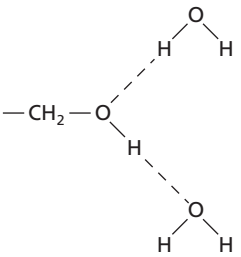
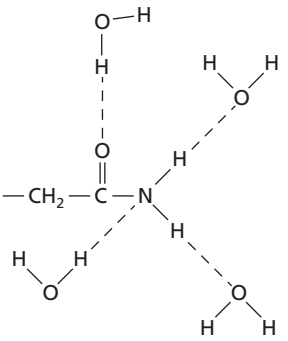
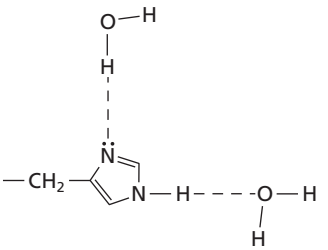
Solutions

Chapter 2 Water

1. Hydrogen bonds involve strongly electronegative atoms such as nitrogen, oxygen, or sulfur.

(a) 

(b) 

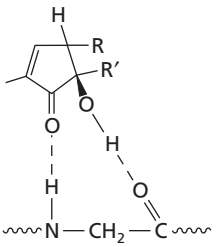
(c) 

2. (a) Glycerol is polar; it is not amphipathic; and it readily dissolves in water.
(b) Hexadecanoyl phosphate is polar; it is amphipathic; and it does not readily dissolve in water but forms micelles.
(c) Laurate is polar; it is amphipathic; and it does not readily dissolve in water but forms micelles.
(d) Glycine is polar; it is not amphipathic; and it readily dissolves in water.

3. There is a larger osmotic pressure inside the cells than outside because the molar concentration of solutes is much greater inside cells than outside. This results in a diffusion of water into cells, causing them to swell and burst.

4. If the pH of a solution is *below* the pK_a of any given ionizable group, the predominant species will be the one with the dissociable proton *on that group*. If the pH of a solution is *above* the pK_a of any given ionizable group, the predominant species will be the one with the dissociable proton *off of that group*.
(a) pH = 11 where the $-COO^{\ominus}$ form predominates.
(b) pH = 2 where the $H^{\oplus}$ form predominates.
(c) pH = 2 where the $H^{\oplus}$ form predominates.
(d) pH = 11 where the $R-O^{\ominus}$ form predominates.

5. (a) Tomato juice. For pH = 4.2, if pH = $-\log [H^{\oplus}]$, then
$[H^{\oplus}] = 10^{-pH}[H^{\oplus}] = 10^{-4.2} = 6.3 \times 10^{-5}$ M.
The ion-product constant of water (K_w) relates the concentrations of $OH^{\ominus}$ and $H^{\oplus}$ (Equation 2.6).
$[OH^{\ominus}] = K_w)/[H^{\oplus}] = 1.0 \times 10^{-14}\ M^2/6.3x^{-5}\ M = 1.6 \times 10^{-10}$ M.
(b) Human blood plasma. If the pH = 7.4, then
$[H^{\oplus}] = 10^{-7.4} = 4.0 \times 10^{-8}$ M. $[OH^{\ominus}] = K_w/[H^{\oplus}] =$
$1.0 \times 10^{-14}\ M^2/4.0x^{-8}\ M = 2.5 \times 10^{-7}$ M.
(c) 1M Ammonia. If the pH = 11.6, then
$[H^{\oplus}] = 10^{-11.6} = 2.5 \times 10^{-12}$ M. $[OH^{\ominus}] = K_w/[H^{\oplus}] =$
$1.0 \times 10^{-14}\ M^2/2.0x^{-12}\ M = 4 \times 10^{-3}$ M.

6. 

7. The total buffer species = [weak acid (HA)] + [conjugate base ($A^{\ominus}$)]

$$\text{Total buffer concentration} = 0.25\ \text{M} + 0.15\ \text{M} = 0.4\ \text{M}$$

The pH can be calculated from the pK_a and the concentrations given using the Henderson-Hasselbalch equation.

$$\text{pH} = \text{p}K_a + \log\frac{[A^{\ominus}]}{[HA]} = 3.90 + \log\frac{(0.15\ \text{M})}{(0.25\ \text{M})} = 3.90 - 0.22 = 3.68$$

8. The pK_a for the ionization of $H_2PO_4^{\ominus}$ is 7.2. The Henderson-Hasselbalch equation (Equation 2.18) indicates that when the concentrations of the acidic form ($H_2PO_4^{\ominus}$) and its conjugate base ($HPO_4^{2\ominus}$) are equivalent, the pH is equal to the pK_a, because the log term is zero (log 1 = 0). Therefore, mixing 50 milliliters of solution A with 50 milliliters of solution B gives a buffer of pH 7.2. Since the concentration of each solution is 0.02 M, mixing equal volumes gives a buffer whose phosphate concentration is also 0.02 M. The reason why this is an effective buffer is that the final pH is at the pK_a value. This means that the buffer will resist changes in pH over a considerable range.

9. (a) The effective range of a buffer is from approximately one pH unit below to one pH unit above the pK_a. The buffering range for MOPS is therefore 6.2–8.2, and the buffering range for SHS is 4.5–6.5. Use the Henderson-Hasselbalch equation to calculate the ratios of basic to acidic species.

For MOPS: $\text{pH} = \text{p}K_a + \log\dfrac{[R_3N]}{[R_3NH^{\oplus}]}$

$$6.5 = 7.2 + \log\frac{[R_3N]}{[R_3NH^{\oplus}]}$$

$$\frac{[R_3N]}{[R_3NH^{\oplus}]} = \frac{1}{5}$$

For SHS:

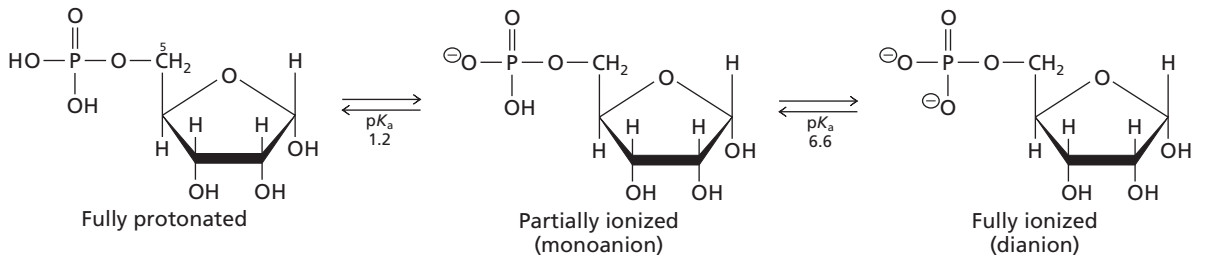
$$6.5 = 5.5 + \log\frac{[RCOO^{\ominus}]}{[RCOOH]}$$

$$\frac{[RCOO^{\ominus}]}{[RCOOH]} = \frac{10}{1}$$

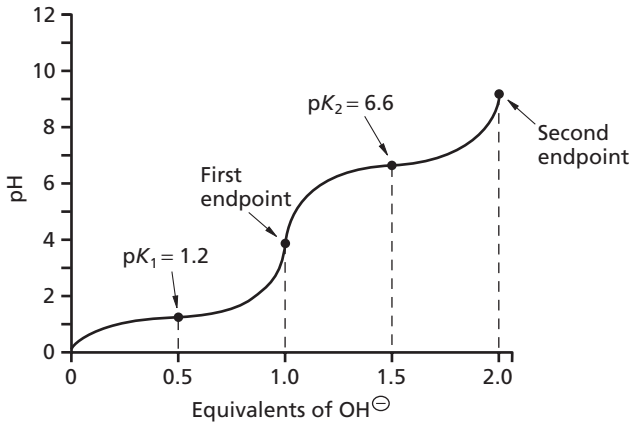
(b) An SHS buffer solution at pH 6.5 contains a much greater proportion of conjugate base relative to acid (10:1) than MOPS does (1:5). Therefore, an SHS buffer would more effectively maintain the pH upon addition of acid: $H^{\oplus} + RCOO^{\ominus} \rightleftharpoons RCOOH$. Conversely, a MOPS buffer at pH 6.5 contains a greater proportion of acid than SHS does; therefore, MOPS would more effectively maintain the pH upon addition of base: $R_3NH^{\oplus} + OH^{\ominus} \rightleftharpoons R_3N + H_2O$.

10.

(a)



(b)



11. Excess gaseous CO_2 rapidly equilibrates with aqueous CO_2 (Equation 2.25), leading to formation of carbonic acid (Equation 2.23). Carbonic acid ionizes to $H^{\oplus}$ and $HCO_3^{\ominus}$ (Equation 2.22). The excess acid, in the form of $H^{\oplus}$, can accumulate in bodily fluids, producing acidosis.

12. Although the metabolism of lactate and other organic acids in the diet can lead to production of CO_2 as shown, CO_2 is efficiently expired from the lungs (except during respiratory acidosis). Thus, the net product of the metabolic process is bicarbonate ($HCO_3^{\ominus}$), a base. Excess $H^{\oplus}$ present during metabolic acidosis can be removed when it combines with $HCO_3^{\ominus}$ to form H_2CO_3 (Equation 2.22), which then forms aqueous CO_2 and H_2O (Equation 2.23).

13. The acidic and conjugate base species of aspirin can be represented as RCOOH and $RCOO^{\ominus}$. Use the Henderson-Hasselbalch equation to calculate the ratio of the two species at pH 2.0 and pH 5.0. Then calculate the fraction of the total species that is unionized and available for absorption. In the stomach at pH 2.0,

$$\text{pH} = \text{p}K_a + \log\frac{[\text{RCOO}^{\ominus}]}{[\text{RCOOH}]}$$

$$2.0 = 3.5 + \log\frac{[\text{RCOO}^{\ominus}]}{[\text{RCOOH}]}$$

$$\frac{[\text{RCOO}^{\ominus}]}{[\text{RCOOH}]} = \frac{0.03}{1}$$

The percentage of the uncharged species (RCOOH) is equal to the amount of RCOOH divided by the total of RCOOH and $RCOO^{\ominus}$, times 100%.

$$\frac{[\text{RCOOH}]}{[\text{RCOOH}] + [\text{RCOO}^{\ominus}]} \times 100\% = \frac{1}{1 + 0.03} \times 100\% = 97\%$$

Therefore, nearly all aspirin in the stomach is in a form available for absorption. In the upper intestine at pH 5.0, however, only a small percentage of aspirin is available for absorption.

$$5.0 = 3.5 + \log\frac{[\text{RCOO}^{\ominus}]}{[\text{RCOOH}]}$$

$$\frac{[\text{RCOO}^{\ominus}]}{[\text{RCOOH}]} = \frac{32}{1}$$

$$\frac{[\text{RCOOH}]}{[\text{RCOOH}] + [\text{RCOO}^{\ominus}]} \times 100\% = \frac{1}{1 + 32} \times 100\% = 3\%$$

Note that aspirin must be in solution in order to be absorbed. For this reason, coated or slow-release forms of aspirin may alter the availability of aspirin in the stomach and intestine.

14. Use the Henderson-Hasselbach equation to calculate the ratio of the two species at each pH

At pH = 7.5

$$\text{pH} = \text{p}K_a + \log\frac{[H_2NCH_2CONH_2]}{[^{+}H_3NCH_2CONH_2]}$$

$$7.5 = 8.2 + \log\frac{[H_2NCH_2CONH_2]}{[^{+}H_3NCH_2CONH_2]}$$

$$\log\frac{[H_2NCH_2CONH_2]}{[^{+}H_3NCH_2CONH_2]} = 7.5 - 8.2 = -0.7$$

$$\frac{[H_2NCH_2CONH_2]}{[^{+}H_3NCH_2CONH_2]} = \frac{1}{5}$$

The ratio of $[H_2NCH_2CONH_2]$ to $[^{+}H_3NCH_2CONH_2]$ is 1 to 5. To determine the percent in the conjugate base form: 1/(1 + 5)*100 = 17%. Therefore, 17% is unprotonated at pH 7.5.

At pH = 8.2

$$\text{pH} = \text{p}K_a + \log\frac{[H_2NCH_2CONH_2]}{[^{+}H_3NCH_2CONH_2]}$$

$$8.2 = 8.2 + \log\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]}$$

$$\log\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]} = 8.2 - 8.2 = 0$$

$$\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]} = \frac{1}{1}$$

The ratio of $[H_2NCH_2CONH_2]$ to $[^+H_3NCH_2CONH_2]$ is 1.0 to 1.0. To determine the percent in the conjugate base form: 1/(1 + 1)*100 = 50%. Therefore, 50% is unprotonated at pH 8.2.

At pH 9.0:

$$pH = pK_a + \log\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]}$$

$$9.0 = 8.2 + \log\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]}$$

$$\log\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]} = 9.0 - 8.2 = 0.8$$

$$\frac{[H_2NCH_2CONH_2]}{[^+H_3NCH_2CONH_2]} = \frac{6.3}{1}$$

The ratio of $[H_2NCH_2CONH_2]$ to $[^+H_3NCH_2CONH_2]$ is 6.3 to 1. To determine the percent of the conjugate base: 6.3/(6.3 + 1)*100 = 86%. That is, 86% is unprotonated at pH 9.0.

15. This titration curve represents a compound with two pK_a values, shown by the two plateaus (near pH 2 and pH 10). Glycine has two pK_a values at 2.4 and at 9.8.

16. Only (a) vitamin C would be soluble in water. Vitamin C contains several hydroxyl groups, each of which can hydrogen-bond with water.

17. At 0°C the ion product for water is 1.14×10^{-15}. At neutral pH,

$$[H^{\oplus}] = [OH^{\ominus}] = \sqrt{1.14 \times 10^{-15}} = 3.38 \times 10^{-8}$$

$$pH = -\log(3.38 \times 10^{-8}) = 7.47$$

At 100°C

$$[H^{\oplus}] = [OH^{\ominus}] = \sqrt{4.0 \times 10^{-13}} = 6.32 \times 10^{-7}$$

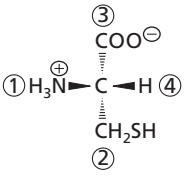
$$pH = -\log(6.32 \times 10^{-7}) = 6.2$$

Note that the density of water changes with temperature but this has very little effect on $[H^+]$.

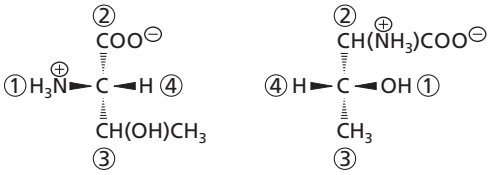
18. HCl dissociates completely in water. In 6 M HCl, $[H^{\oplus}] = 6$ M. The pH is $-\log(6) = -0.78$. The standard pH scale begins at zero ($[H^{\oplus}] = 1$ M) because it's very unusual to encounter more acidic solutions in biology.

Chapter 3 Amino Acids and the Primary Structures of Proteins

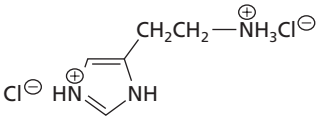
1. By comparing the priorities of L-cysteine (shown here) to those of L-serine (*S* configuration, page 57) you will find that their sequence is clockwise and therefore L-cysteine has the *R* configuration.



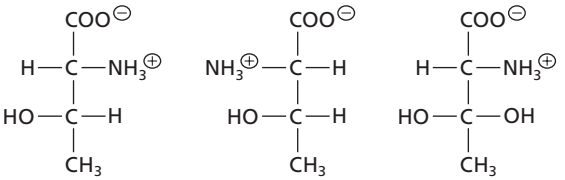
2. The stereochemistry of each chiral carbon must be examined to determine whether it has the *R* or *S* configuration.



C-2, *S*-configuration C-3, *R*-configuration



3. The other stereoisomers are:

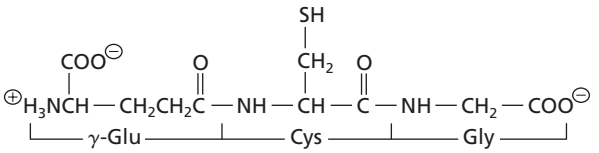


D-Threonine L-Allothreonine D-Allothreonine

4. Methionine.

5. (a) Serine; phosphorylation of the hydroxyl group.
(b) Glutamate; carboxylation of the γ-carbon.
(c) Lysine; acetylation of the ε-amino group.

6. By convention, peptides are designated from the N-terminus → C-terminus, therefore Glu is the N-terminus and Gly is the C-terminus.

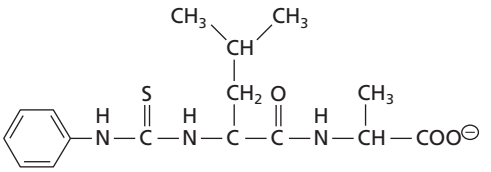


7. The 6 residues at the C-terminus of melittin are highly hydrophilic (Table 3.1). Of the remaining 20 amino acid residues, nearly all are hydrophobic, including 9 with highly hydrophobic side chains (leucine, isoleucine, valine). The hydrophilic portion of melittin is more soluble in aqueous solution, while the hydrophobic portion is more soluble in the membrane lipids.

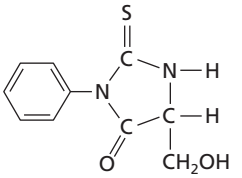
8. Use Table 3.2 to determine the net charge at each pK_a value. The pH at which the net charge is 0 lies midway between the two pK_a values at which the average charges are +0.5 and −0.5.
(a) At pH 9.0, the net charge of arginine is +0.5, and at pH 12.5, the net charge is −0.5. Therefore, $pI_{Arg} = (9.0 + 12.5) \div 2 = 10.8$.
(b) At pH 2.1, the net charge of glutamate is +0.5, and at pH 4.1, the net charge is −0.5. Therefore, $pI_{Glu} = (2.1 + 4.1) \div 2 = 3.1$.

9. The ionizable groups are the free amino group of the N-terminal cysteine residue (pK_a = 10.7), the glutamate side chain (pK_a = 4.1), and the histidine side chain (pK_a = 6.0).
(a) At pH 2.0, the N-terminus and the histidine side chain have positive charges and the glutamate side chain is uncharged. The net charge is +2.
(b) At pH 8.5, the N-terminus has a positive charge, the histidine side chain is uncharged, and the glutamate side chain has a negative charge. The net charge is 0.
(c) At pH 10.7, the charge of the N-terminus is +0.5, the histidine side chain is uncharged, and the glutamate side chain has a negative charge. The net charge is −0.5.

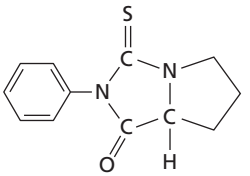
10. (a)



(b)

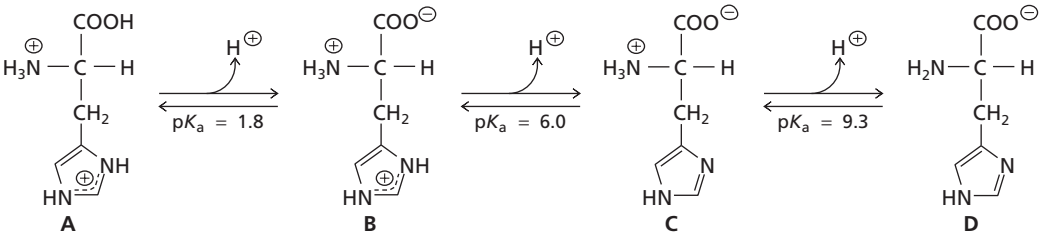


(c)



11. (a) Gly–Ala–Trp–Arg, Asp–Ala–Lys, Glu–Phe–Gly–Gln
(b) Gly–Ala–Trp, Arg–Asp–Ala–Lys–Glu–Phe, Gly–Gln
(c) Gly–Ala–Trp–Arg–Asp, Ala–Lys–Glu, Phe–Gly–Gln

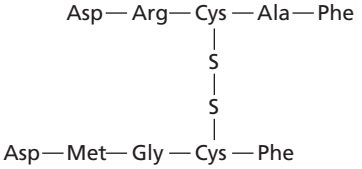
12. (a)



(b) A, 1; B, 3; C, 5; D, 7
(c) 1, 4, 5, 7
(d) 4
(e) 5
(f) Histidine would be a good buffer within one pH unit of any of its three pK_a values: 0.8–2.8, 5.0–7.0, and 8.3–10.3.

13. (a) Because there are two N-terminal groups, there must be two peptide chains, each having an N-terminal aspartate residue.
(b) 2-Mercaptoethanol reduces disulfide bonds, and trypsin catalyzes cleavage on the carboxyl side of arginine residues. Since aspartate is found at both N-termini of FP, the sequence of the dipeptide is Asp–Arg, and the sequence of the pentapeptide is Asp–(Cys, Gly, Met, Phe). The tripeptide has the sequence Cys–(Ala, Phe) and is derived from trypsin–catalyzed cleavage of a pentapeptide whose sequence is Asp–Arg–Cys–(Ala, Phe).
(c) The C-terminal residue of each peptide chain is phenylalanine. Now that the terminal residues are known, one peptide must have the sequence Asp–(Cys, Gly, Met)–Phe, and the other must have the sequence Asp–Arg–Cys–Ala–Phe.
(d) CNBr cleaves on the carbonyl side of methionine residues to produce C-terminal homoserine lactone residues. The peptides are therefore Asp–Met and (Cys, Gly)–Phe. Glycine is the N-terminal residue of the tripeptide, so that pentapeptide sequence is Asp–Met–Gly–Cys–Phe.

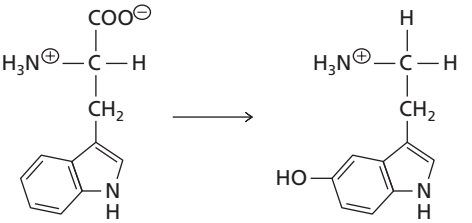
The complete FP structure is



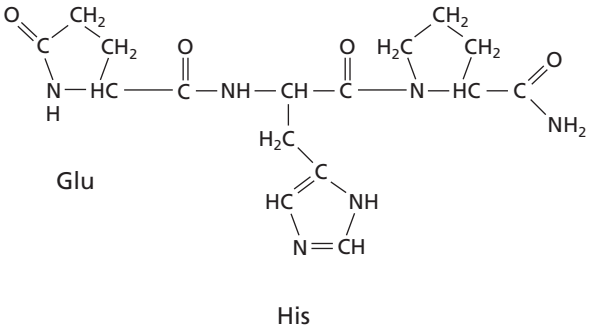
14. (a) The substitution of aspartate (D) for glutamate (E) at position 50 is an example of a conservative change. The amino acids aspartate and glutamate both contain acidic side chains that are negatively charged at physiological pH.
(b) The substitution of tyrosine (Y) for histidine (H) is an example of a nonconservative substitution since tyrosine contains an aromatic side chain and histidine contains a hydrophilic side chain consisting of an imidazole group.

15. The neurotransmitter serotonin is derived from the amino acid tryptophan.

In the conversion, the carboxyl group from tryptophan is removed and a hydroxyl group is added to the aromatic ring.



16. (a) There are two peptide bonds present in TRH. They are marked with the dashed lines.
(b) TRH is derived from the tripeptide Glu–His–Pro. The proline carboxyl group has been modified to an amide (marked with an *). The side chain carboxyl group of the amino terminal Glu forms an amide with the residue's α-amino group (marked with a **).
(c) The amino- and carboxyl-terminal groups have been modified to amide groups and thus are uncharged.



17. (a) L-Dopa is in the *S* configuration.
(b) They are both derived from the amino acid tyrosine.

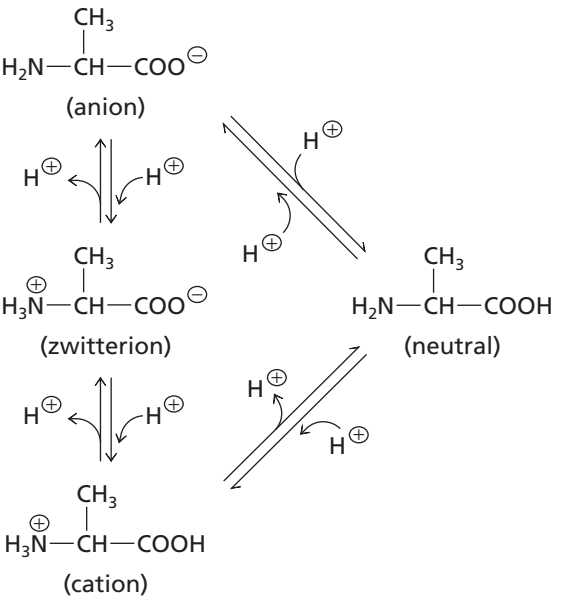
18. Although Figure 3.6 shows only three forms of alanine, there are actually four different forms in equilibrium (see next page). The neutral form will be present at very low concentrations because at any given pH the three other forms are much more stable. We can calculate the relative ratios of the four forms by assuming that the protonation/deprotonation of the two charged groups is independent.

For alanine at pH 2.4 the relative ratio of $R—COO^{\ominus}$ and R—COOH is

$$2.4 = 2.4 + \log\frac{[R—COO^{\ominus}]}{[R—COOH]} \text{ therefore } \frac{[R—COO^{\ominus}]}{[R—COOH]} = 1$$

and the ratio of $H_3N^{\oplus}—R$ to $H_2N—R$ is

$$2.4 = 9.9 + \log\frac{[H_2N—R]}{[H_3N^{\oplus}—R]} \text{ therefore } \frac{[H_2N—R]}{[H_3N^{\oplus}—R]} = 3.1 \times 10^{-8}$$



Thus the relative ratios of the four forms are approximately

$$\text{cation : zwitterion : anion : neutral} \quad 1 : 1 : 10^{-8} : 10^{-8}$$

and the concentration of the neutral form in a 0.01 M solution of alanine is about 10^{-10} M. Neutral molecules exist but their concentration is insignificant.

At pH 9.9 the ratios are

$$\text{anion : zwitterion : cation : neutral} \quad 1 : 1 : 10^{-8} : 10^{-8}$$

At pH 6.15 the relative ratio of $R—COO^{\ominus}$ and R—COOH is

$$6.15 = 2.4 + \log \frac{[R—COO^{\ominus}]}{[R—COOH]} \text{ therefore } \frac{[R—COO^{\ominus}]}{[R—COOH]} = 5.6 \times 10^3$$

and the relative ratio of $H_2N—R$ and $[H_3N^{\oplus}—R]$ is

$$6.15 = 9.9 + \log \frac{[H_2N—R]}{[H_3N^{\oplus}—R]} \qquad \frac{[H_2N—R]}{[H_3N^{\oplus}—R]} = 1.8 \times 10^{-4} \qquad \frac{[H_3N^{\oplus}—R]}{[H_2N—R]} = 5.6 \times 10^{-8}$$

The zwitterion is present in 5600-fold excess over the anion and cation forms and each of these forms is 5600-fold more likely than the neutral form. The ratios are

$$\text{zwitterion : anion : cation : neutral} \quad 3.1 \times 10^7 : 1 : 1.8 \times 10^{-4} : 1.8 \times 10^{-4} : 3.2 \times 10^{-8}$$

The concentration of the neutral form in a solution of 0.01 M alanine is insignificant.

19. The relative concentrations of the zwitterion and the cation are

$$2.4 = 2.4 + \log \frac{[H_3N^{\oplus}—\overset{CH_3}{\overset{|}{C}}—COO^{\ominus}]}{[H_3N^{\oplus}—\overset{CH_3}{\overset{|}{C}}—COOH]} \qquad \frac{[H_3N^{\oplus}—\overset{CH_3}{\overset{|}{C}}—COO^{\ominus}]}{[H_3N^{\oplus}—\overset{CH_3}{\overset{|}{C}}—COOH]} = 1$$

Thus the concentration of the zwitterion in a solution of 0.01 M alanine is 0.005 M. (We can ignore the concentrations of the anion and neutral forms—see previous question.)

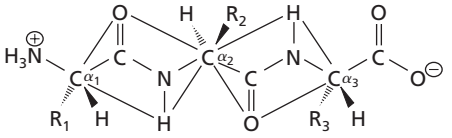
At pH 4.0

$$4.0 = 2.4 + \log \frac{[\text{zwitterion}]}{[\text{cation}]} \qquad \frac{[\text{zwitterion}]}{[\text{cation}]} = 40$$

The concentration of the zwitterion is 0.01 M $\times \frac{40}{41} = 0.00976$ M

Chapter 4 Proteins: Three-Dimensional Structure and Function

1. (a)

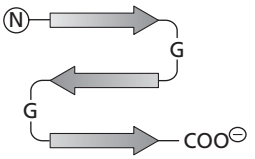


(b) The R groups represent the side chains of the amino acid residues.
(c) The partial double-bond character of the C—N amide bonds prevents free rotation.
(d) Both peptide groups in this tripeptide are in the *trans* conformation, since the α-carbon atoms are on opposite sides of the peptide bonds.
(e) The peptide groups may rotate around the N—C_α and C_α—C bonds.

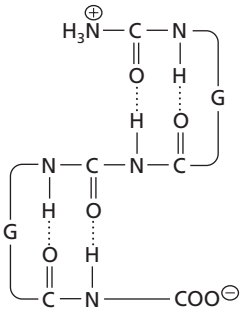
2. (a) (1) In an α helix, intrachain hydrogen bonds form between carbonyl oxygens of certain residues and amide hydrogens of other residues. The hydrogen bonds are approximately parallel to the helix axis (Figure 4.10).
(2) In a collagen triple helix, interchain hydrogen bonds form between amide hydrogens of the glycines in one chain and carbonyl oxygen atoms of residues (which are often proline) in an adjacent chain (Figure 4.41). There are no intrachain hydrogen bonds in a collagen helix.
(b) The side chains of an α helix point outward from the cylinder of the helix (Figure 4.11). In collagen, three chains coil around each other so that every third residue of a given chain makes contact with the other two chains along the central axis of the triple helix (Figure 4.42). Only the small side chain of glycine can fit at these positions. The other side chains point outward from the triple helical coil.

3. (1) The presence of glycine in an α helix destabilizes the helix due to the greater freedom of movement allowed by the small side chain. For this reason, many α helices begin or end with glycine.
(2) Proline tends to disrupt α helices because its rigid, cyclic side chain stereochemically interferes with the space that would normally be occupied by a neighboring residue in the α helix. In addition, proline lacks a hydrogen on its amide nitrogen and cannot participate in normal intrahelical hydrogen bonding.

4. (a) Due to the flexibility resulting from a small side chain (—H), glycine is often found in "hairpin loops" that connect sequential antiparallel β strands. The glycine residues (G) in positions 8 and 14 provide two hairpin-loop regions to connect the three β strands in Betanova.



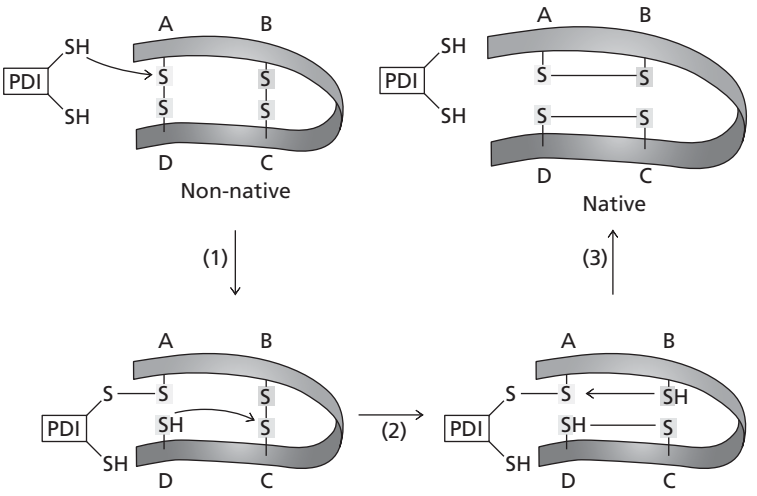
(b) β-sheet structures are stabilized by hydrogen bonds that form between a carbonyl oxygen of one strand and an amide nitrogen of an adjacent strand (Figure 4.15).



5. Helix-loop-helix (HLH) motif (Figure 4.19).

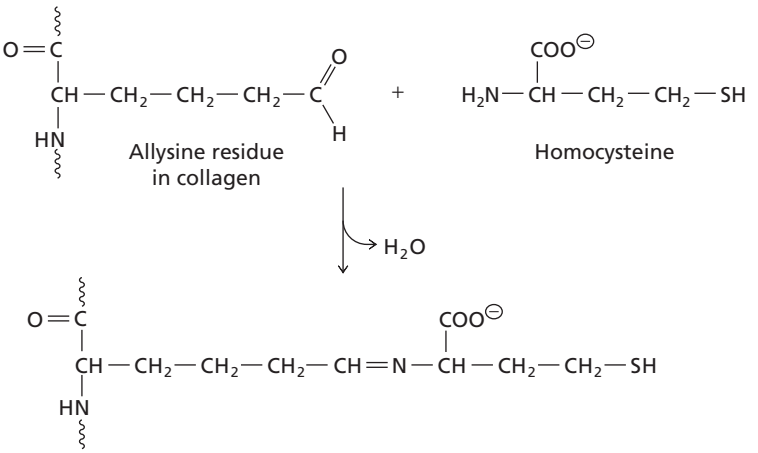
6. (a) α/β. Regions of α helix and β strand alternate in the polypeptide chain.
(b) α/β barrel. Parallel β strands are surrounded by a layer of α helices in a cylindrical shape.
(c) Yeast FMN oxidoreductase and *E. coli* enzyme required for tryptophan biosynthesis (Figure 4.24 (i) and (j) respectively).

7. Protein disulfide isomerase contains two reduced cysteine residues at the active site, and these participate in a reduction and disulfide exchange that allows the misfolded protein to refold into the lower energy native conformation.



8. The highly hydrophobic side chains of methionine, leucine, phenylalanine, and isoleucine are most likely to be on the side of the helix that faces the interior of the protein. Most of the other side chains are polar or charged and can interact with the aqueous solvent. Since the α helix is a repeating structure with approximately 3.6 residues per turn, the hydrophobic groups must be found every three or four residues along the sequence, so that one side of the helix is hydrophobic.

9. Covalent cross-linking contributes significantly to the strength and rigidity of collagen fibers. In one type of cross-link, allysine residues in a collagen molecule condense with lysine residues in an adjacent molecule, forming Schiff bases (Figure 4.38a). When an allysine residue reacts with homocysteine, it is unable to participate in the normal cross-linking of collagen molecules. High levels of homocysteine in blood probably lead to defective collagen structure and skeletal deformities.



10. The sequence –Gly–Pro–X–Y– occurs frequently in collagen, which is found throughout the body, including in the skin. Because the larval enzyme can catalyze cleavage of collagen chains, the parasite is able to enter the host.

11. The reaction of carbon dioxide with water explains why there is a concomitant lowering of pH when the concentration of CO_2 increases. Carbon dioxide produced by rapidly metabolizing tissue reacts with water to produce bicarbonate ions and $H^{\oplus}$.

(a) $CO_2 + H_2O \longleftrightarrow H_2CO_3 \longleftrightarrow HCO_3^{\ominus} + H^{\oplus}$

The $H^{\oplus}$ generated in this reaction decreases the pH of the blood and thus stabilizes the deoxy form (T conformation) of hemoglobin. The net effect is an increase in the P_{50}, that is, a lower affinity of hemoglobin for oxygen, so that more oxygen is released to the tissue (Figure 4.50). Carbon dioxide also lowers the affinity of hemoglobin for oxygen by forming

carbamate adducts with the N-termini of the four chains (Figure 4.51). These adducts contribute to the stability of the deoxy (T) conformation, thereby further increasing the P_{50} and promoting the release of oxygen to the tissue.

(b) Shock victims suffer a critical deficit of oxygen supply to their tissues. Bicarbonate administered intravenously provides a source of carbon dioxide to the tissues. By lowering the affinity of hemoglobin for oxygen, carbon dioxide facilitates a release of oxygen from oxyhemoglobin to the tissues.

12. (a) 2,3BPG binds to positively charged side chains in the central cavity of deoxyhemoglobin (Figure 4.49). Since Hb F lacks two positively charged groups (His-143 of each β chain), 2,3BPG binds less tightly to Hb F than to Hb A.

(b) 2,3BPG stabilizes the deoxy form of hemoglobin, increasing the fraction of molecules in the deoxy form. Since Hb F binds 2,3BPG less tightly than does Hb A, Hb F is less affected by 2,3BPG in the blood and has a greater fraction of molecules in the oxy form. Hb F therefore has a greater affinity than Hb A for oxygen at any oxygen pressure.

(c) At the oxygen pressure of tissues, 20–40 torr, Hb F has a greater affinity for oxygen than does Hb A. The difference in affinity allows efficient transfer of oxygen from maternal blood to the fetus.

13. The low P_{50} value of Hb_{Yakima} indicates a greater than normal affinity for oxygen even at the oxygen pressures found in working muscle. The increased affinity means that Hb_{Yakima} gives up less oxygen to the working muscle.

14. (a) Hydrophilic (italicized) and hydrophobic (underlined) residues are identified:
*E*CG*K*<u>F</u><u>M</u><u>W</u>*K*C*K*N*S*N*D*C*C*<u>*K*</u>*D*<u>L</u><u>V</u>C*S*S*R*<u>W</u>*K*<u>W</u><u>C</u><u>V</u><u>L</u>*A*S<u>P</u><u>F</u>

(b) In the three-dimensional structure of proteins, amino acids that are far from each other in the primary sequence can interact in the globular structure of the protein. Thus the hydrophobic amino acids can be very close to each other in the three-dimensional structure and provide a "hydrophobic" face for interaction with the membrane.

15. (a) The most effective binding of selenoprotein P to heparin is seen at a pH below 6. The binding of selenoprotein P to heparin decreases as the pH is increased to 7. There is very little binding of selenoprotein P to heparin at pH values greater than 7.

(b) Heparin is negatively charged. If selenoprotein P is positively charged, it can bind to heparin. Histidine residues are abundant in selenoprotein P. Histidine has an imidazole side chain that has a pK_a value of 6.0. That is, at a pH of 6.0, 50% of the histidine residues would be protonated and positively charged and 50% would be unprotonated and uncharged. Below a pH of 6.0, there would be a net positive charge on the histidine residues, resulting in effective electrostatic interactions with the heparin. At pH values above 7, almost all of the histidine residues would be unprotonated and uncharged and will not effectively interact with the negatively charged heparin molecule.

16. Collagen is protein consisting of three polypeptide chains that are wound together in a triple helix. The protease bromelin is an enzyme that cleaves some of the peptide bonds in the polypeptide chains. The polypeptide chains are necessary to trap the water molecules in a semisolid state when gelatin cools, and if these are cleaved, the gelatin will not set properly. The cleavage of the polypeptide chains in collagen by bromelin destroys the ability of the gelatin to harden. If the pineapple is first cooked, the heat will denature the protein and thus the enzyme activity will be destroyed. Therefore cooked pineapple can be added to slightly thickened gelatin, and the gelatin will proceed to the semisolid state as desired. (Assume that heat denaturation is irreversible.)

17. The replacement of lysine by methionine results in one less positive charge on each beta subunit in the central cavity (see Figure 4.49). 2,3BPG binds less tightly to HbH. This causes more of the mutant protein to be in the R state (oxyhemoglobin is stabilized). The curve is shifted towards the left (more like myoglobin). Since more is in the R state, the affinity for oxygen has increased.

Chapter 5 Properties of Enzymes

1. The initial velocities are approaching a constant value at the higher substrate concentrations, so we can estimate the V_{max} as 70 mM/min. Since K_m equals the concentration of substrate [S] required to reach half the maximum velocity, we can estimate the K_m to be 0.01 M since that's the concentration of substrate that yields a rate of 35 mM/min ($= V_{max}>2$).

2. (a) The ratio k_{cat}/K_m, or *specificity constant*, is a measure of the preference of an enzyme for different substrates. When two substrates at the same concentration compete for the

active site of an enzyme, the ratio of their rates of conversion to product is equal to the ratio of the k_{cat}/K_m values, since $v_0 = (k_{cat}/K_m)[E][S]$ for each substrate and [E] and [S] are the same.

$$\frac{v_0(S_1)}{v_0(S_2)} = \frac{(k_{cat}>K_m)^1>[E][S]}{(k_{cat}>K_m)^2>[E][S]}$$

(b) The upper limit of k_{cat}/K_m approaches 10^8 to 10^9 s^{-1}, the fastest rate at which two uncharged molecules can approach each other by diffusion at physiological temperatures.

(c) The catalytic efficiency of an enzyme cannot exceed the rate for the formation of ES from E and S. The most efficient enzymes have k_{cat}/K_m values approaching the rate at which they encounter a substrate. At this limiting velocity they have become as efficient catalysts as possible because every encounter produces a reaction. (Most enzymes don't need to catalyze reactions at the maximum possible rates so there's no selective pressure to evolve catalytically perfect enzymes.)

3. The catalytic constant (k_{cat}) is the first-order rate constant for the conversion of ES to E + P under saturating substrate concentrations (Equation 5.26), and CA has a much higher catalytic activity in converting substrate to product than does OMPD. However, the efficiency of an enzyme can also be measured by the *rate acceleration* provided by the enzyme over the corresponding uncatalyzed reaction (k_{cat}/k_n, Table 5.2). The reaction of the substrate for OMPD in the absence of enzyme is very slow ($k_n = 3 \times 10^{-16}$ s^{-1}) compared to the reaction for the CA substrate in the absence of enzyme ($k_n = 1 \times 10^{-1}$ s^{-1}). Therefore, while the OMPD reaction is much slower than the CA reaction in terms of k_{cat}, OMPD is one of the most efficient enzymes known and provides a much higher rate acceleration than does CA when the reactions of each enzyme are compared to the corresponding uncatalyzed reactions.

4. When [S] = 100 μM, [S] $\gg K_m$, so $v_0 = V_{max} = 0.1$ μM min^{-1}.

(a) For any substrate concentration greater than 100 μM, $v_0 = V_{max} = 0.1$ μM min^{-1}.

(b) When [S] = K_m, $v_0 = V_{max}/2$, or 0.05 μM min^{-1}.

(c) Since K_m and V_{max} are known, the Michaelis-Menten equation can be used to calculate v_0 at any substrate concentration. For [S] = 2 μM,

$$v_0 = \frac{V_{max}[S]}{K_m + [S]} = \frac{(0.1\ \mu M\ min^{-1})(2\ \mu M)}{(1\ \mu M + 2\ \mu M)} = \frac{0.2}{3}\mu M\ min^{-1} = 0.067\ \mu M\ min^{-1}$$

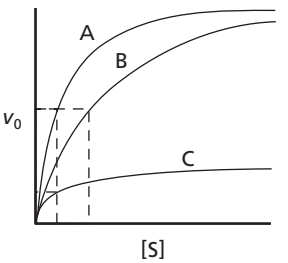
5. (a) Determine $[E]_{total}$ in moles per liter, then calculate V_{max}.

$$[E]_{total} = 0.2\ g\ l^{-1}\left(\frac{1\ mol}{21\ 500\ g}\right) = 9.3 \times 10^{-6}\ M$$

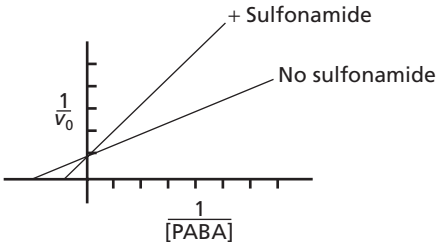
$$V_{max} = k_{cat}[E]_{total} = 1000\ s^{-1}(9.3 \times 10^{-6}\ M) = 9.3 \times 10^{-3}\ M\ s^{-1}$$

(b) Since V_{max} is unchanged in the presence of the inhibitor, competitive inhibition is occurring. Because the inhibitor closely resembles the heptapeptide substrate, competitive inhibition by binding to the enzyme active site is expected (i.e., classical competitive inhibition).

6. Curve A represents the reaction in the absence of inhibitors. In the presence of a competitive inhibitor (curve B), K_m increases and V_{max} is unchanged. In the presence of a noncompetitive inhibitor (curve C), V_{max} decreases and K_m is unchanged.

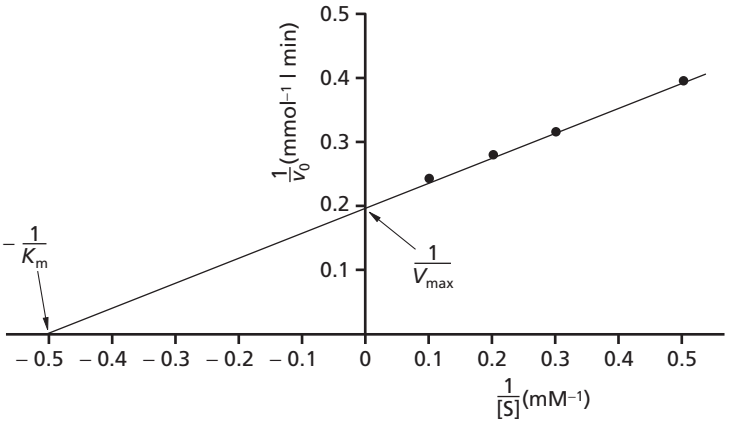


7. Since the inhibitor sulfonamides structurally resemble the PABA substrate we would predict that sulfonamides bind to the enzyme active site in place of PABA and act as competitive inhibitors (Figure 5.9).



8. (a) To plot the kinetic data for fumarase, first calculate the reciprocals of substrate concentrations and initial rates of product formation. (Note the importance of including correct units in calculating and plotting the data.)

Fumarate [S] (mM)	$\frac{1}{[S]}$ (mM^{-1})	Rate of product formation v_0 (mmol l^{-1} min^{-1})	$\frac{1}{v_0}$ (mmol^{-1} l min)
2.0	0.50	2.5	0.40
3.3	0.30	3.1	0.32
5.0	0.20	3.6	0.28
10.0	0.10	4.2	0.24



V_{max} is obtained by taking the reciprocal of $1/V_{max}$ from the *y* intercept (Figure 5.6).

$$1/V_{max} = 0.20 \text{ mmol}^{-1} \text{ l min, so } V_{max} = 5.0 \text{ mmol l}^{-1} \text{ min}^{-1}$$

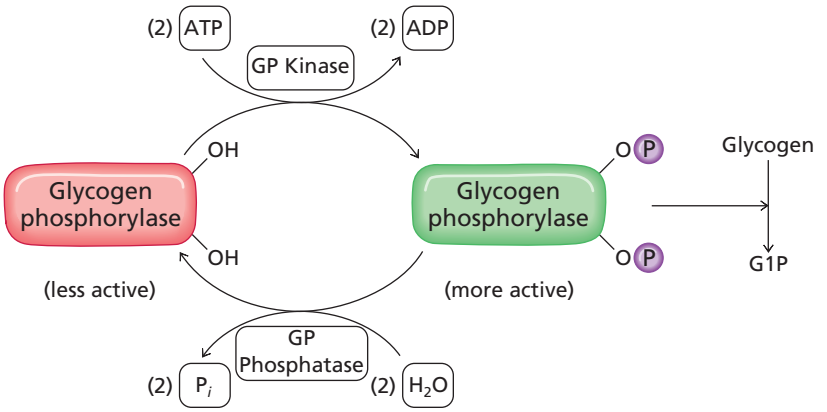
K_m is obtained by taking the reciprocal of $-1/K_m$ from the *x* intercept.

$$-1/K_m = -0.5 \text{ mM}^{-1}, \text{ so } K_m = 2.0 \text{ mM or } 2 \times 10^{-3} \text{ M}$$

(b) The value of k_{cat} represents the number of reactions per second that *one enzyme active site* can catalyze. Although the concentration of enzyme is 1×10^{-8} M, fumarase is a tetramer with four active sites per molecule so the total concentration of enzyme active sites $[E_{total}]$ is 4×10^{-8} M. Using Equation 5.26:

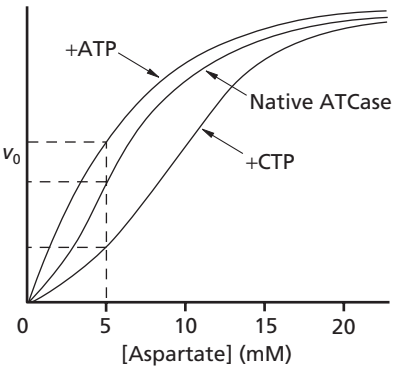
$$k_{cat} = \frac{V_{max}}{[E_{total}]} = \frac{5.0 \text{ mmol l}^{-1} \text{ min}^{-1}}{4 \times 10^{-5} \text{ mmol l}^{-1}} \times \frac{1 \text{ min}}{60 \text{ s}} = 2 \times 10^3 \text{ s}^{-1}$$

9. Like pyruvate dehydrogenase (PDH) (Figure 5.22), glycogen phosphorylase (GP) activity is regulated by alternate phosphorylation by a *kinase* and dephosphorylation by a *phosphatase*. However, unlike PDH, the active form of GP has two phosphorylated serine residues; in the inactive GP form, two serine residues are not phosphorylated.



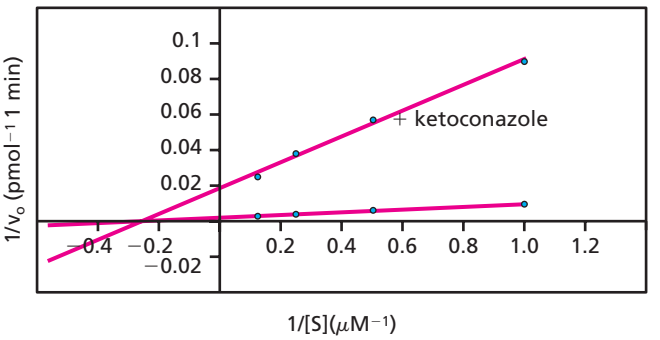
10. Inhibition of the first committed step of a multistep pathway allows the pathway to proceed only when the end product is needed. Since the first committed step is regulated, flux in the pathway is controlled. This type of regulation conserves raw material and energy.

11. When [aspartate] = 5 mM, $v_0 = V_{max}/2$. Therefore, in the absence of allosteric modulators, K_m = [S] = 5 mM. ATP increases v_0, and CTP decreases v_0.



12. (a) To plot the kinetic data for P450 3A4, first calculate the reciprocals of substrate concentrations and initial rates of product formation. The data are plotted in the double reciprocal plot and are shown with the dashed line.

Midazolam [S](μM)	1/[S] (μM^{-1})	Rate of product formation v_0 (pmol l^{-1} min^{-1})	$1/v_0$ (pmol^{-1} l min)
1	1	100	0.01
2	0.5	156	0.0064
4	0.25	222	0.0045
8	0.125	323	0.0031



V_{max} is obtained by taking the reciprocal of $1/V_{max}$ from the y intercept (Figure 5.6).

$$1/V_{max} - 0.0025 \text{ pmol}^{-1} \text{ l min, so } V_{max} = 400 \text{ pmol l}^{-1} \text{ min}^{-1}$$

K_m is obtained by taking the reciprocal of $-1/K_m$ from the x intercept

$$-1/K_m = -0.3\ \mu M^{-1}, \text{ so } K_m = 3.3\ \mu M$$

(b) The reciprocals of the substrate concentration and activity in the presence of ketoconazole are given in the table.

Midazolam [S] (μM)	1/[S] (μM^{-1})	Rate of product formation in the presence of 0.1 μM ketoconazole/ v_0 (pmol l^{-1} min^{-1})	$1/v_0$ (pmol^{-1} l min)
1	1	11	0.091
2	0.5	18	0.056
4	0.25	27	0.037
8	0.125	40	0.025

The plot of the data (solid line) is given in the double reciprocal plot shown in (a). There is an increase in the y intercept and no apparent change in the x intercept. From the double reciprocal plot, it appears that ketoconazole is a noncompetitive inhibitor (see Figure 5.11). These inhibitors are characterized by an apparent decrease in V_{max} (increase in $1/V_{max}$) with no change in K_m.

13. (a) Bergamottin appears to inhibit the activity of P450 3A4 since the P450 activity measured in the presence of 0.1 and 5 μM bergamottin is less than that of the P450 activity in the absence of bergamottin.

(b) It might be dangerous for a patient to take their medication with grapefruit juice since there appears to be an inhibition of P450 activity in the presence of bergamottin. If the bergamottin decreases the P450 activity, and the P450 enzyme is known to metabolize the drug to an inactive form, the time it takes to convert the drug to its inactive form may be increased. This may prolong the effects of the drug, which may lead to adverse consequences for the patient.

14. (a) When $[S] \gg K_m$, then $K_m + [S] \approx [S]$. Substrate concentration has no effect on velocity, and $v_0 = V_{max}$, as shown in the upper part of the curve in Figure 5.4a.

$$v_0 = \frac{V_{max}[S]}{K_m + [S]} \approx \frac{V_{max}[S]}{[S]} = V_{max}$$

(b) When $[S] \ll K_m$, $K_m + [S] \approx K_m$, and the Michaelis-Menten equation simplifies to

$$v_0 = \frac{V_{max}[S]}{K_m + [S]} \approx \frac{V_{max}[S]}{K_m}$$

Velocity is related to [S] by a constant value, and the reaction is first order with respect to S, as shown in the lower part of the curve in Figure 5.4a.

(c) When $v_0 = V_{max}/2$, $K_m = [S]$.

$$v_0 = \frac{V_{max}}{2} = \frac{V_{max}[S]}{K_m + [S]}$$

$$K_m + [S] = 2[S]$$

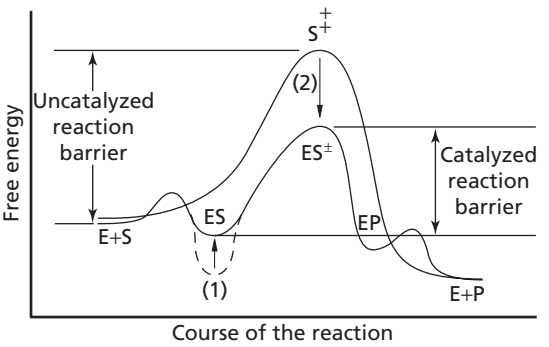
$$K_m = [S]$$

Chapter 6 Mechanisms of Enzymes

1. (a) The major binding forces in ES complexes include charge–charge interactions, hydrogen bonds, hydrophobic interactions, and van der Waals forces. (About 20% of enzymes bind a substrate molecule or part of it covalently.)

(b) Tight binding of a substrate would produce an ES complex that lies in a thermodynamic pit, effectively increasing the activation energy and thereby slowing down the reaction. Tight binding of the transition state, however, lowers the energy of the $ES^{\ddagger}$ complex, thereby decreasing the activation energy and increasing the rate of the reaction.

2. The activation barrier for the reaction is lowered by (1) raising the ground-state energy level (ES) and (2) lowering the transition-state energy level ($ES^{\ddagger}$), resulting in a reaction rate increase.



3. The rate determining step of a multistep reaction is the slowest step, which is the step with the highest activation energy. For Reaction 1, Step 2 is the rate determining step. For Reaction 2, Step 1 is the rate determining step.

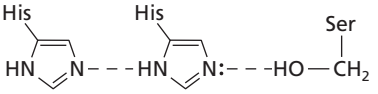
4. The reactive groups in Reaction 2 (—OH and —COOH) are held at close *proximity*. They are oriented in a manner suitable for catalysis by steric crowding of the bulky methyl groups of the ring. The reactive —COOH group cannot rotate away as freely as it can in Reaction 1. Model systems such as these are relevant because they indicate potential rate increases that might be obtained by enzymes that bring substrates and the enzyme's catalytic groups into positions that are optimal for reaction.

5. (1) Binding effects. Lysozyme binds the substrate so that the glycosidic bond to be cleaved is very close to both of the enzyme catalytic groups (Glu-35 and Asp-52). In addition, the energy of the ground-state sugar ring is raised because it is distorted into a half-chair conformation.
(2) Acid–base catalysis. Glu-35 first donates a proton to an oxygen of the leaving sugar (general acid catalysis), and then accepts a proton from the attacking water molecule (general base catalysis).
(3) Transition-state stabilization. Asp-52 stabilizes the developing positive charge on the oxocarbocation intermediate, and subsite D favors the half-chair sugar conformation of this intermediate. The structure proposed for the transition state includes both this charge and sugar conformation in addition to hydrogen bonding to several active-site residues.

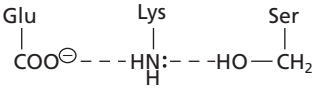
6. Serine 195 is the only serine residue in the enzyme that participates in the catalytic triad at the active site of α-chymotrypsin. The resulting increase in the nucleophilic character of Ser-195 oxygen allows it to react rapidly with DFP.

7. (a) The catalytic triad is composed of an aspartate, a histidine, and a serine residue. Histidine acts as a general acid–base catalyst, removing a proton from serine to make serine a more powerful nucleophile in the initial step. Aspartate forms a low-barrier hydrogen bond with histidine, stabilizing the transition state. An acid catalyst, histidine donates a proton to generate the leaving amine group.
(b) The oxyanion hole contains backbone —NH— groups that form hydrogen bonds with the negatively charged oxygen of the tetrahedral intermediate. The oxyanion hole mediates transition-state stabilization since it binds the transition state more tightly than it binds the substrate.
(c) During catalysis, aspartate forms a low-barrier hydrogen bond with the imidazolium form of histidine. Because asparagine lacks a carboxylate group to form the stabilizing hydrogen bond with histidine, enzyme activity is dramatically decreased.

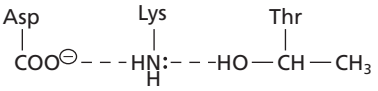
8. (a) Human cytomegalovirus protease: His, His, Ser



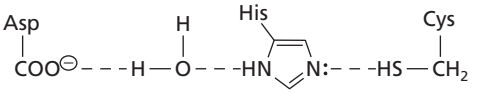
(b) β-Lactamase: Glu, Lys, Ser



(c) Asparaginase: Asp, Lys, Thr

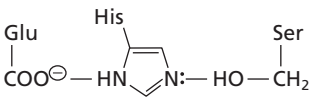


(d) Hepatitis A protease:

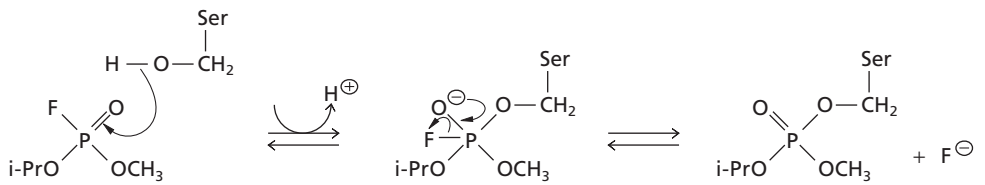


9. When tyrosine was mutated to phenylalanine, the activity of the mutant enzyme was less than 1% of the wild-type enzyme. Thus, the tyrosine residue is involved in the catalytic activity of DDP-IV. Tyrosine contains an -OH group on the aromatic ring of the side chain. As previously stated, this tyrosine is found in the oxyanion hole of the active site. Hydrogen bonds in the oxyanion hole of serine proteases are known to stabilize the tetrahedral intermediate. Tyrosine with an -OH group on the side chain can form a hydrogen bond and stabilize the tetrahedral intermediate. Phenylalanine does not have a side chain that can form a hydrogen bond. Therefore, the tetrahedral intermediate will not be stabilized resulting in a loss of enzyme activity.

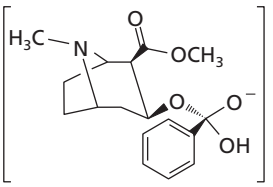
10. (a) Acetylcholinesterase catalytic triad: Glu-His-Ser



(b)



11. Transition-state analogs bound to carrier proteins are used as antigens to induce the formation of antibodies with catalytic activity. The tetrahedral phosphonate ester molecule is an analog of the tetrahedral intermediate structure in the transition state for hydrolysis of the benzyl ester moiety of cocaine. An antibody raised against the phosphonate structure that was able to stabilize the transition state of the cocaine benzyl ester hydrolysis could effectively catalyze this reaction.



Transition state

12. (a) Wild-type α1-proteinase inhibitor is given as treatment to individuals who produce an α1-proteinase inhibitor with substitutions in the amino acid sequence. These changes result in a protein that does not effectively inhibit the protease elastase. Uncontrolled elastase activity leads to increased breakdown of elastin, leading to destructive lung disease. Therefore, these patients are given a functional elastase inhibitor.

(b) The treatment for α1-proteinase inhibitor deficiency is to administer the wild-type protein intravenously. If the protein is given orally, the enzymes present in the digestive tract will cleave the peptide bonds in the α1-proteinase inhibitor. By administering the drug directly into the bloodstream, the protein can circulate to the lungs to act at the site of the neutrophil elastase.

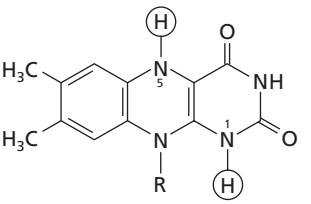
Chapter 7 Coenzymes and Vitamins

1. (a) Oxidation; $NAD^{\oplus}$, FAD, or FMN. (The coenzyme for the reaction shown is $NAD^{\oplus}$.)
(b) Decarboxylation of an α-keto acid; thiamine pyrophosphate.
(c) Carboxylation reaction requiring bicarbonate and ATP; biotin.
(d) Molecular rearrangement; adenosylcobalamin.
(e) Transfer of a hydroxyethyl group from TDP to CoA as an acyl group; lipoic acid.

2. (a) $NAD^{\oplus}$, $NADP^{\oplus}$, FAD, FMN, lipoamide, ubiquinone. Protein coenzymes such as thioredoxin and the cytochromes.
(b) Coenzyme A, lipoamide.
(c) Tetrahydrofolate, *S*-adenosylmethionine, methylcobalamin
(d) Pyridoxal phosphate
(e) Biotin, thiamine pyrophosphate, vitamin K

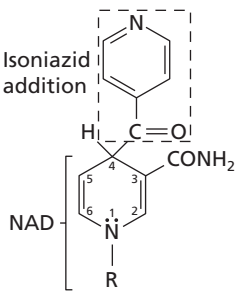
3. No. $NAD^{\oplus}$ acquires two electrons but only one proton. The second proton is released into solution and is reutilized by other proton-requiring reactions.

4.



5. $NAD^{\oplus}$, FAD, and coenzyme A all contain an ADP group (or ADP with 3′-phosphate for coenzyme A).

6.



7. Vitamin B_6 is converted to pyridoxal phosphate, which is the coenzyme for a large number of reactions involving amino acids, including the decarboxylation reactions in the pathways that produce serotonin and norepinephrine from tryptophan and tyrosine, respectively. Insufficient vitamin B_6 can lead to decreased levels of PLP and a decrease in the synthesis of the neurotransmitters.

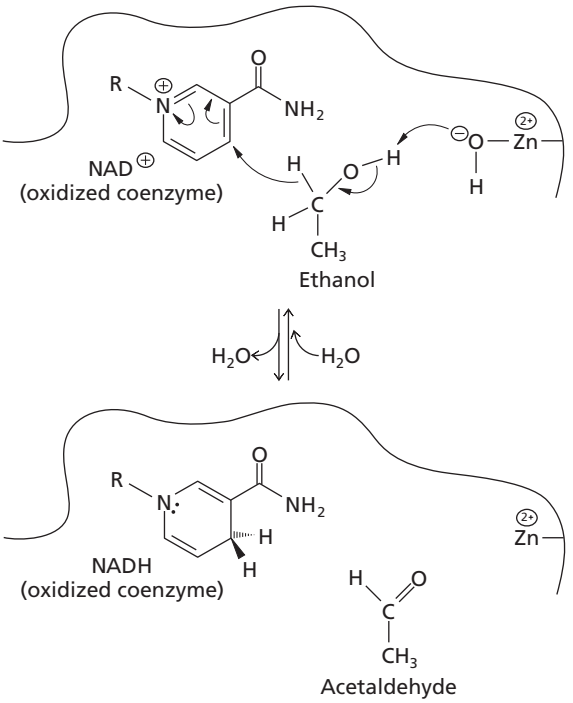
8. The synthesis of thymidylate (dTMP) requires a tetrahydrofolate (folic acid) derivative. Deficiency of folic acid decreases the amount of dTMP available for the synthesis of DNA. Decreased DNA synthesis in red blood cell precursors results in slower cell division, producing macrocytic red blood cells. The loss of cells by rupturing causes anemia.

9. (a) Cobalamin.
(b) The cobalamin derivative adenosylcobalamin is a coenzyme for the intramolecular rearrangement of methylmalonyl CoA to succinyl CoA (Figure 7.28). A deficiency of adenosylcobalamin results in increased levels of methylmalonyl CoA and its hydrolysis product, methylmalonic acid. Another cobalamin derivative, methylcobalamin, is a coen-

zyme for the synthesis of methionine from homocysteine (Reaction 7.5), and a deficiency of cobalamin results in an excess of homocysteine and a deficiency of methionine.

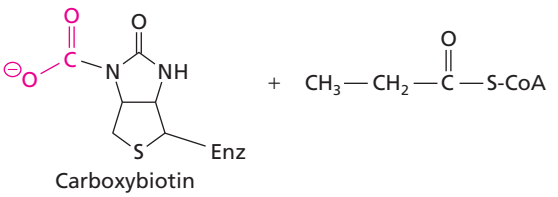
(c) Plants do not synthesize cobalamin and are therefore not a source of this vitamin.

10. (a) In one proposed mechanism, a water molecule bound to the zinc ion of alcohol dehydrogenase forms $OH^{\ominus}$, in the same manner as the water bound to carbonic anhydrase (Figure 7.2). The basic hydroxide ion abstracts the proton from the hydroxyl group of ethanol to form H_2O. (Another mechanism proposes that the zinc also binds to the alcoholic oxygen of the ethanol, polarizing it.)

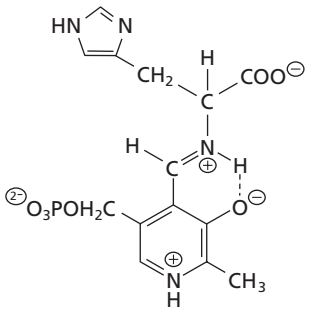


(b) No, a residue such as arginine is not required. Ethanol, unlike lactate, lacks a carboxylate group that can bind electrostatically to the arginine side chain.

11. A carboxyl group is transferred from methylmalonyl CoA to biotin to form carboxybiotin and propionyl CoA.



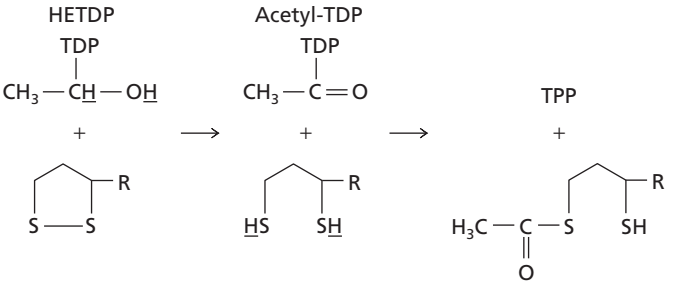
12. (a)



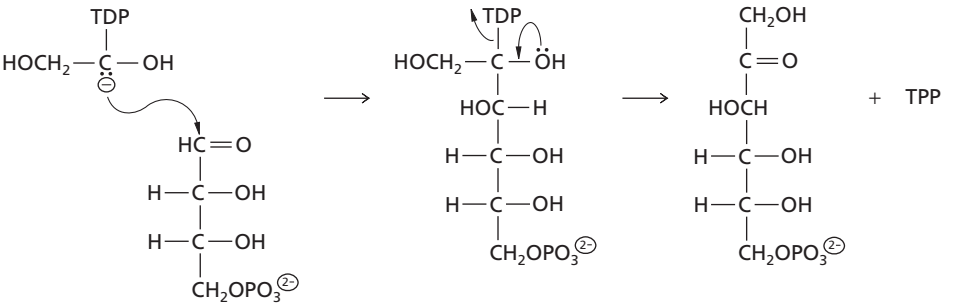
(b) Racemization would not occur. Although a Schiff base forms during decarboxylation as well as racemization, the reactive groups in the histidine decarboxylase active site specifically catalyze decarboxylation, not racemization, of histidine.

13. (a) See Reactions 13.2–13.4 on pages 412 and 413.

(b)



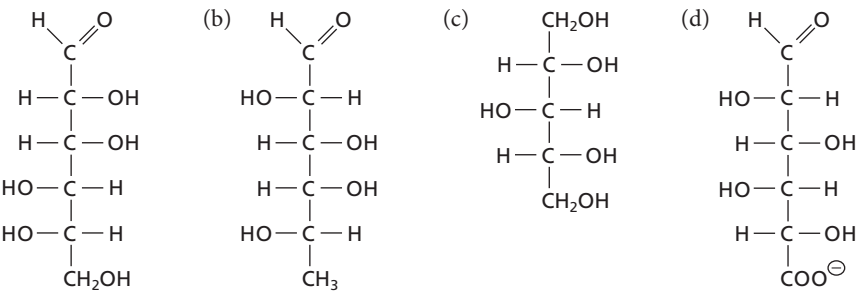
(c)



Chapter 8 Carbohydrates

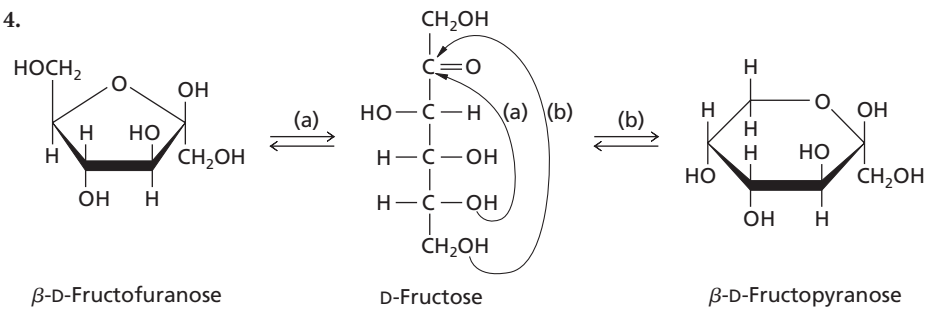
1. (a) D-Glucose and D-mannose
(b) L-Galactose
(c) D-Glucose or D-talose
(d) Dihydroxyacetone
(e) Erythrulose (either D or L)
(f) D-Glucose
(g) *N*-Acetylglucosamine

2. (a) (b) (c) (d)



3. Glycosaminoglycans are unbranched heteroglycans of repeating disaccharide units. One component of the disaccharide is an amino sugar and the other component is usually an al-duronic acid. Specific hydroxyl and amino groups of many glycosaminoglycans are sulfated

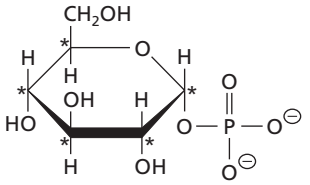
4.



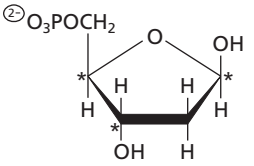
5. (a) α-Anomer
(b) Yes, it will mutorotate.
(c) Yes, it is a deoxy sugar.
(d) A ketone
(e) Four chiral carbons

6. Glucopyranose has five chiral carbons and 2^5, or 32, possible stereoisomers; 16 are D sugars and 16 are L sugars. Fructofuranose has four chiral carbons and 2^4, or 16, possible stereoisomers; 8 are D sugars and 8 are L sugars.

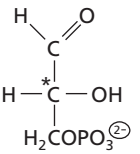
7. (a)



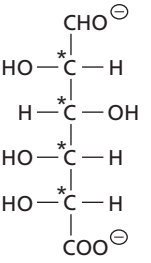
(b)



(c)



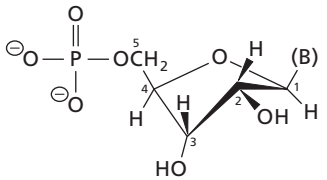
(d)



8. Only the open-chain forms of aldoses have free aldehyde groups that can form Schiff bases with amino groups of proteins. Because relatively few molecules of D-glucose are found in the open-chain form, D-glucose is less likely than other aldoses to react with proteins.

9. A pyranose is most stable when the bulkiest ring substituents are equatorial, minimizing steric repulsion. In the most stable conformer of β-D-glucopyranose, all the hydroxyl groups and the $-CH_2OH$ group are equatorial; in the most stable conformer of α-D-glucopyranose, the C-1 hydroxyl group is axial.

10.

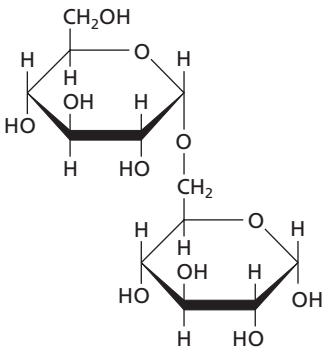


Envelope conformation

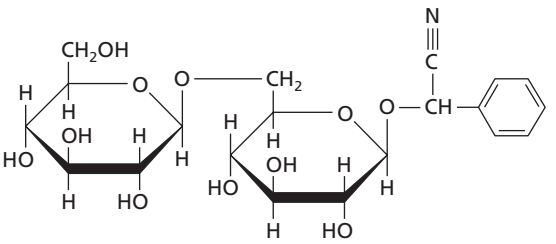
11. The α and β anomers of glucose are in rapid equilibrium. As β-D-glucose is depleted by the glucose oxidase reaction, more β anomer is formed from the α anomer until all the glucose has been converted to gluconolactone.

12. Sucralose is a derivative of the disaccharide sucrose (see Figure 8.20). The two hydroxyl groups on C-1 and C-6 of the fructose molecule have been replaced with chlorine. The hydroxyl group on C-4 of the glucose molecule was removed and then chlorine added. In the chemical synthesis of sucralose from sugar, the configuration of the C-4 substituent of the glucose moiety is reversed.

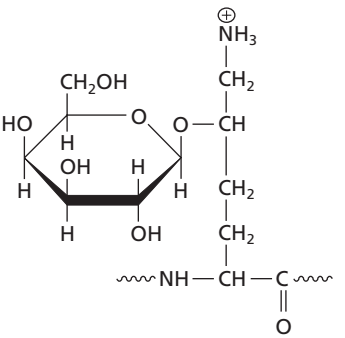
13. (a)



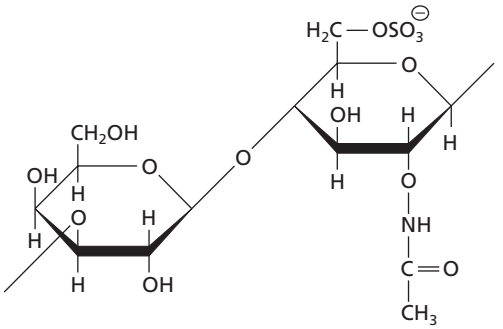
(b)



(c)

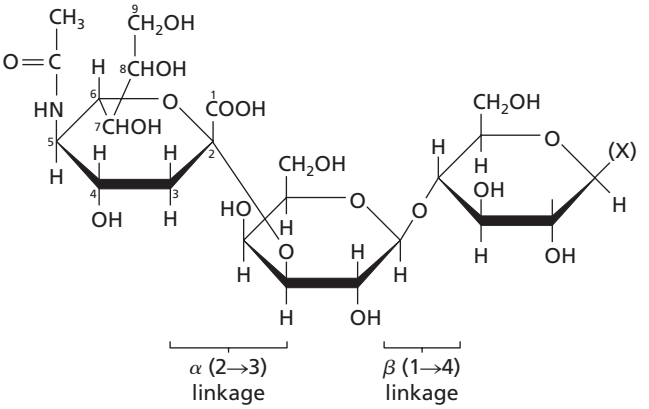


14.



15. (a) a, b, and c; these oligosaccharides contain GlcNAc — Asn bonds.
(b) b and c; these oligosaccharides contain β-galactosidic bonds.
(c) b; this oligosaccharide contains sialic acid.
(d) None, since none of the oligosaccharides shown contains fucose.

16.



17. Paper is made of cellulose and β-glucosidases break down cellulose to glucose residues. If you took a pill, this book would still taste like chewed up paper that tastes like paste (ugh!). That's because your taste buds are in your mouth and the enzyme is in your stomach. If you marinate the book in an enzyme solution, it would taste much sweeter.
Publishers would not print textbooks using flavored ink because they, and the authors, want students to keep their textbooks as valuable resources for future reference in the many advanced courses that you are planning to take. On the other hand, encouraging students to eat their textbooks, instead of selling them, might be a good thing because it promotes better health and nutrition.

Chapter 9 Lipids and Membranes

1.

(a) $CH_3(CH_2)_7-C(H)=C(H)-(CH_2)_{13}COO^{\ominus}$

(b) $CH_3(CH_2)_5-C(H)=C(H)-(CH_2)_9COO^{\ominus}$

(c) $CH_3CH_2(C(H)=C(H)CH_2)_5(CH_2)_2COO^{\ominus}$

2.

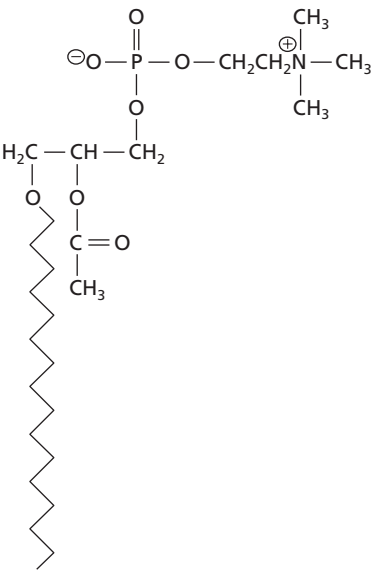
(a) $CH_3CH_2CH_2-O-(CH_2)_9COO^{\ominus}$

(b) $CH_3(CHCH_2CH_2CH_2)_3CHCH_2COO^{\ominus}$ (with CH_3 branches)

(c) $CH_3(CH_2)_5CH-CH(CH_2)_9COO^{\ominus}$ (cyclopropane ring)

3. (a) ω-3; (b) ω-6; (c) ω-6; (d) neither (ω-9); (e) ω-6.

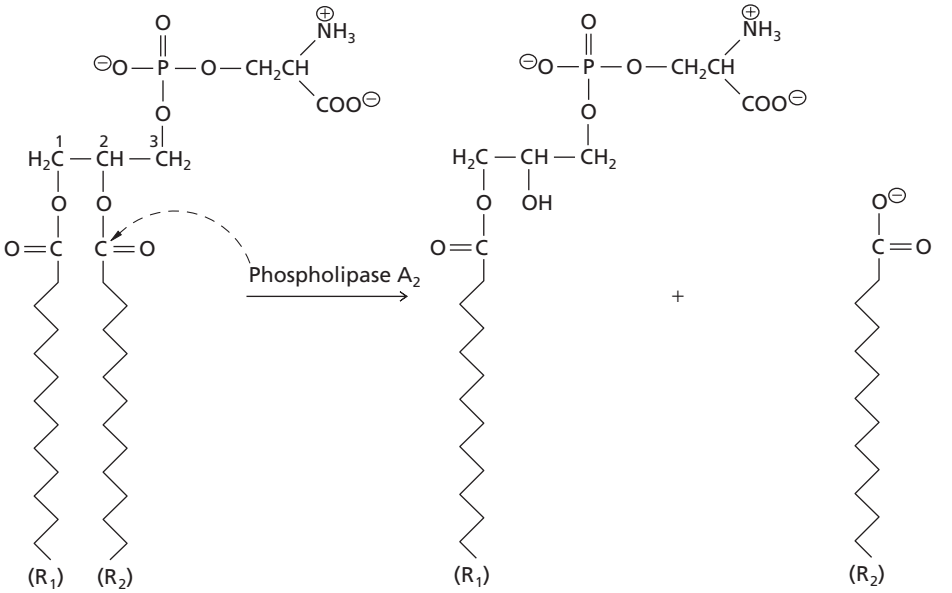
4.



5. (a)

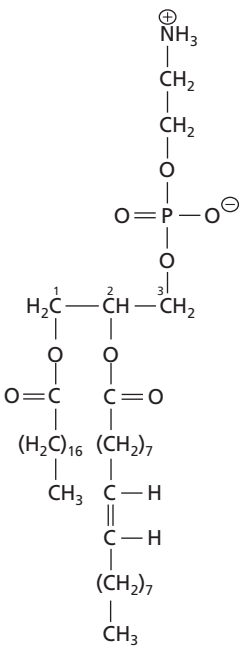
(b) Docosahexaenoic acid is classified as an ω-3 fatty acid

6.

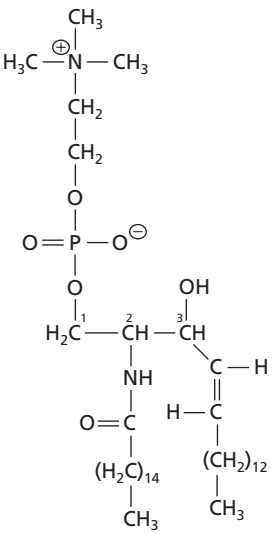


PS A lysolecithin Fatty acid

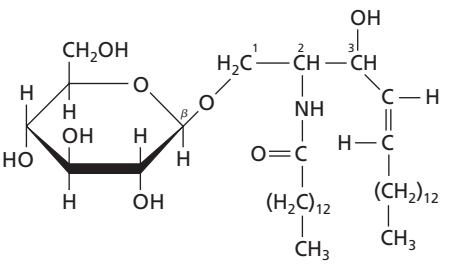
7. (a)



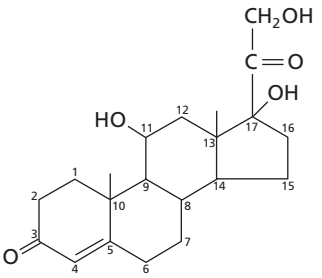
(b)



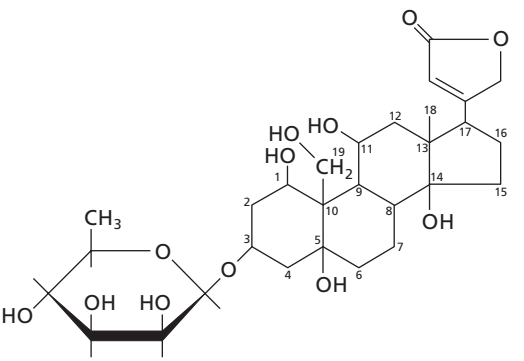
(c)



8. (a)



(b)

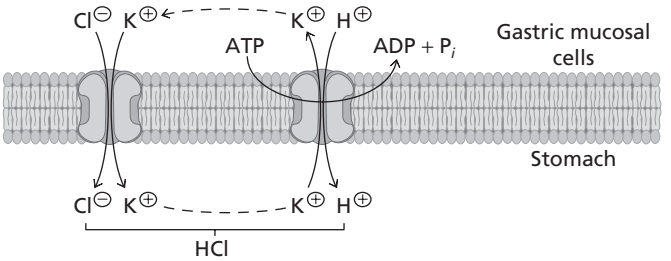


9. PE contains docosahexaenoic acid at position C-2 on the glycerol-3-phosphate backbone at both temperatures. At lower temperatures, the percent of the monounsaturated fatty acyl groups at position C-1 increased from 14% at 30°C to 39% at 10°C. The membrane fluidity must be maintained for the organism, and this is accomplished by changing the composition of the membrane lipids. The increase in the unsaturated lipids at the lower temperature will allow for the proper membrane fluidity.

10. Farnesyl transferase adds a farnesyl or "prenyl" group to a cysteine side chain of the *ras* protein (Figure 9.23b). The *ras* protein is subsequently anchored to the plasma and endoplasmic reticulum membranes and is active in cell signaling processes. Farnesyl transferase is a chemotherapy target because inhibition of this enzyme in tumor cells would disrupt the signaling activity of the mutated *ras* protein. In fact, farnesyl transferase (FT) inhibitors are potent suppressors of tumor growth in mice.

11. Line A represents diffusion of glucose through a channel or pore, and line B represents passive transport. Diffusion through a channel or pore is generally not saturable, with the rate increasing linearly with the concentration of the solute. Transport via a transport protein is saturable at high solute concentrations, much like an enzyme is saturated at high substrate concentrations (Section 9.10C).

12.



13. Theobromine is structurally related to caffeine and theophylline (Figure 9.45). The methylated purines, including the obromine, inhibit cAMP phophodiesterase, a soluble enzyme that catalyzes the hydrolysis of cAMP to AMP (Figure 9.43). These methylated purines inhibit the breakdown of the intracellular messenger cAMP to AMP. Therefore, the effects of the cAMP are prolonged. For dogs, this is combined with the fact that they have slower clearance of the ingested theobromine from their system. Both of these result in the toxicity associated with ingesting the chocolate.

14. The two second messengers IP_3 and DAG are complementary in that they both promote the activation of cellular kinases, which then activate intracellular target proteins by causing their phosphorylation. Diacylglycerol activates protein kinase C directly, whereas IP_3 elevates Ca^{2+} levels by opening a Ca^{2+} channel in the membrane of the endoplasmic reticulum, releasing stored Ca^{2+} into the cytosol (Figure 9.48). The increased Ca^{2+} levels activate other kinase leading to a phosphorylation and activation of certain target proteins.

15. Insulin can still bind normally to the α subunits of the insulin receptor, but due to the mutation, the β subunits lack tyrosine-kinase activity and cannot catalyze autophosphorylation or other phosphorylation reactions. Therefore, insulin does not elicit an intracellular response. The presence of more insulin will have no effect.

16. G proteins are molecular switches with two interconvertible forms, an active GTP-bound form and an inactive GDP-bound form (Figure 9.42). In normal G proteins, GTPase activity converts the active G protein to the inactive form. Because the *ras* protein lacks GTPase activity,

it cannot be inactivated. The result is continuous activation of adenylyl cyclase and prolonged responses to certain extracellular signals.

17. The surface of a sphere is $4\pi r^2$. The surface area of the oocyte is $4\pi(50)^2\mu m$, or $3.9 \times 10^5 \mu m^2$. The surface area of a lipid molecule is $10^{-14} cm^2 = 10^{-6}\mu m^2$. Since only 75% of the membrane is lipid, the total number of lipid molecules is

$$\frac{3.9 \times 10^5}{10^{-6}} \times 0.75 = 2.9 \times 10^{11} \text{ molecules}$$

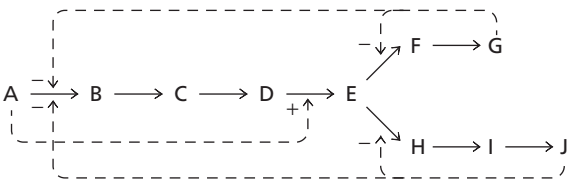
18. Assuming that the lipid molecules made by your grandmother are equally divided between daughter cells at each cell division, then after 30 cell divisions the oocyte (egg cell) produced by your mother will have $1/2^{30}$ of the original lipid molecules. Since the number of lipid molecules she inherited from her mother (your grandmother) was 2.9×10^{11} (see previous question), then the number remaining in each oocyte was

$$1/2^{30} \times 2.9 \times 10^{11} = 270$$

You inherited 270 lipid molecules from your grandmother.

Chapter 10 Introduction to Metabolism

1. (a)



(b) Inhibition of the first step in the common pathway by either G or J prevents the needless accumulation of intermediates in the pathway. When there is ample G or J, fewer molecules of A enter the pathway. By regulating an enzyme after the branch point, G or J inhibits its own production without inhibiting production of the other.

2. Compartmentalizing metabolic processes allows optimal concentrations of substrates and products for each pathway to exist independently in each compartment. In addition, separation of pathway enzymes also permits independent regulation of each pathway without interference by regulators from the other pathway.

3. Bacteria are much smaller than most eukaryotic cells so having separate compartments may not be as much of an advantage. It's also possible that localizing the citric acid cycle in mitochondria may be an historical accident rather than a selective advantage in eukaryotes.

4. In a multistep enzymatic pathway, the product from one enzyme will be the substrate for the next enzyme in the pathway. For independent soluble enzymes, the product of each enzyme must find the next enzyme by random diffusion in solution. By having sequential enzymes located in close proximity to each other, either in a multienzyme complex or on a membrane, the product of each enzyme can be passed directly on to the next enzyme without losing the substrate by diffusion into solution.

5. (a) $\Delta G^{\circ\prime} = RT \ln K_{eq}$

$$\ln K_{eq} = -\frac{\Delta G^{\circ\prime}}{RT} = -\frac{-9000 \text{ J mol}^{-1}}{(8.315 \text{ J K}^{-1} \text{ mol}^{-1})(298 \text{ K})} = 3.63$$

$$K_{eq} = 38$$

(b) $\Delta G^{\circ\prime} = -RT \ln K_{eq}$

$$K_{eq} = \frac{[\text{Glucose}][P_i]}{[\text{Glucose 6-P}][H_2O]} = \frac{(0.1 \text{ M})(0.1 \text{ M})}{(3.5 \times 10^{-5} \text{ M})(1)} = 286$$

$$\Delta G^{\circ\prime} = -(8.315 \text{ JK}^{-1} \text{ mol}^{-1})(298 \text{ K}) \ln 286$$

$$\Delta G^{\circ\prime} = -14\,000 \text{ J mol}^{-1} = -14 \text{ kJ mol}^{-1}$$

6. (a) $\Delta G = \Delta G^{\circ\prime} + RT \ln \frac{[\text{Arginine}][P_i]}{[\text{Phosphoarginine}][H_2O]}$

$$\Delta G = -32\,000 \text{ J mol}^{-1} + (8.315 \text{ J K}^{-1} \text{ mol}^{-1})(298 \text{ K}) \ln \frac{(2.6 \times 10^{-3})(5 \times 10^{-3})}{(6.8 \times 10^{-3})(1)}$$

$$\Delta G = -48 \text{ kJ mol}^{-1}$$

(b) $\Delta G^{\circ\prime}$ is defined under standard conditions of 1 M concentrations of reactants and products. (The concentration of water is assigned a value of 1.) ΔG depends on the actual concentrations of the reactants and products.

(c) Molecules with high free energies of hydrolysis, such as phosphoarginine and acetyl CoA, are thermodynamically unstable but may be kinetically stable. These molecules are hydrolyzed very slowly in the absence of an appropriate catalyst.

7.

	$\Delta G^{\circ\prime}$(kJ mol^{-1})
Glucose 1-phosphate + UTP $\longrightarrow$ UDP-glucose + PP_i	0
$PP_i + H_2O \longrightarrow 2\ P_i$	−29
	$\Delta G^{\circ\prime}$ −29

8. (a) Although ATP is rapidly utilized for energy purposes such as muscle contraction and membrane transport, it is also rapidly resynthesized from ADP and P_i through intermediary metabolic routes. Energy for this process is supplied from the degradation of carbohydrates, fats, and amino acids or from energy storage molecules such as muscle creatine phosphate (CP + ADP → ATP + C). With this rapid recycling, 50 grams total of ATP and ADP is sufficient for the chemical energy needs of the body.

(b) The role of ATP is that of a free energy transmitter rather than an energy storage molecule. As indicated in part (a), ATP is not stored, but is rapidly utilized in energy-requiring reactions.

9. $\Delta G^{\circ\prime}$ for the reaction of ATP and creatine is calculated as

	$\Delta G^{\circ\prime}$(kJ mol^{-1})
Creatine + $P_i \longleftrightarrow$ Phosphocreatine + H_2O	+43
ATP + $H_2O \longleftrightarrow$ ADP + P_i	−32
Creatine + ATP $\longleftrightarrow$ Phosphocreatine + ADP	+11

The ratio of ATP to ADP needed to maintain a 20:1 ratio of phosphocreatine to creatine is calculated from Equation 10.13. At equilibrium, $\Delta G = 0$, so

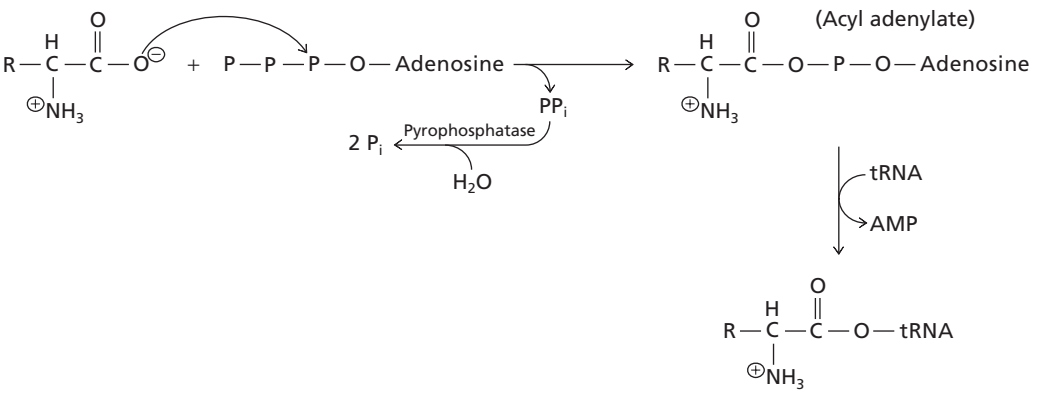
$$\Delta G^{\circ\prime} = -RT \ln \frac{[\text{Phosphocreatine}][\text{ADP}]}{[\text{Creatine}][\text{ATP}]}$$

$$\ln \frac{(20)[\text{ADP}]}{(1)[\text{ATP}]} = -\frac{\Delta G^{\circ\prime}}{RT} = -\frac{(11\ 000\ \text{J mol}^{-1})}{(8.315\ \text{J K}^{-1}\ \text{mol}^{-1})(298\ \text{K})} = -4.44$$

$$\frac{(20)[\text{ADP}]}{(1)[\text{ATP}]} = 1.2 \times 10^{-2}$$

$$\frac{[\text{ATP}]}{[\text{ADP}]} = 1667{:}1$$

10.



11. $\Delta G^{\circ\prime} = -RT \ln K_{eq}$

$$K_{eq} = \frac{[\text{fructose-6-phosphate}]}{[\text{glucose-6-phosphate}]} = \frac{2}{1}$$

$$\Delta G^{\circ\prime} = -(8.315\ \text{J K}^{-1}\ \text{mol}^{-1})(298\ \text{K}) \ln 2$$

$$\Delta G^{\circ\prime} = -1.7\ \text{kJ mol}^{-1}$$

12. (a) $\ln K_{eq} = -\dfrac{\Delta G^{o\prime}}{RT} = -\dfrac{(25\,000\ \text{J mol}^{-1})}{(8.315\ \text{J K}^{-1}\ \text{mol}^{-1})(298\ \text{K})} = -10.1$

$K_{eq} = 4.1 \times 10^{-5}$

(b) $\Delta G^{o\prime}$ for the coupled reaction is calculated as

	$\Delta G^{o\prime}$(kJ mol^{-1})
$A \longrightarrow B$	$+25$
$ATP + H_2O \longleftrightarrow ADP + P_i$	-32
$A + ATP + H_2O \longleftrightarrow B + ADP + P_i$	-7

$\ln K_{eq} = -\dfrac{\Delta G^{o\prime}}{RT} = 2.8$

$K_{eq} = 17$

K_{eq} for the coupled reaction is about 180,000 times larger than K_{eq} in part (a).

(c) $K_{eq} = 17 = \dfrac{[B][ADP][P_i]}{[A][ATP][H_2O]} = \dfrac{[B][ADP]}{[A][ATP]} = \dfrac{[B](1)}{[A](400)}$

$\dfrac{[B]}{[A]} = 6800:1$

Coupling the reaction to ATP hydrolysis increases the ratio of [B] to [A] by a factor of about 166 million ($6800 \div (4.1 \times 10^{-5}) = 1.6 \times 10^{8}$).

13. Electrons flow from the molecule with a more negative standard reduction potential to the molecule with a more positive standard reduction potential.

(a) Cytochrome $b_5(Fe^{2+})$ + Cytochrome $f(Fe^{3+}) \rightarrow$
Cytochrome $b_5(Fe^{3+})$ + Cytochrome $f(Fe^{2+})$

(b) Succinate + Q → Fumarate + QH_2

(c) Isocitrate + $NAD^{\oplus}$ → α-Ketoglutarate + NADH

14. The standard reduction potentials in Table 10.4 refer to half-reactions that are written as $S_{ox} + n\,e^{\ominus} \rightarrow S_{red}$. Two half-reactions can be added to obtain the coupled oxidation–reduction reaction by reversing the direction of the half-reaction involving the reduced species and reversing the sign of its reduction potential.

(a) 2 Cyt $c(Fe^{3+}) + 2e^{\ominus} \longrightarrow$ 2 Cyt $c(Fe^{2+})$ $\quad E^{o\prime}$(V) $+0.23$

$QH_2 \longrightarrow Q + 2\,H^{\oplus} + 2e^{\ominus}$ $\quad -0.04$

2 Cyt $c(Fe^{3+}) + QH_2 \longrightarrow$ 2 Cyt $c(Fe^{2+}) + Q + 2\,H^{\oplus}$ $\quad \Delta E^{o\prime} = 0.19$ V

$\Delta G^{o\prime} = -nF\Delta E^{o\prime} = -(2)(96.48\ \text{kJ V}^{-1}\ \text{mol}^{-1})(0.19\ \text{V})$

$\Delta G^{o\prime} = -37\ \text{kJ mol}^{-1}$

(b) $\frac{1}{2}\,O_2 + 2\,H^{\oplus} + 2\,e^{\ominus} \longrightarrow H_2O$ $\quad E^{o}$(V) $+0.82$

Succinate → Fumarate $2\,H^{\oplus} + 2\,e^{\ominus}$ $\quad -0.03$

$\frac{1}{2}\,O_2$ + Succinate → H_2O + Fumarate $\quad \Delta E^{o\prime} = 0.79$ V

$\Delta G^{o\prime} = -(2)(96.48\ \text{kJ V}^{-1}\ \text{mol}^{-1})(0.79\ \text{V})$

$\Delta G^{o\prime} = -150\ \text{kJ mol}^{-1}$

15. The expected results are as shown in the bottom graph. As NADH is formed in the reaction mixture, the absorbance at 340 nm will increase (see Box 10.1).

16. $Q + 2\,H^{\oplus} + 2\,e^{\ominus} \longrightarrow QH_2$ $\quad E^{o\prime}$(V) $+0.04$

$FADH_2 \longrightarrow FAD + 2\,H^{\oplus} + 2\,e^{\oplus}$ $\quad +0.22$

$Q + FADH_2 \longrightarrow QH_2 + FAD$ $\quad \Delta E^{o\prime} = 0.26$ V

$$\Delta E = \Delta E^{o\prime} - \frac{RT}{nF}\ln\frac{[QH_2][FAD]}{[Q][FADH_2]}$$

$$\Delta E = 0.26\ \text{V} - \frac{0.026\ \text{V}}{2}\ln\frac{(5 \times 10^{-5})(2 \times 10^{-4})}{(1 \times 10^{-4})(5 \times 10^{-3})}$$

$$\Delta E = 0.26\ \text{V} - 0.013(-3.9) = 0.31\ \text{V}$$

$$\Delta G = -nF\Delta E = -(2)(96.48\ \text{kJ V}^{-1}\ \text{mol}^{-1})(0.31\ \text{V})$$

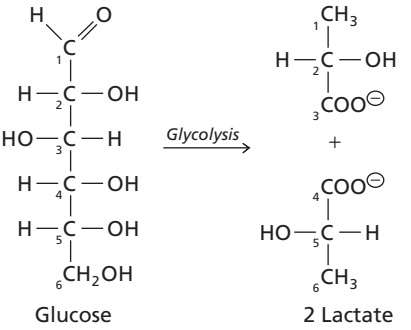
$$\Delta G = -60\ \text{kJ mol}^{-1}$$

Theoretically, the oxidation of $FADH_2$ by ubiquinone liberates more than enough free energy to drive ATP synthesis from ADP and P_i.

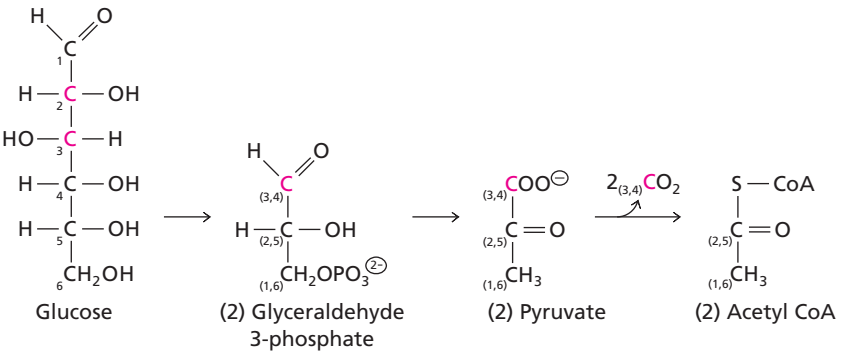
Chapter 11 Glycolysis

1. (a) 2 (see Figure 11.2 and Reaction 11.12)
(b) 2 (1 ATP is consumed by the fructokinase reaction, 1 ATP is consumed by the triose kinase reaction, and 4 ATP are generated by the triose stage of glycolysis)
(c) 2 (2 ATP are consumed in the hexose stage, and 4 ATP are generated by the triose stage)
(d) 5 (2 ATP are obtained from fructose, as in part (b), and 3 ATP—rather than 2—are obtained from the glucose moiety since glucose 1-phosphate, not glucose, is formed when sucrose is cleaved)

2. (a)



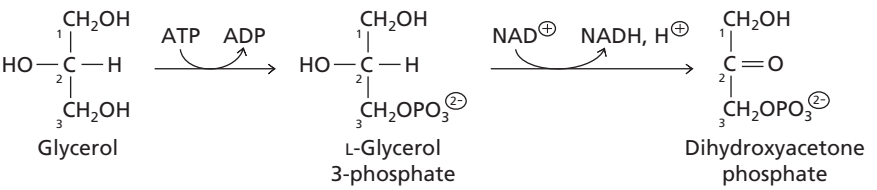
(b) Glucose labeled at either C-3 or C-4 yields $^{14}CO_2$ from the decarboxylation of pyruvate.



3. Inorganic phosphate ($^{32}P_i$) will be incorporated into 1,3-*bis*phosphoglycerate (1,3 BPG) at the C-1 carbon in the glyceraldehyde 3-phosphate dehydrogenase (GADPH) reaction—glyceraldehyde 3-phosphate + $NAD^{\oplus}$ + $P_i \rightarrow$ 1,3 BPG—and then transferred to the γ-position of ATP in the next step: 1,3 BPG + ADP $\rightarrow$ ATP + 3-phosphoglycerate.

4. Since the brain relies almost solely on glucose for energy, it is dependent on glycolysis as the major pathway for glucose catabolism. Since the Huntington protein binds tightly to GAPDH, this suggests that it might inhibit this crucial glycolytic enzyme and thereby impair the production of ATP. Decreased ATP levels would be detrimental to neuronal cells in the brain.

5. (a)



(b) C-2 and C-3 of glycerol 3-phosphate must be labeled. Once dihydroxyacetone phosphate is converted to glyceraldehyde 3-phosphate, C-1 is oxidized to an aldehyde and subsequently lost as CO_2 (Problem 2).

6. Cells that metabolize glucose to lactate by anaerobic glycolysis produce far less ATP per glucose than do cells that metabolize glucose aerobically to CO_2 via glycolysis and the citric acid cycle (Figure 11.1). More glucose must be utilized via anaerobic glycolysis to produce a sufficient amount of ATP for cellular needs, and the rate of conversion of glucose to lactate is

much higher than under aerobic conditions. Cancer cells in an anaerobic environment take up far more glucose and may overproduce some glycolytic enzymes to compensate for the increase in the activity of this pathway of carbohydrate metabolism.

7. No. The conversion of pyruvate to lactate, catalyzed by lactate dehydrogenase, oxidizes NADH to $NAD^{\oplus}$, which is required for the glyceraldehyde 3-phosphate dehydrogenase reaction of glycolysis.

8. In the reactions catalyzed by these enzymes, the bond between the γ-phosphorus atom and the oxygen of the β-phosphoryl group is cleaved when the γ-phosphoryl group of ATP is transferred (Figure 11.3). The analog cannot be cleaved in this way and therefore inhibits the enzymes by competing with ATP for the active site.

9. The free energy change for the aldolase reaction under standard conditions ($\Delta G^{\circ\prime}$) is $+22.8$ kJ mol^{-1}. The concentrations of fructose 1,6-*bis*phosphate, dihydroxyacetone phosphate, and glyceraldehyde 3-phosphate in heart muscle, however, are much different than the 1 M concentrations assumed under standard conditions. The actual free energy change under cellular concentrations ($\Delta G^{\circ\prime} = -5.9$ kJ mol^{-1}) is much different than $\Delta G^{\circ\prime}$, and the aldolase reaction readily proceeds in the direction necessary for glycolysis: Fructose 1, 6-*bis*phosphate ⟶ glyceraldehyde 3-phosphate + dihydroxyacetone phosphate.

10. The standard Gibbs free energy change is + 28 kJ mol^{-1}. The equilibrium constant is

$$28 = RT \ln K_{eq} \approx 10^{-5} \quad \text{(Equation 1.12)}$$

(a) $$\frac{[\text{DHAP}][\text{G3P}]}{[\text{FBP}]} = 10^{-5} \quad \frac{[5 \times 10^{-6}][5 \times 10^{-6}]}{[\text{FBP}]} = 10^{-5} \quad \text{FBP} = 2.5\ \mu\text{M}$$

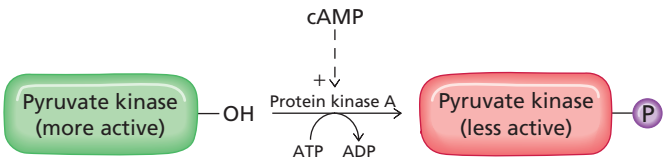
(b) 250μM

(c) 25,000 μM = 25 *m*M

11. (a) ATP is both a substrate and an allosteric inhibitor for PFK-1. Higher concentrations of ATP result in a decrease in the activity of PFK-1 due to an increase in the K_m. AMP is an allosteric activator that acts by relieving the inhibition caused by ATP, thus raising the curve when AMP is present with ATP.

(b) F2,6P is an allosteric activator of PFK-1. In the presence of F2,6P the activity of PFK-1 is increased due to a decrease in the apparent K_m for fructose 6-phosphate.

12. Increased [cAMP] activates protein kinase A, which catalyzes the phosphorylation and inactivation of pyruvate kinase.



13. (a) A decrease in glycolysis in the liver makes more glucose available for export to other tissues.

(b) Decreased activity of the glucagon transducer system decreases the amount of cAMP formed. As existing cAMP is hydrolyzed by the activity of a phosphodiesterase, cAMP-dependent protein kinase A becomes less active. Under these conditions, PFK-2 activity increases and fructose 2,6-*bis*phosphatase activity decreases (Figure 11.18). The resulting increase in fructose 2,6-*bis*phosphate activates PFK-1, increasing the overall rate of glycolysis. A decrease in cAMP also leads to the activation of pyruvate kinase (Problem 12).

14. Chemoautotrophs use glycolysis to generate energy from stored glucose residues in glycogen as described in Chapter 12.

Chapter 12 Gluconeogenesis, The Pentose Phosphate Pathway, and Glycogen Metabolism

1. 2 pyruvate + 2NADH + 4 ATP + 2 GTP + 6 H_2O + 2 $H^{\oplus}$ ⟶
   glucose + 2 $NAD^{\oplus}$ + 4 ADP + 2 GDP + 6 P_i

   2 NADH ≡ 5 ATP equivalents

   4 ATP = 4 ATP

   $$2\text{GTP} \equiv \frac{2\ \text{ATP}}{11\ \text{ATP}}$$

The energy required to synthesize one molecule of glucose 6-phosphate from CO_2 can be calculated from Reaction 12.7.

$$12\ \text{NADPH} \equiv 30\ \text{ATP}$$

The conversion of G6P to glucose does not require or produce ATP equivalents. The synthesis of glucose from pyruvate via the gluconeogenesis pathway is only about one third (11/30) as expensive as the synthesis of glucose from CO_2.

2. Reducing power in the form of NADH (2), and ATP (4) and GTP (2) are required for the synthesis of glucose from pyruvate (Equation 12.1). The NADH and GTP are direct products of the citric acid cycle, and ATP can be generated from NADH and QH_2($FADH_2$) during the oxidative phosphorylation process.

3. Epinephrine interacts with the liver β-adrenergic receptors and activates the adenylyl cyclase signaling pathway, leading to cAMP production and activation of protein kinase A (Figure 12.15). Protein kinase A activates phosphorylase kinase, which in turn activates glycogen phosphorylase (GP), leading to glycogen degradation (Figure 12.16). Glucose can then be transported out of the liver and into the bloodstream, where it is taken up by muscles for needed energy production.

$$\text{Liver[Glycogen} \xrightarrow{\text{(GP)}} \text{GIP} \longrightarrow \text{G6P} \longrightarrow \text{Glucose]} \longrightarrow \text{Bloodstream} \longrightarrow \text{Muscles}$$

4. (a) Protein phosphatase-1 activated by insulin catalyzes the hydrolysis of the phosphate ester bonds on glycogen synthase (activating it) and on glycogen phosphorylase and phosphorylase kinase (inactivating them), as shown in Figure 12.17. Therefore, insulin stimulates glycogen synthesis and inhibits glycogen degradation in muscle cells.

(b) Only liver cells are rich in glucagon receptors, so glucagon selectively exerts its effects on liver enzymes.

(c) The binding of glucose to the glycogen phosphorylase–protein phosphatase-1 complex in liver cells relieves the inhibition of protein phosphatase-1 and makes glycogen phosphorylase more susceptible to dephosphorylation (inactivation) by protein phosphatase-1 (Figure 12.18). Protein phosphatase-1 also catalyzes the dephosphorylation of glycogen synthase, making it more active. Therefore, glucose stimulates glycogen synthesis and inhibits glycogen degradation in the liver.

5. Decreased concentrations of fructose 2,6-*bis*phosphate (F2,6BP) lead to a decreased rate of glycolysis and an increased rate of gluconeogenesis. F2,6BP is an activator of the glycolytic enzyme phosphofructokinase-1 (PFK-1), and lower F2,6BP levels will result in decreased rates of glycolysis. In addition, F2,6BP is an inhibitor of the gluconeogenic enzyme fructose 1,6-*bis*phosphatase, and therefore decreased levels of F2,6BP will decrease the inhibition and increase the rate of gluconeogenesis (Figure 12.4).

6. When glucagon binds to its receptor, it activates adenylyl cyclase. Adenylyl cyclase catalyzes the synthesis of cAMP from ATP. The cAMP activates protein kinase A. Protein kinase A catalyzes the phosphorylation of PFK-2, which inactivates the kinase activity and activates the phosphatase activity. Fructose 2,6-*bis*phosphatase catalyzes the hydrolytic dephosphorylation of fructose 2,6-*bis*phosphate to form fructose 6-phosphate. The resulting decrease in the concentration of fructose 2,6-*bis*phosphate relieves the inhibition of fructose 1,6-*bis*phosphatase, thereby activating gluconeogenesis. Thus, the kinase activity of PFK-2 is decreased.

7. (a) Yes. The synthesis of glycogen from glucose 6-phosphate requires the energy of one phosphoanhydride bond (in the hydrolysis of PP_i; Figure 12.10). However, when glycogen is degraded to glucose 6-phosphate, inorganic phosphate (P_i) is used in the phosphorolysis reaction. No "high energy" phosphate bond is used.

(b) One fewer ATP molecule is available for use in the muscle when liver glycogen is the source of the glucose utilized. Liver glycogen is degraded to glucose phosphates and then to glucose without consuming ATP. After transport to muscle cells, the glucose is converted to glucose 6-phosphate by the action of hexokinase in a reaction that consumes one molecule of ATP. Muscle glycogen, however, is converted directly to glucose 1-phosphate by the action of glycogen phosphorylase, which does not consume ATP. Glucose 1-phosphate is isomerized to glucose 6-phosphate by the action of phosphoglucomutase.

8. A deficiency of glycogen phosphorylase in the muscle prevents the mobilization of glycogen to glucose. Insufficient glucose prevents the production of ATP by glycolysis. Existing ATP used for muscle contraction is not replenished, thus increasing the levels of ADP and P_i. Since no glucose is available from glycogen in the muscle, no lactate is produced.

9. Converting glucose 1-phosphate to two molecules of lactate yields 3 ATP equivalents (1 ATP expended in the phosphofructokinase-1 reaction, 2 ATP produced in the phosphoglycerate

kinase reaction, and 2 ATP produced in the pyruvate kinase reaction). Converting two molecules of lactate to one molecule of glucose 1-phosphate requires 6 ATP equivalents (2 ATP in the pyruvate carboxylase reaction, 2 GTP in the PEP carboxykinase reaction, and 2 ATP in the phosphoglycerate kinase reaction).

10. (a) Muscle pyruvate from glycolysis or amino acid catabolism is converted to alanine by transamination. Alanine travels to the liver, where it is reconverted to pyruvate by transamination with α-ketoglutarate. Gluconeogenesis converts pyruvate to glucose, which can be returned to muscles.

(b) NADH is required to reduce pyruvate to lactate in the Cori cycle, but it is not required to convert pyruvate to alanine in the glucose-alanine cycle. Thus, the glucose-alanine cycle makes more NADH available in muscles for the production of ATP by oxidative phosphorylation.

11. (a) Inadequate glucose 6-phosphatase activity (G6P $\rightarrow$ glucose + P_i) leads to accumulation of intracellular G6P, which inhibits glycogen phosphorylase and activates glycogen synthase. This prevents liver glycogen from being mobilized. This results in increased glycogen storage (and enlargement of the liver) and low blood glucose levels (hypoglycemia).

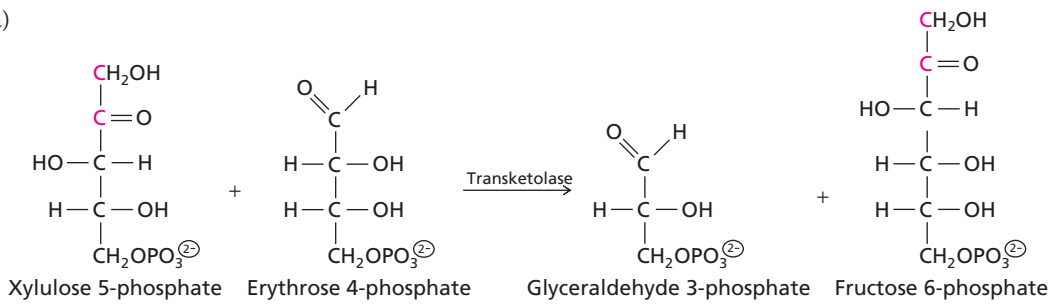
(b) Yes. A defective branching enzyme leads to accumulation of glycogen molecules with defective, short outer branches. These molecules cannot be degraded, so there will be much less efficient glycogen degradation for glucose formation. Low blood glucose levels result due to the impaired glycogen degradation.

(c) Inadequate liver phosphorylase activity leads to an accumulation of liver glycogen since the enzyme cleaves a glucose molecule from the nonreducing end of a glycogen chain. Low blood glucose levels result, due to the impaired degradation of glycogen.

12. Glucose 6-phosphate, glyceraldehyde 3-phosphate, and fructose 6-phosphate.

13. The repair of tissue injury requires cell proliferation and synthesis of scar tissue. NADPH is needed for the synthesis of cholesterol and fatty acids (components of cellular membranes), and ribose 5-phosphate is needed for the synthesis of DNA and RNA. Since the pentose phosphate pathway is the primary source of NADPH and ribose 5-phosphate, injured tissue responds to the increased demands for these products by increasing the level of synthesis of the enzymes in the pentose phosphate pathway.

14. (a)



(b) C-2 of glucose 6-phosphate becomes C-1 of xylulose 5-phosphate. After C-1 and C-2 of xylulose 5-phosphate are transferred to erythrose 4-phosphate, the label appears at C-1 of fructose 6-phosphate, as shown in part (a).

Chapter 13 The Citric Acid Cycle

1. (a) No net synthesis is possible since two carbons from acetyl CoA enter the cycle in the citrate synthase reaction and two carbons leave as CO_2 in the isocitrate dehydrogenase and α-ketoglutarate dehydrogenase reactions.

(b) Oxaloacetate can be replenished by the pyruvate carboxylase reaction, which carries out a net synthesis of OAA,

$$\text{Pyruvate} + CO_2 + \text{ATP} + H_2O \longrightarrow \text{Oxaloacetate} + \text{ADP} + P_i$$

This is the major anaplerotic reaction in some mammalian tissues. Many plants and some bacteria supply oxaloacetate via the phosphoenolpyruvate carboxykinase reaction,

$$\text{Phosphoenolpyruvate} + HCO_3^{\ominus} \longrightarrow \text{Oxaloacetate} + P_i$$

In most species, acetyl CoA can be converted to malate and oxaloacetate via the glyoxylate pathway.

2. Aconitase would be inhibited by fluorocitrate formed from fluoroacetate, leading to increased levels of citric acid and decreased levels of all subsequent citric acid cycle intermediates from

isocitrate to oxaloacetate. Since fluorocitrate is a competitive inhibitor, very high levels of citrate would at least partially overcome the inhibition of aconitase by fluorocitrate and permit the cycle to continue at some level.

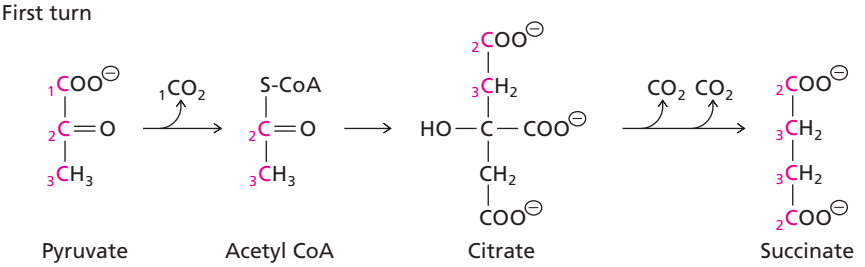
3. (a) 12.5; 10.0 from the cycle and 2.5 from the pyruvate dehydrogenase reaction.
(b) 10.0; 7.5 from oxidation of 3 NADH, 1.5 from oxidation of 1 QH_2, and 1.0 from the substrate-level phosphorylation catalyzed by CoA synthetase.

4. 87.5% (28 of 32) of the ATP is produced by oxidative phosphorylation, and 12.5% (4 of 32) is produced by substrate-level phosphorylation.

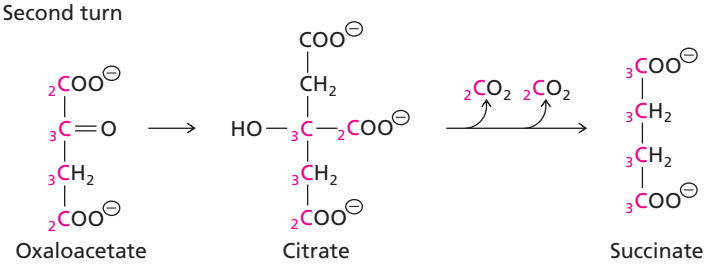
5. Thiamine is the precursor of the coenzyme thiamine pyrophosphate (TPP), which is found in two enzyme complexes associated with the citric acid cycle: the pyruvate dehydrogenase complex and the α-ketoglutarate dehydrogenase complex. A deficiency of TPP decreases the activities of these enzyme complexes. Decreasing the conversion of pyruvate to acetyl CoA and of α-ketoglutarate to succinyl CoA causes accumulation of pyruvate and α-ketoglutarate.

6. Since C-1 of pyruvate is converted to CO_2 in the reaction catalyzed by the pyruvate dehydrogenase complex, 1-[^{14}C]-pyruvate is the first to yield $^{14}CO_2$. Neither of the two acetyl carbon atoms of acetyl CoA is converted to CO_2 during the first turn of the citric acid cycle (Figure 13.5). However, the carboxylate carbon atoms of oxaloacetate, which arise from C-2 of pyruvate, become the two carboxylates of citrate that are removed as CO_2 during a second turn of the cycle. Therefore, 2-[^{14}C]-pyruvate is the second labeled molecule to yield $^{14}CO_2$. 3-[^{14}C]-Pyruvate is the last to yield $^{14}CO_2$, in the third turn of the cycle.

First turn



Second turn



Half of the ^{14}C is eliminated by the third turn of the cycle. An additional one-fourth is eliminated in the fourth turn, then one-eighth in the fifth turn, etc. It will take a very long time to eliminate all of the ^{14}C from the citric acid cycle intermediates.

7. (a) The NADH produced by the oxidative reactions of the citric acid cycle must be recycled back to $NAD^{\oplus}$, which is required for the pyruvate dehydrogenase reaction. When O_2 levels are low, fewer NADH molecules are reoxidized by O_2 (via the process of oxidative phosphorylation), so the activity of the pyruvate dehydrogenase complex decreases.
(b) Pyruvate dehydrogenase kinase catalyzes phosphorylation of the pyruvate dehydrogenase complex, thereby inactivating it (Figure 13.12). Inhibiting the kinase shifts the pyruvate dehydrogenase complex to its more active form.

8. A deficiency in the citric acid cycle enzyme fumarase would result in abnormally high concentrations of fumarate and prior cycle intermediates including succinate and α-ketoglutarate, which could lead to excretion of these molecules.

9. The different actions of acetyl CoA on two components of the pyruvate dehydrogenase (PDH) complex both lead to an inhibition of the pyruvate to acetyl CoA reaction. Acetyl CoA inhibits the E_2 component of the PDH complex directly (Figure 13.11). Acetyl CoA causes inhibition of the E_1 component indirectly by activating the pyruvate kinase (PK) component of

the PDH complex, and PK phosphorylates the E_1 component of the PDH complex, thus inactivating it (Figure 13.12).

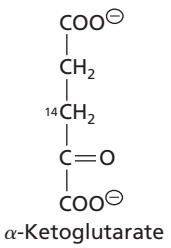
10. The pyruvate dehydrogenase complex catalyzes the oxidation of pyruvate to form acetyl CoA and CO_2. If there is reduced activity of this complex, then the pyruvate concentration will increase. Pyruvate will be converted to lactate through the action of lactate dehydrogenase. Lactate builds up since glycolytic metabolism is increased to synthesize ATP since oxidation of pyruvate to acetyl CoA is impaired. In addition, pyruvate is converted to alanine, as shown in Reaction 12.6.

11. Calcium activates both isocitrate dehydrogenase and α-ketoglutarate dehydrogenase in the citric acid cycle, thereby increasing this catabolic process and producing more ATP. In addition, Ca^{2+} activates the pyruvate dehydrogenase phosphatase enzyme of the PDH complex, which activates the E_1 component (Figure 13.12). Activation of the PDH complex converts more pyruvate into acetyl CoA for entry into the citric acid cycle, resulting in an increased production of ATP.

12. (a) Alanine degradation replenishes citric acid cycle intermediates, since pyruvate can be converted to oxaloacetate via the pyruvate carboxylase reaction, the major anaplerotic reaction in mammals (Reaction 13.19). Leucine degradation cannot replenish intermediates of the citric acid cycle, since for every molecule of acetyl CoA that enters the cycle, two molecules of CO_2 are lost.

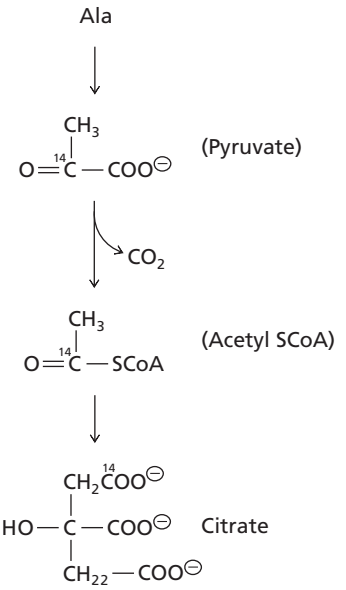
(b) By activating pyruvate carboxylase, acetyl CoA increases the amount of oxaloacetate produced directly from pyruvate. The oxaloacetate can react with the acetyl CoA produced by the degradation of fatty acids. As a result, flux through the citric acid cycle increases to recover the energy stored in the fatty acids.

13. (a)

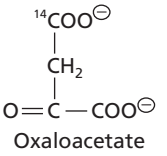


α-Ketoglutarate

(b)



(c)



Oxaloacetate

14. (a) Two molecules of acetyl CoA yield 20 ATP molecules via the citric acid cycle (Figure 13.10) or 6.5 ATP molecules via the glyoxylate cycle (from the oxidation of two molecules of NADH and one molecule of QH_2; Reaction 13.22).

(b) The primary function of the citric acid cycle is to oxidize acetyl CoA to provide the reduced coenzymes necessary for the generation of energy-rich molecules such as ATP. The primary function of the glyoxylate cycle is not to produce ATP, but to convert acetyl groups to four-carbon molecules that can be used to produce glucose.

15. The protein that controls the activity of isocitrate dehydrogenase in *E. coli* is a bifunctional enzyme with kinase and phosphatase activities in the same protein molecule. The kinase activity phosphorylates isocitrate dehydrogenase to inhibit the activity of isocitrate dehydrogenase, and the phosphatase activity dephosphorylates isocitrate dehydrogenase to activate isocitrate dehydrogenase. When concentrations of glycolytic and citric acid cycle intermediates are high, isocitrate dehydrogenase is not phosphorylated and is active. When phosphorylation decreases the activity of isocitrate dehydrogenase, isocitrate is diverted to the glyoxylate cycle.

Chapter 14 Electron Transport and Oxidative Phosphorylation

1. The formula for calculating protonmotive force is

$$\Delta G = F\,\Delta\psi - 2.303\,RT\,\Delta\ \text{pH}$$

If $G = -21{,}000$ kJ and $\Delta\psi = -0.15$ V, then at 25°C

$$-21{,}200 = (96485 \times -0.15) - 2.303(8.315 \times 298)\,\Delta\text{pH}$$

$$5707\ \Delta\ \text{pH} = 6727$$

$$\Delta\ \text{pH} = 1.2$$

Since the outside pH is 6.35 and the inside is negative (higher pH), then the cytoplasmic pH is $6.35 + 1.2 = 7.55$.

2. The reduction potential of an iron atom in a heme group depends on the surrounding protein environment, which differs for each cytochrome. The differences in reduction potentials allow electrons to pass through a series of cytochromes.

3. Refer to Figure 14.6.
(a) Complex III. The absence of cytochrome *c* prevents further electron flow.
(b) No reaction occurs since Complex I, which accepts electrons from NADH, is missing.
(c) O_2
(d) Cytochrome *c*. The absence of Complex IV prevents further electron flow.

4. UCP-2 leaks protons back into the mitochondria, thereby decreasing the protonmotive force. The metabolism of foodstuffs provides the energy for electron transport, which in turn creates the protonmotive gradient used to produce ATP. An increase in UCP-2 levels would make the tissue less metabolically efficient (i.e., less ATP would be produced per gram of foodstuff metabolized). As a result, more carbohydrates, fats, and proteins would have to be metabolized in order to satisfy the basic metabolic needs, and this could "burn off" more calories and potentially cause weight loss.

5. (a) Demerol interacts with Complex I and prevents electron transfer from NADH to Q. The concentration of NADH increases since it cannot be reoxidized to $NAD^{\oplus}$. The concentration of Q increases since electrons from QH_2 are transferred to O_2 but Q is not reduced back to QH_2.

(b) Myxothiazole inhibits electron transfer from QH_2 to cytochrome c_1 and from QH_2 (via $\cdot Q^-$) to cytochrome b_{566} in Complex III (Figure 14.14). The oxidized forms of both cytochromes predominate since Fe^{3+} cannot be reduced by electrons from QH_2.

6. (a) Oxygen (O_2) must bind to the Fe^{3+} of cytochrome a_3 in order to accept electrons (Figure 14.19), and it is prevented from doing so by the binding of $CN^{\ominus}$ to the iron atom.
(b) The methemoglobin (Fe^{3+}) generated from nitrite treatment competes with cytochrome a_3 for the $CN^{\ominus}$ ions. This competition effectively lowers the concentration of cyanide available to inhibit cytochrome a_3 in Complex IV, and decreases the inhibition of the electron transport chains in the presence of $CN^{\ominus}$.

7. A substrate is usually oxidized by a compound with a more positive reduction potential. Since $E^{\circ\prime}$ for the fatty acid is close to $E^{\circ\prime}$ for FAD in Complex II (0.0 V, as shown in Table 14.1), electron transfer from the fatty acid to FAD is energetically favorable.

$$\Delta E^{\circ\prime} = 0.0\ \text{V} - (-0.05\ \text{V}) = +0.05\ \text{V}$$

$$\Delta G^{\circ\prime} = -nF\Delta E^{\circ\prime}$$

$$\Delta G^{\circ\prime} = -(2)(96.48\ \text{kJ V}^{-1})(0.05\ \text{V}) = -9.6\ \text{kJ mol}^{-1}$$

Since $E^{\circ\prime}$ for NADH in Complex I is -0.32 V, the transfer of electrons from the fatty acid to NADH is unfavorable.

$$\Delta E^{\circ\prime} = -0.32\ \text{V} - (-0.05\ \text{V}) = -0.27\ \text{V}$$

$$\Delta G^{\circ\prime} = -(2)(96.48\ \text{kJ V}^{-1}\ \text{mol}^{-1})(-0.27\ \text{V}) = 52\ \text{kJ mol}^{-1}$$

8. (a) 10 protons; 2.5 ATP; P : O = 2.5.
(b) 6 protons; 1.5 ATP; P : O = 1.5.
(c) 2 protons; 0.5 ATP; P : O = 0.5.

9. (a) The inner mitochondrial membrane has a net positive charge on the cytosolic side (outside). The exchange of one ATP^{4-} transferred out for one ADP^{3-} transferred in yields a

net movement of one negative charge from the inner matrix side to the positive cytosolic side. The membrane potential thereby assures that outward transport of a negatively charged ATP is favored by the outside positive charge.

(b) Yes. The electrochemical potential with a net positive charge outside the membrane is a result of proton pumping, which is driven by the electron transport chain. This in turn requires oxidation of metabolites to generate NADH and QH_2 as electron donors.

10. ATP synthesis is normally associated with electron transport. Unless ADP can continue to be translocated into the mitochondrial matrix for the ATP synthesis reaction (ADP + $P_i \rightarrow$ ATP), ATP synthesis will not occur and the proton gradient will not be dissipated. Electron transport will be inhibited as the proton concentration increases in the intermembrane space.

11. (a) $\Delta G = F\Delta\Psi - 2.303\ RT\ \Delta$ pH (Equation 14.6)

$\Delta G = ((96485)(-0.18)) - ((2.303)(8.315)(0.7))$

$\Delta G = -17367 - 3995$

$\Delta G = -2136 = 21$ kJ mol^{-1}

(b) $\Delta G_{total} = 21.36$ kJ mol^{-1}

Charge gradient contribution is 17.367 kJ mol^{-1}, or $17.367 \div 21.36 \times 100 = 81.3\%$

pH gradient contribution is 3.995 kJ mol^{-1}, or $3.995 \div 21.36 \times 10 = 18.7\%$

12. (a) In the malate-aspartate shuttle, the reduction of oxaloacetate in the cytosol consumes a proton that is released in the matrix by the oxidation of malate (Figure 14.27). Therefore, one fewer proton is contributed to the proton concentration gradient for every cytosolic NADH oxidized (9 versus 10 for mitochondrial NADH). The ATP yield from two molecules of cytoplasmic NADH is about 4.5 rather than 5.0.

(b) *Cytoplasmic reactions*

Glucose $\longrightarrow$ 2 Pyruvate 2.0 ATP

2 NADH $\longrightarrow$ 4.5 ATP

Mitochondrial reactions

2 Pyruvate $\longrightarrow$ 2 Acetyl CoA + 2 CO_2 2 NADH $\longrightarrow$ 5.0 ATP

2 Acetyl CoA $\longrightarrow$ 4 CO_2 2.0 GTP

6 NADH $\longrightarrow$ 15.0 ATP

2 QH_2 $\longrightarrow$ 3.0 ATP

Total 31.5 ATP

Chapter 15 Photosynthesis

1. Because in photosynthesis there are *two* steps where light energy is absorbed to produce "high energy" electrons, thus PS II transfers 6 $H^{\oplus}$ insead of 10 $H^{\oplus}$ in respiration but PSI produces 2.5 ATP equivalents—the same as respiration.

2. Plant chlorophylls absorb energy in the red region of the spectrum (Figure 15.2). The dragonfish chlorophyll derivatives absorb the red light energy (667 nm), and pass the signals on to the visual pigments in much the same manner that plant antenna chlorophylls and related molecules capture light energy and transfer it to a reaction center where electrons are promoted into excited states for transfer to acceptors of the electron transport chain.

3. (a) Rubisco is the world's most abundant protein and the principal catalyst for photosynthesis, the basic means by which living organisms acquire the carbon necessary for life. Its importance in the process of providing food for all living things can be well justified.

(b) Photorespiration is a process that wastes ribulose 1,5-*bis*phosphate, consumes the NADPH and ATP generated by the light reactions, and can greatly reduce crop yields. As much as 20% to 30% of the carbon fixed in photosynthesis can be lost to photorespiration. This process results from the lack of specificity of Rubisco, which can use O_2 instead of CO_2 (Figure 15.8) to produce phosphoglycolate and 3-phosphoglycerate (Figure 15.18) instead of two triose phosphate molecules. In addition, Rubisco has low catalytic activity ($K_{cat} \approx 3\ s^{-1}$). This lack of specificity and low activity earns Rubisco the title of a relatively incompetent, inefficient enzyme.

4. $6\ CO_2 + 6\ H_2S \rightarrow C_6H_{12}O_6 + 3\ O_2 + 6\ S$

$6\ CO_2 + 12\ H^{\oplus} \rightarrow C_6H_{12}O_6 + 3\ O_2$

5. (a) $CO_2 + 2\,H_2S \xrightarrow{\text{Light}} (CH_2O) + H_2O + 2\,S$

$CO_2 + 2\,CH_3CH_2OH \xrightarrow{\text{Light}} (CH_2O) + H_2O + 2\,CH_3CHO$

Ethanol Acetaldehyde

(b) When H_2O is the proton donor, O_2 is the product, but when other proton donors such as H_2S and ethanol are used, oxygen cannot be produced. Most photosynthetic bacteria do not produce O_2 and are obligate anaerobes that are poisoned by O_2.

(c) $CO_2 + 2\,H_2A \xrightarrow{\text{Light}} (CH_2O) + H_2O + 2A$

6. Rubisco is not active in the dark because it requires alkaline conditions. Those conditions only occur when photosynthesis is active so there's nothing (except light) that can be added to the chloroplast suspension in the dark that will activate the calvin cycle.

7. (a) Two H_2O molecules provide the oxygens for one O_2 during the photosynthetic process. A total of four electrons must be removed from two H_2O and passed through an electron transport system to two NADPH. One quantum of light is required to transfer one electron through PSI and one quantum for PSII. Therefore, a total of eight photons will be required to move four electrons through both reaction centers (four photons for PHI and four photons for PHII).

(b) Six NADPH are required for the synthesis of one triose phosphate by the Calvin cycle (Figure 15.21). Therefore, 12 electrons must be transferred through the two reaction centers of the electron transport system and this will require the absorption of 24 hν.

8. (a) Yes. (Refer to the Z-scheme, Figure 15.14). When DCMU blocks electron flow, PSII in the P680* state will not be reoxidized to the $P680^{\oplus}$ state, which is required as an acceptor of electrons from H_2O. If H_2O is not oxidized by $P680^{\oplus}$, then no O_2 will be produced. In the absence of electron flow through the cytochrome *bf* complex, no protons will be translocated across the membrane. Without a pH gradient no photophosphorylation (ATP synthesis) will be possible.

(b) External electron acceptors for PSII will permit P680 to be reoxidized to $P680^{\oplus}$ and will restore O_2 evolution. No electrons will flow through the cytochrome *bf* complex, however, so no photophosphorylation will occur.

9. (a) When the external pH rises to 8.0, the stromal pH also rises quickly, but the luminal pH remains low initially because the thylakoid membrane is relatively impermeable to protons. The pH gradient across the thylakoid membrane drives the production of ATP via proton translocation through chloroplast ATP synthase (Figure 15.16).

(b) Protons are transferred from the lumen to the stroma by ATP synthase, driving ATP synthesis. The pH gradient across the membrane decreases until it is insufficient to drive the phosphorylation of ADP, and ATP synthesis stops.

10. During cyclic electron transport, reduced ferredoxin donates its electrons back to P700 via the cytochrome *bf* complex (Figure 15.11). As these electrons cycle again through photosystem I, the proton concentration gradient generated by the cytochrome *bf* complex drives ATP synthesis. However, no NADPH is produced because there is no *net* flow of electrons from H_2O to ferredoxin. No O_2 is produced because photosystem II, the site of O_2 production, is not involved in cyclic electron transport.

11. The light absorbing complexes, electron transport chain, and chloroplast ATP synthase all reside in the thylakoid membranes, and the structure and interactions of any of these photosynthetic components could be affected by a change in the physical nature of the membrane lipids.

12. The compound is acting as an uncoupler. The electron transfer is occurring without the synthesis of ATP. The compound destroys the proton gradient that is produced through electron transfer.

13. (a) The synthesis of one triose phosphate from CO_2 requires 9 molecules of ATP and 6 molecules of NADPH (Equation 15.5). Since two molecules of triose phosphate can be converted to glucose, glucose synthesis requires 18 molecules of ATP and 12 molecules of NADPH.

(b) Incorporating glucose 1-phosphate into starch requires one ATP equivalent during the conversion of glucose 1-phosphate to ADP-glucose (Figure 15.24), bringing the total requirement to 19 molecules of ATP and 12 molecules of NADPH.

14. Refer to Figure 15.21. (a) C-1. (b) C-3 and C-4. (c) C-1 and C-2. C-1 and C-2 of fructose 6-phosphate are transferred to glyceraldehyde 3-phosphate to form xylulose 5-phosphate. C-3 and C-4 of fructose 6-phosphate become C-1 and C-2 of erythrose 4-phosphate.

15. (a) In the C_4 pathway (Figure 15.29), the pyruvate-phosphate dikinase reaction consumes two ATP equivalents for each CO_2 fixed (since PP_i is hydrolyzed to 2 P_i). Therefore, C_4

plants require 12 more molecules of ATP per molecule of glucose synthesized than C_3 plants require.

(b) Because C_4 plants minimize photorespiration, they are more efficient than C_3 plants in using light energy to fix CO_2 into carbohydrates, even though the chemical reactions for fixing CO_2 in C_4 plants require more ATP.

16. (a) An increase in stromal pH increases the rate of the Calvin cycle in two ways.

(1) An increase in stromal pH increases the activity of ribulose 1,5-*bis*phosphate carboxylase-oxygenase (Rubisco), the central regulatory enzyme of the Calvin cycle, and the activities of fructose 1,6-*bis*phosphatase and sedoheptulose 1,7-*bis*phosphatase. It also increases the activity of phosphoribulokinase. Phosphoribulokinase is inhibited by 3-phosphoglycerate (3PG) in the $3PG^{(2-)}$ ionization state but not in the $3PG^{(3-)}$ ionization state, which predominates at higher pH.

(2) An increase in stromal pH also increases the proton gradient that drives the synthesis of ATP in chloroplasts. Since the reactions of the Calvin cycle are driven by ATP, an increase in ATP production increases the rate of the Calvin cycle.

(b) A decrease in the stromal concentration of $Mg^{(2+)}$ decreases the rate of the Calvin cycle by decreasing the activity of Rubisco, fructose 1,6-*bis*phosphatase, and sedoheptulose 1,7-*bis*phosphatase.

Chapter 16 Lipid Metabolism

1. (a) LDLs are rich in cholesterol and cholesterol esters and transport these lipids to peripheral tissues. Delivery of cholesterol to tissues is moderated by LDL receptors on the cell membranes. When LDL receptors are defective, receptor-mediated uptake of cholesterol does not occur (Section 16.10B). Because cholesterol is not cleared from the blood it accumulates and contributes to the formation of atherosclerotic plaques.

(b) Increased cholesterol levels normally repress transcription of HMG-CoA reductase and stimulate the proteolysis of this enzyme as well. With defective LDL, however, cholesterol synthesis continues in spite of high blood cholesterol levels because the extracellular cholesterol cannot enter the cells to regulate intracellular synthesis.

(c) HDLs remove cholesterol from plasma and cells of nonhepatic tissues and transport it to the liver where it can be converted into bile salts for disposal. In Tangier patients, defective cholesterol-poor HDLs cannot absorb cholesterol, and the normal transport process to the liver is disrupted.

2. (a) Carnitine is required to transport fatty acyl CoA into the mitochondrial matrix for β-oxidation (Figure 16.24). The inhibition of fatty acid transport caused by a deficiency in carnitine diminishes energy production from fats for muscular work. Excess fatty acyl CoA can be converted to triacylglycerols in the muscle cells.

(b) Since carnitine is not required to transport pyruvate, a product of glycolysis, into mitochondria for oxidation, muscle glycogen metabolism is not affected in individuals with a carnitine deficiency.

3. (a) Activation of the C_{12} fatty acid to a fatty acyl CoA consumes 2 ATP. Five rounds of β-oxidation generate 6 acetyl CoA, 5 QH_2 (which yield 7.5 ATP via oxidative phosphorylation), and 5 NADH (which yield 12.5 ATP). Oxidation of the 6 acetyl CoA by the citric acid cycle yields 60 ATP. Therefore, the net yield is 78 ATP equivalents.

(b) Activation of the C_{16} monounsaturated fatty acid to a fatty acyl CoA consumes 2 ATP. Seven rounds of β-oxidation generate 8 acetyl CoA, 6 QH_2 (which yield 9 ATP via oxidative phosphorylation), and 7 NADH (which yield 17.5 ATP). The fatty acid contains a *cis*-β,γ double bond that is converted to a *trans*-α,β double bond, so the acyl-CoA dehydrogenase-catalyzed reaction, which generates QH_2, is bypassed in the fifth round. Oxidation of the 8 acetyl CoA by the citric acid cycle yields 80 ATP. Therefore, the net yield is 104.5 ATP equivalents.

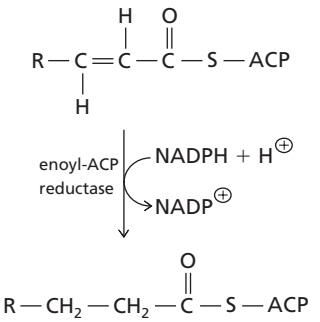
4. When triacylglycerols are ingested in our diets, the hydrolysis of the dietary lipids occurs mainly in the small intestine. Pancreatic lipase catalyzes the hydrolysis at the C-1 and C-3 positions of triacylglycerol, producing free fatty acids and 2-monoacylglycerol. These molecules are transported in bile-salt micelles to the intestine, where they are absorbed by intestinal cells. Within these cells, the fatty acids are converted to fatty acyl CoA molecules, which eventually form a triacylglycerol that is incorporated into chylomicrons for transport to other tissues. If the pancreatic lipase is inhibited, the ingested dietary triglyceride cannot be absorbed. The triglyceride will move through the digestive tract and will be excreted without absorption.

5. (a) Oleate has a *cis*-Δ^9 double bond, so oxidation requires enoyl-CoA isomerase (as in Step 2 of Figure 16.26).
(b) Arachidonate has *cis* double bonds at both odd (Δ^5, Δ^{11}) and even (Δ^8, Δ^{14}) carbons, so oxidation requires both enoyl-CoA isomerase and 2,4-dienoyl-CoA reductase (as in Step 5 of Figure 16.26).
(c) This C_{17} fatty acid contains a *cis* double bond at an even-numbered carbon (Δ^6), so oxidation requires 2,4-dienoyl-CoA reductase. In addition, three enzymes are required to convert the propionyl CoA product into succinyl CoA: propionyl-CoA carboxylase, methylmalonyl-CoA racemase, and methylmalonyl-CoA mutase (Figure 16.25).

6. Even-chain fatty acids are degraded to acetyl CoA, which is not a gluconeogenic precursor. Acetyl CoA cannot be converted directly to pyruvate because for every two carbons of acetyl CoA that enter the citric acid cycle, two carbons in the form of two CO_2 molecules leave as products. The last three carbons of odd-chain fatty acids, on the other hand, yield a molecule of propionyl CoA upon degradation in the fatty acid oxidation cycle. Propionyl CoA can be carboxylated and converted to succinyl CoA in three steps (Figure 16.25). Succinyl CoA can be converted to oxaloacetate by citric acid cycle enzymes, and oxaloacetate can be a gluconeogenic precursor for glucose synthesis.

7. (a) The labeled carbon remains in $H^{14}CO_3^{\ominus}$; none is incorporated into palmitate. Although $H^{14}CO_3^{\ominus}$ is incorporated into malonyl CoA (Figure 16.2), the same carbon is lost as CO_2 during the ketoacyl-ACP synthase reaction in *each* turn of the cycle (Figure 16.5).
(b) All the even-numbered carbons are labeled. Except for the acetyl CoA that becomes C-15 and C-16 of palmitate, the acetyl CoA is converted to malonyl CoA and then to malonyl-ACP before being incorporated into a growing fatty acid chain with the loss of CO_2.

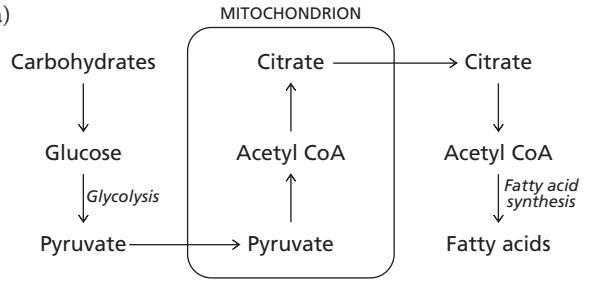
8. (a) Enoyl ACP reductase catalyzes the second reductive step in the fatty acid biosynthesis pathway, converting a *trans*-2,3 enoyl moiety into a saturated acyl chain, and uses NADPH as cofactor.



(b) Fatty acids are essential for membranes in bacteria. If fatty acid synthesis is inhibited, there will be no new membranes and no growth of the bacteria.
(c) The fatty acid synthesis systems are different in animals and bacteria. Animals contain a type I fatty acid synthesis system (FAS I) where the various enzymatic activities are localized to individual domains in a large, multifunctional enzyme. In bacteria, each reaction in fatty acid synthesis is catalyzed by a separate monofunctional enzyme. Understanding some of the differences in these two systems, would allow for the design of specific inhibitors of the bacterial FAS II.

9. Eating stimulates the production of acetyl CoA from the metabolism of carbohydrates (glycolysis and pyruvate dehydrogenase) and fats (FA oxidation). Normally, increased acetyl CoA results in the elevation of malonyl CoA levels (acetyl CoA carboxylase reaction, Figure 16.2), which may act to inhibit appetite. By blocking fatty acid synthase enzyme, C75 prevents the removal of malonyl CoA for the synthesis of fatty acids, thereby elevating the levels of malonyl-CoA and further suppressing appetite.

10. (a)



(b) The NADH generated by glycolysis can be transformed into NADPH by a variety of different reactions and pathways.

11. (a) Plentiful citrate and ATP levels promote fatty acid synthesis. High citrate levels activate ACC by preferential binding and stabilization of the active dephosphorylated filamentous form. On the other hand, high levels of fatty acyl CoAs indicate that there is no further need for more fatty acid synthesis. Palmitoyl CoA inactivates ACC by preferential binding to the inactive protomeric dephosphorylated form.

(b) Glucagon and epinephrine inhibit fatty acid synthesis by inhibiting the activity of acetyl CoA carboxylase. Both hormones bind to cell receptors and activate cAMP synthesis, which in turn activates protein kinases. Phosphorylation of ACC by protein kinases converts it to the inactive form, thus inhibiting fatty acid synthesis. On the other hand, the active protein kinases catalyze phosphorylation and activation of triacylglycerol lipases that catalyze hydrolysis of triacylglycerols, releasing fatty acids for β-oxidation.

12. (a) An inhibitor of acetyl-CoA acetylase will affect a key regulatory reaction for fatty acid synthesis. The concentration of malonyl CoA, the product of the acetyl-CoA carboxylase-catalyzed reaction, will be decreased in the presence of the inhibitor. The decrease in the concentration of malonyl CoA will relieve the inhibition of carnitine acyltransferase I, which is a key regulatory site for the oxidation of fatty acids. Thus, with an active carrier system, fatty acids will be translocated to the mitochondrial matrix where the reactions of β-oxidation occur. In the presence of an inhibitor of acetyl-CoA carboxylase, fatty acid synthesis will decrease and β-oxidation will increase.

(b) CABI is a structural analog of biotin. Acetyl-CoA carboxylase is a biotin-dependent enzyme. A biotin analog may bind in place of biotin and inhibit the activity of acetyl-CoA carboxylase.

13. The overall reaction for the synthesis of palmitate from acetyl CoA is the sum of two processes: (1) the formation of seven malonyl CoA by the action of acetyl-CoA carboxylase and (2) seven cycles of the fatty acid biosynthetic pathway.

$$7\ \text{Acetyl CoA} + 7\ CO_2 + 7\ \text{ATP} \longrightarrow 7\ \text{Malonyl CoA} + 7\ \text{ADP} + 7\ P_i$$

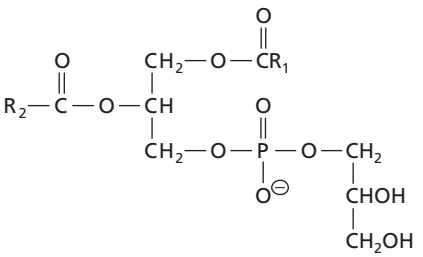
$$\text{Acetyl CoA} + 7\ \text{Malonyl CoA} + 14\ \text{NADPH} + 14\ H^{\oplus} \longrightarrow \text{Palmitate} + 7\ CO_2 + 14\ \text{NADP}^{\oplus} + 8\ \text{HS} - \text{CoA} + 6\ H_2O$$

$$8\ \text{Acetyl CoA} + 7\ \text{ATP} + 14\ \text{NADPH} + 14\ H^{\oplus} \longrightarrow \text{Palmitate} + 7\ \text{ADP} + 7\ P_i + 14\ \text{NADP}^{\oplus} + 8\ \text{HS} - \text{CoA} + 6\ H_2O$$

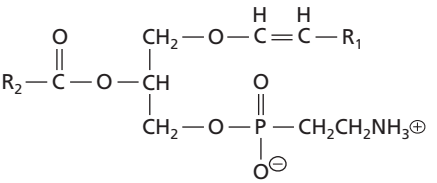
14. (a) Arachidonic acid is a precursor for synthesis of eicosanoids including "local regulators" such as prostaglandins, thromboxanes, and leukotrienes (Figure 16.14). These regulators are involved in mediation of pain, inflammation, and swelling responses resulting from injured tissues.

(b) Both prostaglandins and leukotrienes are derived from arachidonate, which is released from membrane phospholipids by the action of phospholipases. By inhibiting a phospholipase, steroidal drugs block the biosynthesis of *both* prostaglandins and leukotrienes. Aspirin-like drugs block the conversion of arachidonate to prostaglandin precursors by inhibiting cyclooxygenase but do not affect leukotriene synthesis.

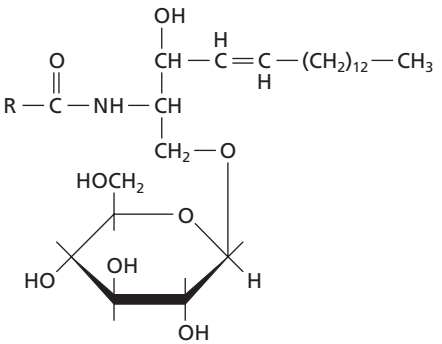
15. (a)



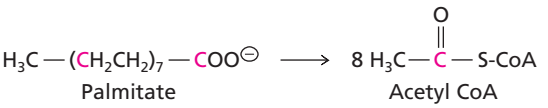
(b)

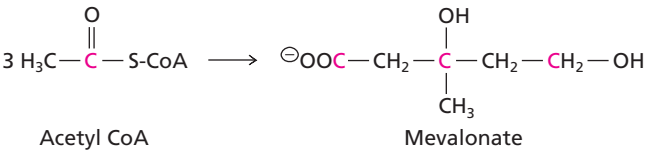


(c)

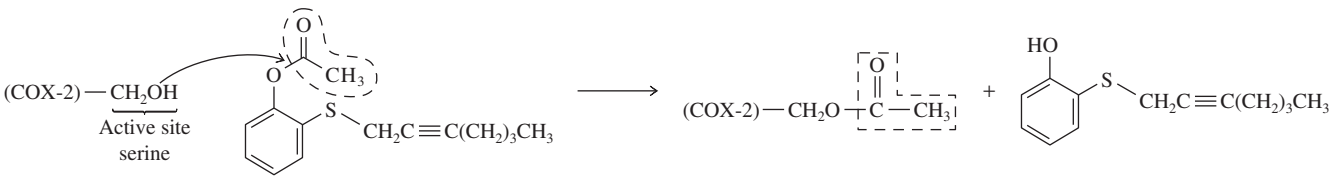


16. Palmitate is converted to eight molecules of acetyl CoA labeled at C-1. Three acetyl CoA molecules are used to synthesize one molecule of mevalonate (Figure 16.17).





17. Both APHS and aspirin transfer an acetyl group to a serine residue on COX enzymes. Since APHS is an irreversible inhibitor, it does not exhibit competitive inhibition kinetics even though it acts at the active site of COX enzymes.



Chapter 17 Amino Acid Metabolism

1. PSII contains the oxygen evolving complex and oxygen is produced during photosynthesis. Since oxygen inhibits nitrogenase, the synthesis of O_2 in hetocysts must be avoided. PSI is retained because it can still generate a light-induced proton gradient by cyclic electron transport and it is not involved in the production of O_2.

2. (a) Glutamate dehydrogenase + glutamine synthetase

$$\begin{aligned} NH_4^{\oplus} + \alpha\text{-Ketoglutarate} + NAD(P)H + H^{\oplus} &\longrightarrow \text{Glutamate} + NAD(P)^{\oplus} + H_2O \\ NH_3 + \text{Glutamate} + ATP &\longrightarrow \text{Glutamine} + ADP + P_i \\ \hline 2\ NH_4^{\oplus} + \alpha\text{-Ketoglutarate} + NAD(P)H + ATP &\longrightarrow \text{Glutamine} + NAD(P)^{\oplus} + ADP + P_i + H_2O \end{aligned}$$

(b) Glutamine synthetase + glutamate synthase

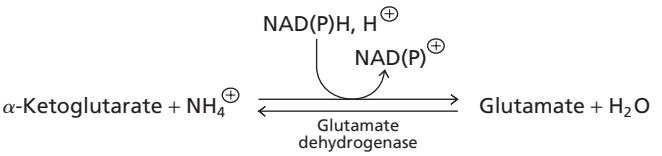
$$\begin{aligned} 2\ NH_3 + 2\ \text{Glutamate} + 2\ ATP &\longrightarrow 2\ \text{Glutamine} + 2\ ADP + 2\ P_i \\ \text{Glutamine} + \alpha\text{-Ketoglutarate} + NAD(P)H + H^{\oplus} &\longrightarrow 2\ \text{Glutamate} + NAD(P)^{\oplus} \\ \hline 2\ NH_3 + \alpha\text{-Ketoglutarate} + NAD(P)H + 2\ ATP + H^{\oplus} &\longrightarrow \text{Glutamine} + NAP(P)^{\oplus} + 2\ ADP + 2\ P_i \end{aligned}$$

The coupled reactions in (b) consume one more ATP molecule than the coupled reactions in (a). Because the K_m of glutamine synthetase for NH_3 is much lower than the K_m of glutamate dehydrogenase for $NH_4^{\oplus}$, the coupled reactions in (b) predominate when $NH_4^{\oplus}$ levels are low. Thus, more energy is spent to assimilate ammonia when its concentration is low.

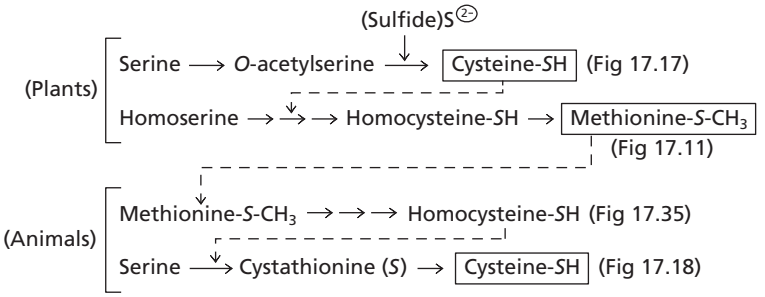
3. The ^{15}N-labeled amino group is transferred from aspartate to α-ketoglutarate, producing glutamate in a reaction catalyzed by aspartate transaminase (Figure 17.10). Since transaminases catalyze near-equilibrium reactions and many transaminases use glutamate as the α-amino group donor, the labeled nitrogen is quickly distributed among the other amino acids that are substrates of glutamate-dependent transaminases.

4. (a) α-Ketoglutarate + Amino acid $\rightleftharpoons$ Glutamate + α-Keto acid
Oxaloacetate + Amino acid $\rightleftharpoons$ Aspartate + α-Keto acid
Pyruvate + Amino acid $\rightleftharpoons$ Alanine + α-Keto acid

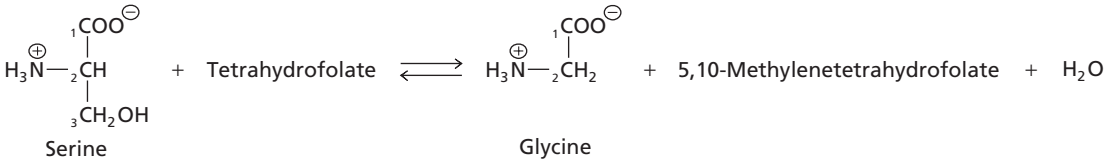
(b)

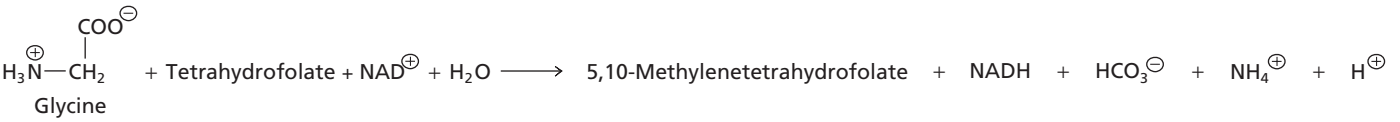


5.



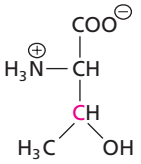
6. (a) C-3 of serine is transferred to tetrahydrofolate during the synthesis of glycine, and C-2 is transferred to tetrahydrofolate when glycine is cleaved to produce ammonia and bicarbonate.



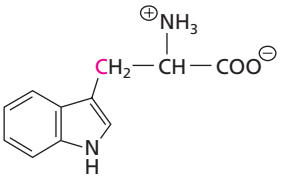


(b) Serine is synthesized from 3-phosphoglycerate (Figure 17.15), an intermediate of glycolysis. C-3 of both 3-phosphoglycerate and serine is derived from either C-1 or C-6 of glucose, and C-2 of both 3-phosphoglycerate and serine is derived from either C-2 or C-5 of glucose.

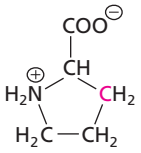
7. (a)



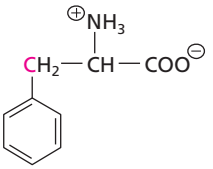
(b)



(c)



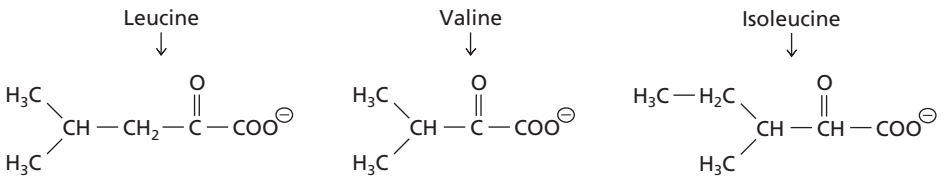
(d)



8. (a) Glutamic acid. PPI inhibits glutamine synthetase.
(b) Histidine biosynthesis pathway (Figure 17.23).

9. Aspartame is a dipeptide consisting of an asparate and a phenylalanine residue joined by a peptide bond. This bond is eventually hydrolyzed inside the cell producing aspartate and phenylalanine. Phenylketonuria patients must avoid any excess phenylalanine.

10. (a)



(b) Lysine degradation pathway. α-Aminoadipate δ-semialdehyde synthase is deficient (Figure 17.39).
(c) Urea cycle. Argininosuccinate synthetase is deficient (Figure 17.43).

11. (a) Alanine (c) Glycine
(b) Aspartate (d) Cysteine

12. The urea cycle does not operate in muscle, so ammonia from the deamination of amino acids cannot be converted to urea. Because high concentrations of ammonia are toxic, ammonia is converted to other products for disposal. In the first pathway, ammonia is incorporated into glutamine by the action of glutamine synthetase (Figure 17.5). Glutamine can then be

transported to the liver or kidneys. The second pathway is the glucose-alanine cycle (Figure 17.45). Pyruvate accepts the amino group of amino acids by transamination, and the alanine produced is transported to the liver where it can be deaminated back to pyruvate. The amino group is used for urea synthesis, and the pyruvate can be converted to glucose.

13. Inhibition of nitric oxide synthase (NOS) can prevent excess amounts of nitric oxide from being produced in cells lining the blood vessels. Nitric oxide causes relaxation of the vessels and in excess amounts can cause reduced blood pressure leading to shock. Thiocitrulline and *S*-methylthocitrulline inhibit NOS because they are unreactive analogs of the NOS reaction product citrulline (Figure 17.25).

14. There are two reasons. Firstly, many of the amino acid biosynthesis pathways aren't found in humans, so there won't be any metabolic diseases of nonexistent essential amino acid pathways. Secondly, the remaining pathways are probably crucial pathways during development so that any defects in these pathways are likely to be lethal. This is the same reasoning that we used to explain the lack of metabolic diseases in the sphingolipid biosynthesis pathways (Box 16.2).

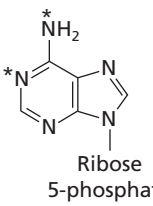
15. The 21st, 22nd, and 23rd amino acids are *N*-formylmethionine, selenocysteine, and pyrrolysine. *N*-formylmethionine and selenocysteine are synthesized during translation on aminoacylated tRNA and not by the standard metabolic pathways covered in this chapter. Pyrrolysine may also be synthesized on aminoacylated tRNA. The precursors are methionine, serine, and lysine.

16. The precursor in the serine biosynthesis pathway is 3-phosphoglycerate. This precursor can be derived from glyceraldehyde-3-phosphate (G3P) in the glycolytic pathway, where the conversion is associated with the *gain* of 1 ATP + 1 NADH. This gain must be subtracted from the total cost of G3P synthesis. Therefore, the cost of making 3-phosphoglycerate is 24 − 3.5 = 20.5 ATP equivalents, assuming that each NADH is equivalent to 2.5 ATPs. (The same cost can be derived from the Calvin cycle pathway.) The serine biosynthesis pathway produces one NADH when 3-phosphoglycerate is oxidized to 3-phosphohydroxypyruvate, so the next cost of making serine is 20.5 − 2.5 = 18 ATP equivalents. This value is identical to the value given in Box 17.3. (Note that the transamination reaction in the serine biosynthesis pathway is cost-free.)

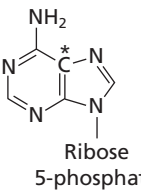
Alanine is made from pyruvate in a simple, cost-free, transamination reaction. The cost of making pyruvate can be estimated from the conversion of 3-phosphoglycerate to pyruvate in the glycolytic pathway. This conversion is associated with a gain of 1 ATP, so the cost of pyruvate is 20.5 − 1 = 19.5 ATP equivalents. Thus, the cost of synthesizing alanine is 19.5 ATP equivalents, or 20 ATP equivalents when rounded to two significant figures. This value is the same as that given in Box 17.3.

Chapter 18 Nucleotide Metabolism

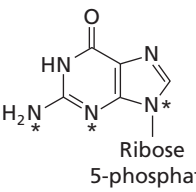
1. (a)



(b)



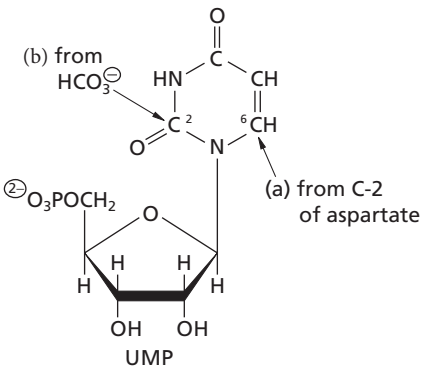
(c)



See Figure 18.10 for the reactions in the pathway of UMP synthesis.

(d) Labeled C-2 from aspartate, which is incorporated into carbamoyl aspartate, appears at C-6 of the uracil of UMP.

(e) The labeled carbon from $HCO_3^{\ominus}$, which is incorporated into carbamoyl phosphate, appears at C-2 of the pyrimidine ring of UMP.



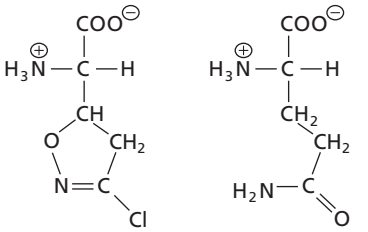
2. Seven ATP equivalents are required. One ATP is cleaved to AMP when PRPP is synthesized (Figure 18.3). The pyrophosphoryl group of PRPP is released in step 1 of the IMP biosynthetic pathway and subsequently hydrolyzed to 2 P_i (Figure 18.5), accounting for the second ATP equivalent. Five ATP molecules are consumed in steps 2, 4, 5, 6, and 7.

3. Purines: Reaction 3: GAR transformylase 10-formyl-THF, C-8 position.

Reaction 9: AICAR transformylase, 10-formyl-THF, C-2 position.

Pyrimidines: Thymidylate synthase, 5,10-methylene-THF, 5-CH_3 of thymidylate.

4. (a)



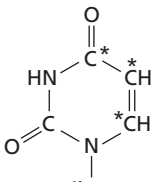
Acivicin Glutamine

(b) Acivicin inhibits glutamine-PRPP amidotransferase, the first enzyme in the purine biosynthetic pathway, so PRPP accumulates.

(c) Acivicin inhibits the carbamoyl phosphate synthetase II activity of dihydroorotate synthase that catalyzes the first step in the pyrimidine biosynthetic pathway.

5. (a) When β-alanine is used instead of aspartate, no decarboxylation reaction (step 6 of the *E. coli* pathway) would be required.

(b)



Ribose 5-phosphate

6. (a) dUMP + $NH_4^{\oplus}$

(b) Synthesis of DNA requires certain ratios of A, T, G and C. If dTTP levels are higher than necessary, dTTP will act to decrease its own synthesis pathway by inhibiting the conversion of dCMP to dUMP by dCMP deaminase. dUMP is the precursor to dTMP (thymidylate synthase, Figure 18.16), and the subsequent conversion to dTDP and dTTP (needed for DNA synthesis). On the other hand, if dCTP levels are high, activation of dCMP deaminase will lead to an increased conversion of dCMP to dUMP and this diverts any dCMP that might have been converted to more dCTP by phosphorylation (Figure 18.20).

7. Four ATP equivalents are required. One ATP equivalent is required for the synthesis of PRPP from ribose 5-phosphate (Figure 18.3). Carbamoyl phosphate synthesis requires 2 ATP (Figure 18.10, step 1). One ATP equivalent is consumed in step 5, when PP_i is hydrolyzed to 2 P_i.

8. In the absence of adenosine deaminase, adenosine and deoxyadenosine are not degraded via inosine and hypoxanthine to uric acid (Figure 18.19 and 18.21). This leads to an increase in the concentration of deoxyadenosine, which can be converted to dATP. High concentrations of dATP inhibit ribonucleotide reductase (Table 18.1). The inhibition of ribonucleotide reductase results in decreased production of all deoxynucleotides and therefore inhibits DNA synthesis.

9. Glutamine-PRPP amidotransferase is the first enzyme and the principal site of regulation in the *de novo* pathway to IMP (Figure 18.5). In humans, PRPP is both a substrate and a positive effector of this enzyme. An increase in the cellular levels of PRPP due to increased PRPP synthetase activity will therefore enhance the activity of the amidotransferase. This will result in an increased synthesis of IMP and other purine nucleosides and nucleotides. Overproduction of purine nucleotides and subsequent degradation can lead to elevated uric acid levels characteristic of gout.

10. (a) ATP (b) ATP (c) ATP (d) GTP (e) UTP (f) GTP (g) C TP (h) UTP (i) ATP (j) IMP (k) IMP

11. Purines and pyrimidines are not significant sources of energy. The carbon atoms of fatty acids and carbohydrates can be oxidized to yield ATP, but there are no comparable energy-yielding pathways for nitrogen-containing purines and pyrimidines. However, the NADH produced when hypoxanthine is converted to uric acid may indirectly generate ATP via

oxidative phosphorylation. The degradation of uracil and thymine yields acetyl CoA and succinyl CoA, respectively, which can be metabolized via the citric acid cycle to generate ATP.

12. The sugar D-ribose exists as an equilibrium mixture of α-D-ribopyranose, α-D-ribofuranose, β-D-ribopyranose, and β-D-ribofuranose. These forms freely interconvert with each through the open-chain form (Section 8.2).

13. Xanthine is 2,6-dioxopurine; hypoxanthine is 6-oxopurine; orotate is 2,4-dioxo-6-carboxylpyrimidine.

14. SAICAR synthetase + adenylosuccinate lyase in the IMP biosynthesis pathway (Figure 18.5) and argininosuccinate synthetase + argininosuccinate lyase in the arginine biosynthesis pathway (urea cycle: Figure 17.43).

Chapter 19 Nucleic Acids

1. In the α helix, hydrogen bonds form between the carbonyl oxygen of one residue and the amine hydrogen four residues, or one turn, away. These hydrogen bonds between atoms in the backbone are roughly parallel to the axis of the helix. The amino acid side chains, which point away from the backbone, do not participate in intrahelical hydrogen bonding. In double-stranded DNA, the sugar-phosphate backbone is not involved in hydrogen bonding. Instead, two or three hydrogen bonds, which are roughly perpendicular to the helix axis, form between complementary bases in opposite strands.

In the α helix, the individual hydrogen bonds are weak, but the cumulative forces of these bonds stabilize the helical structure, especially within the hydrophobic interior of a protein where water does not compete for hydrogen bonding. In DNA, the principal role of hydrogen bonding is to allow each strand to act as a template for the other. Although the hydrogen bonds between complementary bases help stabilize the helix, stacking interactions between base pairs in the hydrophobic interior make a greater contribution to helix stability.

2. If 58% of the residues are (G + C), 42% of the residues must be (A + T). Since every A pairs with a T on the opposite strand, the number of adenine residues equals the number of thymine residues. Therefore, 21%, or 420, of the residues are thymine ($2000 \times 0.21 = 420$).

3. (a) The base compositions of complementary strands of DNA are usually quite different. For example, if one strand is poly dA (100% A), the other strand must be poly dT (100% T). However, since the two strands are complementary, the amount of (A + T) must be the same for each strand, and the amount of (G + C) must be the same for each strand.
(b) (A + G) = (T + C). Complementarity dictates that for every purine (A or G) on one strand, there must be a pyrimidine (T or C) on the complementary strand.

4. Since the DNA strands are anti-parallel, the complementary strand runs in the opposite direction. The sequence of the double-stranded DNA is

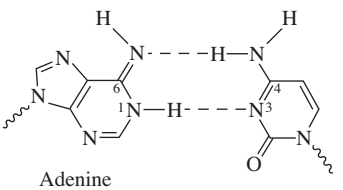
ATCGCGTAACATGGATTCGG
TAGCGCATTGTACCTAAGCC

By convention, DNA sequences are written in the 5′ → 3′ direction. Therefore, the sequence of the complementary strand is

CCGAATCCATGTTACGCGAT

5. The stability of the single-stranded helix is largely due to stacking interactions between adjacent purines. Hydrophobic effects also contribute, since the stacked bases form an environment that is partially shielded from water molecules.

6.



Adenine (imino form) Cytosine

7. There will be two discrete melting points separated by a plateau. When the extra strand of poly dT is released, the absorbance of the solution at 260 nm will increase as the stacked bases leave the largely hydrophobic interior of the triple helix. A second increase in the absorbance occurs when the remaining two DNA strands denature.

8. The sequence is

5′ ACGCACGUAUAUGUACUUAUACGUGGCU 3′

The underlined sequences are palindromic.

9. The main products will be a mixture of mononucleotides *and* pieces of single-stranded DNA approximately 500 bp in length. A piece of DNA with an enzyme molecule bound at each end will be degraded until the two strands can no longer base-pair; at that point the single strands cease to be a substrate for the enzyme.

10. In the 30 nm fiber, DNA is packaged in nucleosomes, each containing about 200 bp of DNA; therefore, the DNA in a nucleosome has a molecular weight of 130,000 (200 × 650 = 130,000). Assuming there is one molecule of histone H1 per nucleosome, the molecular weight of the protein component of the nucleosome would be 129,800.

Histone H1	21,000
Histone H2A (×2)	28,000
Histone H2 B (×2)	27,600
Histone H3 (×2)	30,600
Histone H4 (×2)	22,600
Total	129,800

Thus, the ratio by weight of protein to DNA is 129,800:130,000, or approximately 1:1.

11. Nucleosomes are composed of histones plus 200 base pairs of DNA. Since you inherited half your chromosomes from your mother, the oocyte contained

$$(3.2 \times 10^9 \text{ bp}) \times \frac{1 \text{ nucleosome}}{200 \text{ bp}} = 8 \times 10^6 \text{ nucleosomes}$$

(You inherited no nucleosomes from your father since nucleosomes are replaced by small, positively charged polypeptides during spermatogenesis.)

12. (a) pdApdGpdT + pdC
(b) pdAp + dGpdTpdC
(c) pdA + pdGpdTpdC

13. Since the supercoiled plasmid DNA is in equilibrium with relaxed DNA containing short unwound regions, the *Aspergillus* enzyme will slowly convert the DNA into nicked circles. Eventually the enzyme will convert the relaxed circles into unit-length linear fragments of double-stranded DNA.

14. Yes. The sugar-phosphate backbone in both RNA and DNA contains phosphodiester bonds that link the sugar residues.

15. pppApCpUpCpApUpApGp + CpUpApUpGp + ApGp + U

16. Bacteriophages have evolved several mechanisms to protect their DNA from restriction endonucleases. In general, bacteriophage DNA contains few restriction sites. Restriction endonuclease recognition sites are strongly selected and any mutations that alter these sites will be favored. In addition, restriction sites are often methylated, as in the bacterial chromosome. This is presumably due to a fortuitous event in the distant past when the phage DNA became methylated before it could be cleaved.

Some bacteriophages incorporate modified nucleotides into their DNA. The modified nucleotides (e.g., 5-hydroxymethylcytosine in bacteriophage T4) are not recognized by restriction endonucleases.

Phage genomes may also encode an enzyme that inactivates restriction endonucleases, or they may encode proteins that bind to restriction sites to prevent cleavage.

17. (a) The probability can be estimated from the probability of each nucleotide in the *Hind* III restriction site. (G = C = 0.18 and A = T = 0.32)
For the sequence AAGCTT there will be, on average, one *Hind*III site every
1/(0.32)(0.32)(0.18)(0.18)(0.32)(0.32) = 2943 bp
Thus, in a 100 Mb genome there will be, on average,
100,000/2943 = 33,070 sites
(b) 24,414

18. Although the recognition sites for *Bgl*II and *Bam*H1 differ, the enzymes produce fragments with identical sticky ends. These fragments can be ligated as easily as fragments produced by a single enzyme.

*Bgl*II
~~~A GATCT~~~
~~~TCTAG A~~~

*Bam*HI
~~~G GATCC~~~
~~~CCTAG G~~~

19. Restriction enzymes present in normal host cells might cleave newly introduced recombinant molecules, making it impossible to clone certain fragments of DNA. Using a host strain that does not make restriction endonucleases avoids this problem.

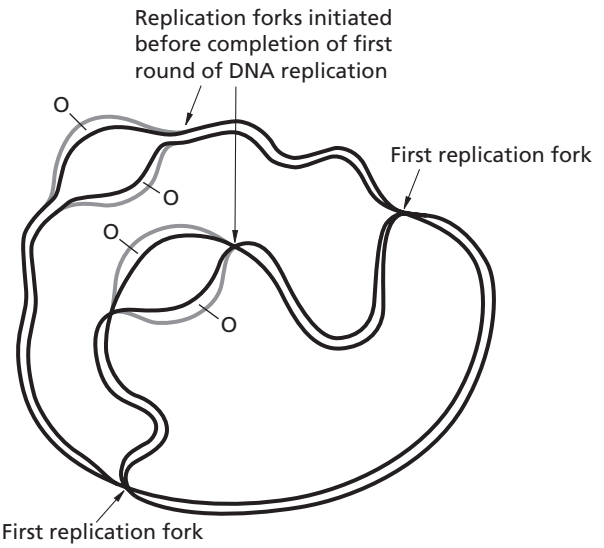
A mutation in RecA reduces recombination, thereby preventing the rearrangement of recombinant DNA molecules during propagation in the host cells. Rearrangement is often a problem, particularly when the cloned fragment of DNA contains repetitive sequences that can serve as sites for homologous recombination.

Chapter 20 DNA Replication, Repair, and Recombination

1. (a) Two replication forks form at the origin of replication and move in opposite directions until they meet at a point opposite the origin. Therefore, each replisome replicates half the genome (2.6×10^6 base pairs). The time required to replicate the entire chromosome is

$$\frac{2.6 \times 10^6 \text{ base pairs}}{1\,000 \text{ base pairs s}^{-1}} = 2600 \text{ s} = 43 \text{ min and } 20 \text{ s}$$

(b) Although there is only one origin (O), replication can be reinitiated before the previous replication forks have reached the termination site. Thus, the chromosome can contain more than two replication forks. Replication of a single chromosome still requires approximately 43 minutes, but completed copies of each chromosome can appear at shorter intervals, depending on the rate of initiation.



2. T4 DNA polymerase should be an early gene product because it is required for replication of the viral genome.

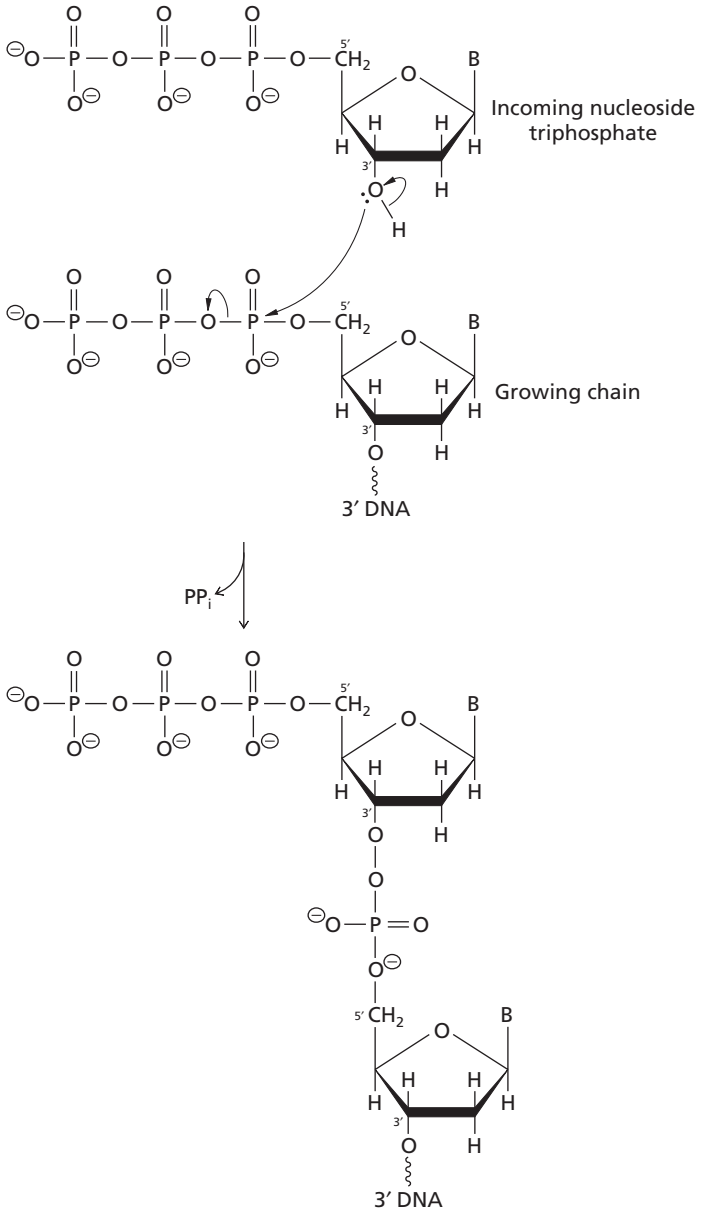
3. (a) The single-stranded DNA template used for DNA synthesis *in vitro* can form secondary structures such as hairpins. SSB prevents the formation of double-stranded structure by binding to the single-stranded template. SSB thus renders the DNA a better substrate for DNA polymerase.

(b) The yield of DNA *in vitro* is improved at higher temperatures because formation of secondary structure in the template is less likely. A temperature of 65°C is high enough to prevent formation of secondary structure but not high enough to denature the newly synthesized DNA. DNA polymerases from bacteria that grow at high temperatures are used because they are active at 65°C, a temperature at which DNA polymerases from other bacteria would be inactive.

4. Extremely accurate DNA replication requires a proofreading mechanism to remove errors introduced during the polymerization reaction. Synthesis of an RNA primer by a primase, which does not have proofreading activity, is more error prone than DNA synthesis. However,

because the primer is RNA, it can be removed by the 5′ → 3′ exonuclease activity of DNA polymerase I and replaced with accurately synthesized DNA when Okazaki fragments are joined. If the primer were composed of DNA made by a primase without proofreading activity, it would not be removed by DNA polymerase I and the error rate of DNA replication would be higher at sites of primer synthesis.

5. (a) In the hypothetical nucleotidyl group transfer reaction, the nucleophilic 3′-hydroxyl group of the *incoming* nucleotide would attack the triphosphate group of the growing chain. Pyrophosphate would be released when a new phosphodiester linkage was formed.



(b) If the hypothetical enzyme had 5′ → 3′ proofreading activity, removal of a mismatched nucleotide would leave a 5′-monophosphate group at the end of the growing chain. Further DNA synthesis, which would require a terminal triphosphate group, could not occur.

6. Topisomerase II or gyrase relieves supercoiling ahead of and behind the replication fork. If this enzyme is inhibited, the unwinding of the parental DNA cannot occur. Therefore, the DNA of the *E. coli* cannot be replicated.

7. (a) Assume that the genome is one large linear molecule of DNA and that the origin of replication is at the midpoint of this chromosome. Since the replication forks move in opposite directions, 60 base pairs can be replicated per second. The time required to replicate the entire genome would be

$$\frac{1.65 \times 10^8 \text{ base pairs}}{60 \text{ base pairs s}^{-1}} = 2.75 \times 10^6 \text{ s} = 764 \text{ h} = 32 \text{ days}$$

(b) Assuming that the 2000 bidirectional origins are equally spaced along the DNA molecule and that initiation occurs simultaneously at all origins, the rate would be $2000 \times 2 \times 30$ base pairs per second, or 1.2×10^5 base pairs per second. The time required to replicate the entire genome would be

$$\frac{1.65 \times 10^8 \text{ base pairs}}{1.2 \times 10^5 \text{ base pairs s}^{-1}} = 1375 \text{ s} = 23 \text{ min}$$

(c) Assume that the origins are equally spaced and that initiation at all origins is simultaneous. The required rate of replication is

$$\frac{1.65 \times 10^8 \text{ base pairs}}{300 \text{ s}} = 5.5 \times 10^5 \text{ base pairs s}^{-1}$$

Bidirectional replication from each fork proceeds at an overall rate of 60 base pairs per second. The minimum number of origins would be

$$\frac{5.5 \times 10^5 \text{ base pairs s}^{-1}}{60 \text{ base pairs s}^{-1} \text{ origin}^{-1}} = 9170 \text{ origins}$$

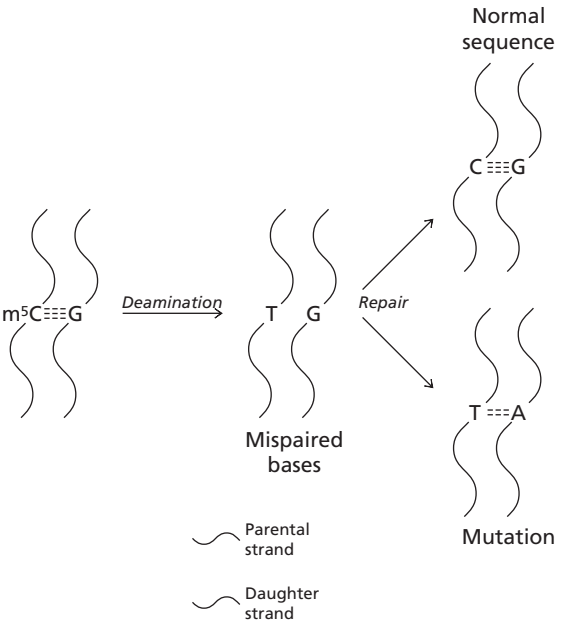
8. The modified G can no longer form a productive Watson-Crick base pair with C but can now base-pair with T. Therefore, one of the daughter strands of DNA will contain a T across from the modified base. After further rounds of replication, the T will base-pair with A and what was originally a G/C base pair will have mutated into an A/T base pair.

9. Ultraviolet light can damage DNA by causing dimerization of thymidylate residues. One mechanism for repairing thymine dimers is enzymatic photoreactivation, catalyzed by DNA photolyase. This enzyme uses energy from visible light to cleave the dimer and repair the DNA. Thus, cells that are exposed to visible light following ultraviolet irradiation are better able to repair DNA than cells kept in the dark.

10. (a) DNA from a *dut*$^-$ strain will appear normal because the Ung enzyme will remove any uracil that gets incorporated.
(b) DNA from a *dut*$^-$, *ung*$^-$ strain will contain dU residues in the place of some dT residues.

11. The DNA repair enzyme uracil *N*-glycosylase removes uracil formed by the hydrolytic deamination of cytosine. Because the enzyme does not recognize thymine or the other three bases normally found in DNA, it cannot repair the damage when 5-methylcytosine is deaminated to thymine.

12. High mutation rates occur at methylcytosine-containing regions because the product of deamination of 5-methylcytosine is thymine, which cannot be recognized as abnormal. When the mismatched T/G base pair that results from deamination of methylcytosine is repaired, the repair enzymes may delete either the incorrect thymine or the correct guanine. When the guanine is replaced by adenine, the resulting A/T base pair is a mutation.



13. Proofreading during replication results in excision of 99% of misincorporated nucleotides, thus reducing the overall error rate to 10^{-7}. Of those errors that escape the proofreading step, a further 99% are corrected by repair enzymes. The overall mutation rate is therefore 10^{-9}.

14. Yes. The *E. coli* enzyme DNA ligase is required to seal the nicks left in the DNA strands following DNA repair. This enzyme has a strict requirement for $NAD^{\oplus}$.

15. The dimers can be removed by excision repair. UvrABC endonuclease removes a 12–13 residue segment containing the pyrimidine dimer. The DNA oligonucleotide is removed with the help of a helicase. The gap is filled by the action of DNA polymerase I, and the nick sealed by the action of DNA ligase. The dimers can also be repaired through direct repair. DNA photolyase binds to the distorted double helix at the site of the dimer. As the DNA–enzyme complex absorbs light, the dimerization reaction is reversed.

16. The repair enzymes need an undamaged template in order to repair mutations in DNA. If both strands of the DNA molecule have been damaged, there is not a template to use for repair.

17. The proteins that catalyze strand exchange recognize regions of high sequence similarity and promote formation of a triple-stranded intermediate in which the invading strand base-pairs with a complementary strand. This pairing would not be possible if the sequences of the two DNA molecules were different.

18. DNA polymerase III is a component of the replisome that synthesizes the leading strand and the lagging strand during replication of the *E. coli* chromosome. DNA polymerase I is required to remove the short RNA primers on the lagging strand.

Chapter 21 Transcription and RNA Processing

1. (a) Since the rate of transcription is 70 nucleotides per second and each transcription complex covers 70 base pairs of DNA, an RNA polymerase completes a transcript and leaves the DNA template each second (assuming that the complexes are densely packed). Therefore, when the gene is loaded with transcription complexes, 60 molecules of RNA are produced per minute.

(b) Since each transcription complex covers 70 base pairs, the maximum number of complexes is

$$\frac{6000 \text{ base pairs}}{70 \text{ base pairs transcription complex}} = 86 \text{ transcription complexes}$$

2. (a) Since the average *E. coli* gene is 1 kb (1000 bp) long, 4000 genes account for 4000 kb of DNA. The percentage of DNA that is not transcribed is

$$\frac{500 \text{ kb}}{4600 \text{ kb}} \times 100\% = 10.9\%$$

Most of the nontranscribed DNA consists of promoters and regions that regulate transcription initiation.

(b) Since the gene products in mammals and bacteria are similar in size, the amount of DNA in the exons of a typical mammalian gene must also be 1000 bp. The total amount of DNA in exons is

$$5 \times 10^4 \text{ genes} \times 1.0 \text{ kb gene}^{-1} = 5 \times 10^4 \text{ kb}$$

This DNA represents about 1.7% of the mammalian genome.

$$\frac{5 \times 10^4 \text{ kb}}{3 \times 10^6 \text{ kb}} \times 100\% = 1.7\%$$

The remaining 97.5% of DNA consists of introns and other sequences.

3. No. It is extremely unlikely that the eukaryotic gene's promoter will contain the correct sequences in the correct location to permit accurate initiation by the prokaryotic RNA polymerases. Likewise, it is extremely unlikely that the prokaryotic gene's promoter will contain the correct sequence in the correct location to permit accurate initiation by RNA polymerase II.

4. No. A typical eukaryotic triose phosphate isomerase gene contains introns. The prokaryotic cell contains no spliceosomes and therefore will not be able to correctly process the primary transcript. Therefore, translation of the RNA will yield an aberrant protein fragment.

5. (a) In the presence of both lactose and glucose, the *lac* operon is transcribed at a low level because *lac* repressor forms a complex with allolactose (an isomer of lactose). Because the

allolactose–repressor complex cannot bind to the promoter region of the *lac* operon, the repressor does not prevent initiation of transcription.

(b) In the absence of lactose, no allolactose is formed. Thus, *lac* repressor binds near the *lac* operon promoter and prevents transcription.

(c) When lactose is the sole carbon source, the *lac* operon is transcribed at the maximum rate. In the presence of allolactose, transcription is allowed since *lac* repressor does not bind to the promoter region of the *lac* operon. Also, in the absence of glucose, the transcription rate increases because cAMP production increases, making more CRP-cAMP available to bind to the promoter region of the *lac* operon. The absence of the repressor and the enhancement of transcription initiation by CRP-cAMP allow the cell to synthesize the quantities of enzymes required to support growth when lactose is the only carbon source.

6. Since the wild-type *lac* promoter is relatively weak, maximal transcription requires the activator CRP. The UV5 mutations alters the −10 region such that it now resembles the consensus −10 sequence, making it a much stronger promoter. In the absence of the *lac* repressor, the promoter is independent of CRP.

7. ^{32}P appears only at the 5′ end of mRNA molecules that have ATP as the first residue. It does not appear in any other residues because pyrophosphate, which includes the β-phosphoryl group, is released when nucleoside triphosphates are added to the 3′ end of a growing RNA chain (Figure 21.3).

When the 5′ end of mRNA is capped, only the γ-phosphoryl group of the initial residue is removed when the cap forms. The β-phosphoryl group, which contains the label, is retained and receives the GMP group from GTP (Figure 21.26).

8. The lack of proofreading activity in RNA polymerase makes the error rate of transcription greater than the error rate of DNA replication. However, the defective RNA molecules produced are not likely to affect cell viability because most copies of RNA synthesized from a given gene are normal. In the case of defective mRNA, the number of defective proteins is only a small percentage of the total number of proteins synthesized. Also, mistakes made during transcription are quickly eliminated since most mRNA molecules have a short half-life.

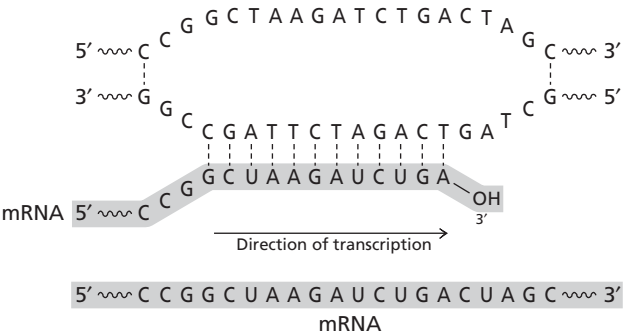
9. During maturation, eukaryotic mRNA precursors are modified at their 3′ ends by the addition of a poly A tail. When a mixture of components from a cell extract is passed over the column, the poly A tail will hybridize with oligo dT on the column. The other components in the cell extract will pass through the column. The bound mature mRNA with the poly A tail is removed from the column by changing the pH or the ionic strength of the buffer. This will disrupt the hydrogen bonds between the A and T nucleotides.

10. (a) A much lower concentration of rifampicin stopped the growth of the wild-type *E. coli* (<5 μg/mL) as compared to the concentration of rifampicin that stopped the growth of the mutant (>50 μg/mL).

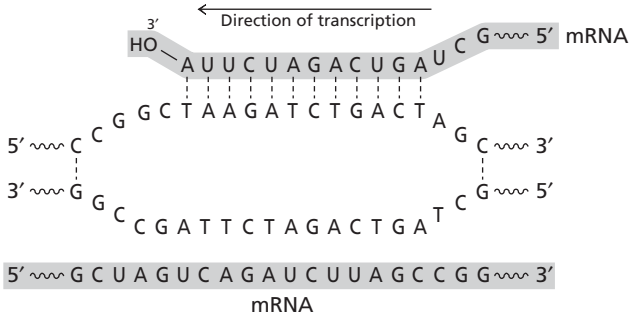
(b) RNA polymerase consists of a core enzyme with a stoichiometry of $\alpha_2\beta\beta'\omega$ that participates in many of the transcription reactions. The large β and β' subunits make up the active site of the enzyme.

(c) The rifampicin-resistant bacteria could arise from mutations that occur in the gene for the β subunit of RNA polymerase.

11. Since either strand can serve as a template, two mRNA molecules can be transcribed from this DNA segment. When the bottom strand is the template, the mRNA sequence is complementary to the bottom strand.



When the top strand is the template, the mRNA sequence is complementary to the top strand.



12. A gene was defined as a DNA sequence that is transcribed. By this definition, the entire ribosomal RNA operon is a gene. However, it is sometimes more convenient to restrict the term *gene* to the segment of RNA that encodes a functional product, for example, one of the enzymes encoded by the *lac* operon. The operon in Figure 21.25 therefore contains tRNA and 16S, 23S, and 5S rRNA genes. The DNA sequences between these genes, although transcribed, are not considered part of any gene.

13. The genomic DNA sequence provides an accurate rendition of the primary RNA sequence as expected. However, sequencing a purified tRNA reveals that many of the nucleotides have been specifically modified post-transcriptionally. The same is true for eukaryotes.

14. The gene for triose phosphate isomerase in maize contains about 3400 base pairs. If the spliceosome assembles at the first intron, then 2900 base pairs remain to be transcribed. The time required to transcribe 2900 base pairs is 97 seconds (2900 nucleotides ÷ 30 nucleotides per second). If the spliceosome assembles immediately after transcription of the first intron, and if splicing cannot begin until transcription of the entire gene is complete, the spliceosome must be stable for at least 97 seconds.

15. The CRP-cAMP binding site probably overlaps the promoter of the gene. When CRP-cAMP binds, the promoter is blocked and transcription cannot occur.

16. When the sequence of the 5′ or 3′ splice site or the branch point is altered by mutation, proper splicing cannot occur and no functional mRNA can be produced.

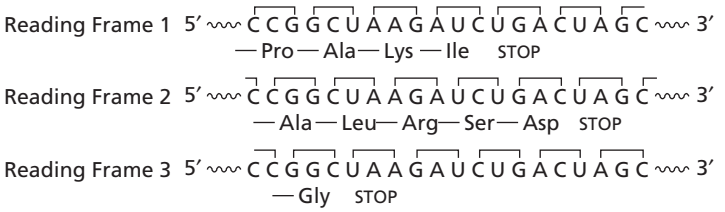
17. Yes. Once the U2 snRNP binds to the branch site it will occlude the U5 snRNP from binding to the 3′ splice acceptor and interfere with splicing. Furthermore, the deletion will have removed a large part of the pyrimidine stretch required for binding to the 3′ splice site. Both of these will prevent proper mRNA processing and the aberrant RNA will not be properly translated.

Chapter 22 Protein Synthesis

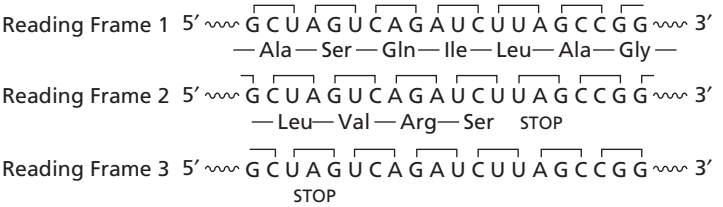
1. One strand of DNA has three different overlapping reading frames, therefore a double-stranded DNA has six reading frames. This can be seen by examining the DNA sequence beginning at the 5′ end of each strand and marking off the triplet codons. This identifies one reading frame on each strand. Now start at the second nucleotide in from the 5′ ends and mark off triplet codons; that is reading frame 2. The third reading frame on each strand begins at the third nucleotide in from the 5′ ends. The "fourth" reading frame is identical to the first—test this for yourself.

Using similar logic, it follows that if the genetic code were read in codons four nucleotides in length, then one strand of DNA could be read in four different reading frames and therefore a double-stranded piece of DNA would contain eight reading frames (four on each strand).

2. Each mRNA sequence could be translated in three different reading frames. For the first mRNA sequence, the possible codons and polypeptide sequences are



For the second mRNA sequence, the possible codons and polypeptide sequences are



Since only a reading frame without a stop codon can encode a polypeptide, the second mRNA sequence corresponds to the actual transcript. The sequence of the encoded polypeptide is –Ala–Ser–Gln–Ile–Leu–Ala–Gly–.

3. Two phosphoanhydride bonds are hydrolyzed for each amino acid activated by an aminoacyl-tRNA synthetase.

$$\text{Amino acid} + \text{tRNA} + \text{ATP} \longrightarrow \text{Aminoacyl} - \text{tRNA} + \text{AMP} + PP_i$$

$$PP_i + H_2O \longrightarrow 2\ P_i$$

The rest of the energy needed to synthesize the protein is provided by hydrolysis of GTP: one "high energy" bond is hydrolyzed in the formation of the 70S initiation complex, another during the insertion of each aminoacyl-tRNA into the A site of the ribosome, and another at each translocation step. Since the initial methionyl-tRNA is inserted into the P site, 599 new insertions and 599 translocations occur during the synthesis of a 600-residue protein. Finally, one phosphoanhydride bond is hydrolyzed during release of the completed polypeptide chain from the ribosome. The total number of phosphoanhydride bonds hydrolyzed during synthesis of the protein is

Activation (600×2)	1200
Initiation	1
Insertion	599
Translocation	599
Termination	1
Total	2400

4. The answer depends on your frame of reference. For example, relative to the ribosome, the mRNA and both tRNAs get translocated by one triplet codon. Relative to the mRNA, it is the ribosome that is shifted by three nucleotides.

5. The region of the mRNA molecule upstream of the true initiation codon contains the purine-rich Shine-Dalgarno sequence, which is complementary to a pyrimidine-rich sequence at the 3′ end of the 16S rRNA component of the 30S ribosomal subunit (Figure 22.17). By correctly positioning the 30S subunit on the mRNA transcript, the Shine-Dalgarno sequence allows fMct-$tRNA_f^{Mct}$ to bind to the initiation codon. Once protein synthesis begins, all subsequent methionine codons are recognized by Met-$tRNA^{Met}$.

6. No, because proper translation initiation in an *E. coli* cell requires a Shine-Dalgarno sequence located in the 5′ untranslated region of the mRNA. Since eukaryotic ribosomes do not have this requirement, it is extremely unlikely that an mRNA from a plant would fortuitously contain a Shine-Dalgarno sequence in the proper location.

If, however, the part of the gene encoding the plant mRNA were fused to a bacterial Shine-Dalgarno sequence, then the open reading frame for the plant protein would be properly translated in the bacterial cell.

7. The transcript of each rRNA gene is an rRNA molecule that is directly incorporated into a ribosome. Thus, multiple copies of rRNA genes are needed to assemble the large number of ribosomes that the cell requires. In contrast, the transcript of each ribosomal protein gene is an mRNA that can be translated many times. Because of this amplification of RNA to protein, fewer genes are needed for each ribosomal protein than for rRNA.

8. Possible suppressor tRNA species include all those that recognize codons differing from UAG by a single nucleotide, that is, tRNAs whose anticodons differ by a single nucleotide from the sequence CUA, which is complementary to the stop codon UAG. $tRNA^{Gln}$, $tRNA^{Lys}$, and $tRNA^{Gln}$ all recognize codons that differ only at the first position (codons CAG, AAG, and GAG, respectively). $tRNA^{Leu}$, $tRna^{Ser}$, and $tRNA^{Trp}$ recognize codons that differ only at the second position (UUG, UCG, and UGG, respectively). $tRNA^{Tyr}$ recognizes codons that differ only at the third position (UAU or UAC).

A cell that contains a suppressor tRNA can survive despite the loss of a normal tRNA because the cell also contains isoacceptor tRNA molecules that carry the same amino acid. Although the suppressor tRNA may occasionally insert an amino acid at a normally occurring stop codon, the resulting protein, which is larger than the normal gene product, is usually not lethal to the cell. In fact, strains of *E. coli* that contain suppressor tRNAs do survive but are often not as healthy as wild-type strains.

9. (a) Aminoacyl-tRNA synthetases—the enzymes that bind to tRNAs and catalyze aminoacylation.
(b) IF-2 in bacteria and eIF-2 in eukaryotes, a protein that binds to aminoacylated initiator-tRNA and loads it into the ribosome's P site during translation initiation.
(c) EF-Tu in bacteria and EF-1α in eukaryotes—a protein that binds to charged tRNAs and loads them into the ribosome's A site during polypeptide elongation.
(d) Ribosomes. These large complexes of RNA and protein contain two sites that can bind specifically to tRNAs, the A site and the P site.
(e) mRNA—tRNAs bind to mRNA through codon-anticodon hydrogen bonds.

The enzymes that modify specific residues on individual tRNAs during the maturation process must also be able to bind to tRNAs.

10. Under normal circumstances, when the translation machinery encounters UGA in RF-2 mRNA, RF-2 recognizes the stop codon and terminates protein synthesis. When the cellular concentration of RF-2 is low, however, the ribosome pauses at the termination codon, shifts frame, and continues translating the RF-2 mRNA to produce the full-length functional protein. Thus, the presence of the stop codon encourages translational frameshifting in the absence of RF-2 and allows RF-2 to regulate its own production.

11. (a) If the entire leader region were deleted, attenuation would not be possible, and transcription would be controlled exclusively by *trp* repressor. The overall rate of transcription of the *trp* operon would increase.
(b) If the region encoding the leader peptide were deleted, transcription would be controlled exclusively by *trp* repressor. Deletion of the sequence encoding the leader peptide would remove Sequence 1, thus allowing the stable 2–3 hairpin to form. Since neither the pause site (1–2 hairpin) nor the terminator (3–4 hairpin) could form, initiated transcripts would always continue into the *trp* operon.
(c) If the leader region did not contain an AUG codon, the operon would be rarely transcribed. Because of the absence of the initiation codon, the leader peptide would not be synthesized, and 1–2 hairpins and 3–4 hairpins would almost always form, leading to termination of transcription.

12. No, this is difficult to imagine. One of the important features of the attenuation model is that one or more codons in the leader peptide usually encode the amino acid that is synthesized by that operon. It is the relative shortage or abundance of particular aminoacylated tRNAs that modulates the attenuation. The products of the *lac* operon are not directly involved in amino acid biosynthesis, so we would not expect cellular levels of one class of aminoacylated tRNAs to vary with the activity of the operon.

13. The presence of codons specifying valine and leucine in the leader regions of isoleucine operons suggests that a scarcity of these amino acids would promote transcription of the genes for isoleucine biosynthesis. Many of the enzymes required to synthesize isoleucine are also required in the pathways to valine and leucine (Section 18.5A). Thus, even when the isoleucine concentration is high, a low concentration of valine or leucine ensures that transcription of the isoleucine operon does not terminate prematurely.

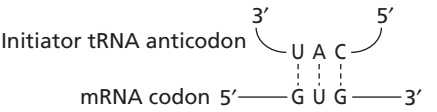
14. As the newly synthesized protein is extruded from the ribosome, the *N*-terminal signal peptide is recognized and bound by a signal-recognition particle (SRP). Further translation is inhibited until the SRP binds to its receptor on the cytosolic face of the endoplasmic reticulum. Ribophorins anchor the ribosome to the endoplasmic reticulum. When translation resumes, the polypeptide chain passes through a pore into the lumen. If the polypeptide does not pass completely through the membrane, the result is an integral membrane protein with its *N*-terminus in the lumen of the endoplasmic reticulum and its *C*-terminus in the cytosol.

Glycosylation of specific residues takes places in the lumen of the endoplasmic reticulum and in the Golgi apparatus. The protein, still embedded in the membrane, is transported between the endoplasmic reticulum and the Golgi apparatus in transfer vesicles that bud off the endoplasmic reticulum.

Secretory vesicles transport the fully glycosylated protein from the Golgi apparatus to the plasma membrane. When the vesicles fuse with the plasma membrane, the *N*-terminal portion of the protein, which was in the lumen, is now exposed to the extracellular space, and the *C*-terminal portion remains in the cytosol.

15. Yes. A hydrophobic secretion signal sequence located at the *N*-terminus of a protein is necessary and sufficient for entry into the cell's secretory pathway.

16. The initiator tRNA anticodon pairs with GUG by forming a G/U base pair between the 5′ nucleotide of the codon and the 3′ position of the anticodon.



This interaction is unrelated to wobble since the 5′ position of the anticodon is the wobble position.

Glossary of Biochemical Terms

A site. Aminoacyl site. The site on a ribosome that is occupied during protein synthesis by an aminoacyl-tRNA molecule.

acceptor stem. The sequence at the 5′ end and the sequence near the 3′ end of a tRNA molecule that are base paired, forming a stem. The acceptor stem is the site of amino acid attachment. Also known as the amino acid stem.

accessory pigments. Pigments other than chlorophyll that are present in photosynthetic membranes. The accessory pigments include carotenoids and phycobilins.

acid. A substance that can donate protons. An acid is converted to its conjugate base by loss of a proton. (The Lewis theory defines an acid as an electron-pair acceptor [Lewis acid].)

acid anhydride. The product formed by condensation of two molecules of acid.

acid dissociation constant (K_a). The equilibrium constant for the dissociation of a proton from an acid.

acid-base catalysis. Catalysis in which the transfer of a proton accelerates a reaction.

ACP. *See* acyl carrier protein.

activation energy. The free energy required to promote reactants from the ground state to the transition state in a chemical reaction.

activator. *See* transcriptional activator.

active site. The portion of an enzyme that contains the substrate-binding site and the amino-acid residues involved in catalyzing the conversion of substrate(s) to product(s). Active sites are usually located in clefts between domains or subunits of proteins or in indentations on the protein surface.

active transport. The process by which a solute specifically binds to a transport protein and is transported across a membrane against the solute concentration gradient. Energy is required to drive active transport. In primary active transport, the energy source may be light, ATP, or electron transport. Secondary active transport is driven by ion concentration gradients.

acyl carrier protein (ACP). A protein (in prokaryotes) or a domain of a protein (in eukaryotes) that binds activated intermediates of fatty acid synthesis via a thioester linkage.

adipocyte. A triacylglycerol-storage cell found in animals. An adipocyte consists of a fat droplet surrounded by a thin shell of cytosol in which the nucleus and other organelles are suspended.

adipose tissue. Animal tissue composed of specialized triacylglycerol-storage cells known as adipocytes.

A-DNA. The conformation of DNA commonly observed when purified DNA is dehydrated. A-DNA is a right-handed double helix containing approximately 11 base pairs per turn.

aerobic. Occurring in the presence of oxygen.

affinity chromatography. A chromatographic technique used to separate a mixture of proteins or other macromolecules in solution based on specific binding to a ligand that is covalently attached to the chromatographic matrix.

affinity labeling. A process by which an enzyme (or other macromolecule) is covalently inhibited by a reaction with a molecule that specifically interacts with the active site (or other binding site).

aldoses. A class of monosaccharides in which the most oxidized carbon atom, designated C-1, is aldehydic.

allosteric effector. *See* allosteric modulator.

allosteric interaction. The modulation of activity of a protein that occurs when a molecule binds to the regulatory site of the protein.

allosteric modulator. A biomolecule that binds to the regulatory site of an allosteric protein and thereby modulates its activity. An allosteric modulator may be an activator or an inhibitor. Also known as an allosteric effector.

allosteric protein. A protein whose activity is modulated by the binding of another molecule.

allosteric site. *See* regulatory site.

allosteric transitions. The changes in conformation of a protein between the active (R) state and the inactive (T) state.

α helix. A common secondary structure of proteins, in which the carbonyl oxygen of each amino acid residue (residue n) forms a hydrogen bond with the amide hydrogen of the fourth residue further toward the C-terminus of the polypeptide chain (residue $n + 4$). In an ideal right-handed α helix, equivalent positions recur every 0.54 nm, each amino acid residue advances the helix by 0.15 nm along the long axis of the helix, and there are 3.6 amino acid residues per turn.

amino acid. An organic acid consisting of an α-carbon atom to which an amino group, a carboxylate group, a hydrogen atom, and a specific side chain (R group) are attached. Amino acids are the building blocks of proteins.

amino acid analysis. A chromatographic procedure used for the separation and quantitation of amino acids in solutions such as protein hydrolysates.

amino terminus. *See* *N*-terminus.

aminoacyl site. *See* A site

aminoacyl-tRNA synthetase. An enzyme that catalyzes the activation and attachment of a specific amino acid to the 3′ end of a corresponding tRNA molecule.

amphibolic reaction. A metabolic reaction that can be both catabolic and anabolic.

amphipathic. Describes a molecule that has both hydrophobic and hydrophilic regions.

amyloplast. Modified chloroplasts that specialize in starch synthesis.

anabolic reaction. A metabolic reaction that synthesizes a molecule needed for cell maintenance and growth.

anaplerotic reaction. A reaction that replenishes metabolites removed from a central metabolic pathway (cf. cataplerotic).

angstrom (Å). A unit of length equal to 1×10^{-10} m, or 0.1 nm.

anion. An ion with an overall negative charge.

anode. A positively charged electrode. In electrophoresis, anions move toward the anode.

anomeric carbon. The most oxidized carbon atom of a cyclized monosaccharide. The anomeric carbon has the chemical reactivity of a carbonyl group.

anomers. Isomers of a sugar molecule that have different configurations only at the anomeric carbon atom.

antenna pigments. Light-absorbing pigments associated with the reaction center of a photosystem. These pigments may form a separate antenna complex or may be bound directly to the reaction-center proteins.

antibiotic. A compound, produced by one organism, that is toxic to other organisms. Clinically useful antibiotics must be specific for pathogens and not affect the human host.

antibody. A glycoprotein synthesized by certain white blood cells as part of the immunological defense system. Antibodies specifically bind to foreign compounds, called antigens, forming antibody-antigen complexes that mark the antigen for destruction. Also known as an immunoglobulin.

anticodon. A sequence of three nucleotides in the anticodon loop of a tRNA molecule. The anticodon binds to the complementary codon in mRNA during translation.

anticodon arm. The stem-and-loop structure in a tRNA molecule that contains the anticodon.

antigen. A molecule or part of a molecule that is specifically bound by an antibody.

antiport. The cotransport of two different species of ions or molecules in opposite directions across a membrane by a transport protein.

antisense strand. In double-stranded DNA the antisense strand is the strand that does not contain codons. Also called the template strand. The opposite strand is called the sense strand or the coding strand.

antisense RNA. An RNA molecule that binds to a complementary mRNA molecule, forming a double-stranded region that inhibits translation of the mRNA.

apoprotein. A protein whose cofactor(s) is absent. Without the cofactor(s), the apoprotein lacks the biological activity characteristic of the corresponding holoprotein.

apoptosis. The programed death of a cell.

atomic mass unit. The unit of atomic weight equal to 1/12th the mass of the ^{12}C isotope of carbon. The mass of the ^{12}C nuclide is exactly 12 by definition.

attenuation. A mechanism of regulation of gene expression that couples translation and transcription. Generally, the translation of a short reading frame at the beginning of a prokaryotic operon will determine whether transcription terminates before the rest of the operon is transcribed.

autophosphorylation. Phosphorylation of a protein kinase catalyzed by another molecule of the same kinase.

autosome. A chromosome other than a sex chromosome.

autotroph. An organism that can grow and reproduce using only inorganic substances (such as CO_2) as its only source of essential elements.

backbone. 1. The repeating $N—C_\alpha—C$ units connected by peptide bonds in a polypeptide chain. **2.** The repeating sugar-phosphate units connected by phosphodiester linkages in a nucleic acid.

bacteriophage. A virus that infects a bacterial cell.

base. 1. A substance that can accept protons. A base is converted to its conjugate acid by addition of a proton. (The Lewis theory defines a base as an electron-pair donor [Lewis base].) **2.** The substituted pyrimidine or purine of a nucleoside or nucleotide. The heterocyclic bases of nucleosides and nucleotides can participate in hydrogen bonding.

base pairing. The interaction between the bases of nucleotides in single-stranded nucleic acids to form double-stranded molecules, such as DNA, or regions of double-stranded secondary structure. The most common base pairs are formed by hydrogen bonding of adenine (A) with thymine (T) or uracil (U) and of guanine (G) with cytosine (C).

B-DNA. The most common conformation of DNA and the one proposed by Watson and Crick. B-DNA is a right-handed double helix with a diameter of 2.37 nm and approximately 10.4 base pairs per turn.

***β*-oxidation pathway.** The metabolic pathway that degrades fatty acids to acetyl CoA, producing NADH and QH_2 and thereby generating large amounts of ATP. Each round of *β*-oxidation of fatty acids consists of four steps: oxidation, hydration, further oxidation, and thiolysis.

***β* pleated sheet.** *See β* sheet.

***β* sheet.** A common secondary structure of proteins that consists of extended polypeptide chains stabilized by hydrogen bonds between the carbonyl oxygen of one peptide bond and the amide hydrogen of another on the same or an adjacent polypeptide chain. The hydrogen bonds are nearly perpendicular to the extended polypeptide chains, which may be either parallel (running in the same *N*- to *C*-terminal direction) or antiparallel (running in opposite directions).

***β* strand.** An extended polypeptide chain within a *β* sheet secondary structure or having the same conformation as a strand within a *β* sheet.

***β* turn.** *See* turn.

bile. A suspension of bile salts, bile pigments, and cholesterol that originates in the liver and is stored in the gall bladder. Bile is secreted into the small intestine during digestion.

binding-change mechanism. A proposed mechanism for the phosphorylation of ADP and release of ATP from F_0F_1 ATP synthase. The mechanism proposes three different binding-site conformations for ATP synthase: an open site from which ATP has been released, an ATP-bearing tight-binding site that is catalytically active, and an ADP and P_i loose-binding site that is catalytically inactive. Inward passage of protons through the ATP synthase complex into the mitochondrial matrix causes the open site to become a loose site; the loose site, already filled with ADP and P_i, to become a tight site; and the ATP-bearing site to become an open site.

bioenergetics. The study of energy changes in biological systems.

biological membrane. *See* membrane.

biopolymer. A biological macromolecule in which many identical or similar small molecules are covalently linked to one another to form a long chain. Proteins, polysaccharides, and nucleic acids are biopolymers.

Bohr effect. The phenomenon observed when exposure to carbon dioxide, which lowers the pH inside the cells, causes the oxygen affinity of hemoglobin in red blood cells to decrease.

branch migration. The movement of a crossover, or branch point, resulting in further exchange of DNA strands during recombination.

branch site. The point within an intron that becomes attached to the 5′ end of the intron during splicing of mRNA precursors.

buffer. A solution of an acid and its conjugate base that resists changes in pH.

buffer capacity. The ability of a solution to resist changes in pH. For a given buffer, maximum buffer capacity is achieved at the pH at which the concentrations of the weak acid and its conjugate base are equal (i.e., when $pH = pK_a$).

C_4 pathway. A pathway for carbon fixation in several plant species that minimizes photorespiration by concentrating CO_2. In this pathway, CO_2 is incorporated into C_4 acids in the mesophyll cells, and the C_4 acids are decarboxylated in the bundle sheath cells, releasing CO_2 for use by the reductive pentose phosphate cycle.

calorie (cal). The amount of energy required to raise the temperature of 1 gram of water by 1°C (from 14.5°C to 15.5°C). One calorie is equal to 4.184 J.

Calvin cycle. A cycle of reactions that involve the fixation of carbon dioxide and the net production of glyceraldehyde-3-phosphate. Usually associated with photosynthesis. Also known as the Calvin-Benson cycle, the C3 pathway, and the reductive pentose phosphate (RPP) cycle.

Calvin-Benson cycle. *See* Calvin cycle.

CAM. *See* Crassulacean acid metabolism.

cap. A 7-methylguanosine residue attached by a pyrophosphate linkage to the 5′ end of a eukaryotic mRNA molecule. The cap is added posttranscriptionally and is required for efficient translation. Further covalent modifications yield alternative cap structures.

carbanion. A carbon anion that results from the cleavage of a covalent bond between carbon and another atom in which both electrons from the bond remain with the carbon atom.

carbocation. A carbon cation that results from the cleavage of a covalent bond between carbon and another atom in which the carbon atom loses both electrons from the bond.

carbohydrate. Loosely defined as a compound that is a hydrate of carbon in which the ratio of C:H:O is 1:2:1. Carbohydrates include monomeric sugars (i.e., monosaccharides) and their polymers. Also known as a saccharide.

carboxyl terminus. *See C*-terminus.

carnitine shuttle system. A cyclic pathway that shuttles acetyl CoA from the cytosol to the mitochondria by formation and transport of acyl carnitine.

cascade. Sequential activation of several components, resulting in signal amplification.

catabolic reaction. A metabolic reaction that degrades a molecule to provide smaller molecular building blocks and energy to an organism.

catabolite repression. A regulatory mechanism that results in increased rates of transcription of many bacterial genes and operons when glucose is present. A complex between cAMP and cAMP regulatory protein (CRP) activates transcription.

catalytic antibodies. Antibody molecules that have been genetically manipulated so that they catalyze reactions involving the antigen.

catalytic center. The polar amino acids in the active site of an enzyme that participate in chemical changes during catalysis.

catalytic constant (k_{cat}). A kinetic constant that is a measure of how rapidly an enzyme can catalyze a reaction when saturated with its

substrate(s). The catalytic constant is equal to the maximum velocity (V_{max}) divided by the total concentration of enzyme ($[E]_{total}$), or the number of moles of substrate converted to product per mole of enzyme active sites per second, under saturating conditions. Also known as the turnover number.

catalytic proficiency. The ratio of the rate constants for a reaction in the presence of enzyme (k_{cat}/K_m) to the rate constant for the chemical reaction in the absence of enzyme.

cataplerotic reaction. A reaction that removes intermediates in a pathway, especially the citric acid cycle (cf., anaplerotic).

cathode. A negatively charged electrode. In electrophoresis, cations move toward the cathode.

cation. An ion with an overall positive charge.

cDNA. *See* complementary DNA.

Central Dogma. The concept that the flow of information from nucleic acid to protein is irreversible. The term is often applied incorrectly to the actual pathway of information flow from DNA to RNA to protein.

ceramide. A molecule that consists of a fatty acid linked to the C-2 amino group of sphingosine by an amide bond. Ceramides are the metabolic precursors of all sphingolipids.

cerebroside. A glycosphingolipid that contains one monosaccharide residue attached via a β-glycosidic linkage to C-1 of a ceramide. Cerebrosides are abundant in nerve tissue and are found in myelin sheaths.

channel. An integral membrane protein with a central aqueous passage, which allows appropriately sized molecules and ions to traverse the membrane in either direction. Also known as a pore.

channeling. *See* metabolite channeling.

chaotropic agent. A substance that enhances the solubility of nonpolar compounds in water by disrupting regularities in hydrogen bonding among water molecules. Concentrated solutions of chaotropic agents, such as urea and guanidinium salts, decrease the hydrophobic effect and are thus effective protein denaturants.

chaperone. A protein that forms complexes with newly synthesized polypeptide chains and assists in their correct folding into biologically functional conformations. Chaperones may also prevent the formation of incorrectly folded intermediates, prevent incorrect aggregation of unassembled protein subunits, assist in translocation of polypeptide chains across membranes, and assist in the assembly and disassembly of large multiprotein structures.

charge-charge interaction. A noncovalent electrostatic interaction between two charged particles.

chelate effect. The phenomenon by which the constant for binding of a ligand having two or more binding sites to a molecule or atom is greater than the constant for binding of separate ligands to the same molecule or atom.

chemiosmotic theory. A theory proposing that a proton concentration gradient established during oxidation of substrates provides the energy to drive processes such as the formation of ATP from ADP and P_i.

chemoautotroph. An autotroph that derives chemical energy by oxidizing inorganic compounds (cf., photoautotroph).

chemoheterotroph. Non-photosynthetic organism that requires organic molecules as a carbon source and derives energy from oxidizing organic molecules.

chemotaxis. A mechanism that couples signal transduction to flagella movement in bacteria causing them to move toward a chemical (positive chemotaxis) or away from a chemical (negative chemotaxis).

chiral atom. An atom with asymmetric substitution that can exist in two different configurations.

chloroplast. A chlorophyll-containing organelle in algae and plant cells that is the site of photosynthesis.

chromatin. A DNA-protein complex in the nuclei of eukaryotic cells.

chromatography. A technique used to separate components of a mixture based on their partitioning between a mobile phase, which can be gas or liquid, and a stationary phase, which is a liquid or solid.

chromosome. A single DNA molecule containing many genes. An organism may have a genome consisting of a single chromosome or many.

chylomicron. A type of plasma lipoprotein that transports triacylglycerols, cholesterol, and cholesteryl esters from the small intestine to the tissues.

citric acid cycle. A metabolic cycle consisting of eight enzyme-catalyzed reactions that completely oxidizes acetyl units to CO_2. The energy released in the oxidation reactions is conserved as reducing power when the coenzymes NAD^1 and ubiquinone (Q) are reduced. Oxidation of one molecule of acetyl CoA by the citric acid cycle generates three molecules of NADH, one molecule of QH_2, and one molecule of GTP or ATP. Also known as the Krebs cycle and the tricarboxylic acid cycle.

clone. One of the identical copies derived from the replication or reproduction of a single molecule, cell, or organism.

cloning. The generation of many identical copies of a molecule, cell, or organism. Cloning sometimes refers to the entire process of constructing and propagating a recombinant DNA molecule.

cloning vector. A DNA molecule that carries a segment of foreign DNA. A cloning vector introduces the foreign DNA into a cell where it can be replicated and sometimes expressed.

coding strand. The strand of DNA within a gene whose nucleotide sequence is identical to that of the RNA produced by transcription (with the replacement of T by U in RNA).

codon. A sequence of three nucleotide residues in mRNA (or DNA) that specifies a particular amino acid according to the genetic code.

coenzyme. An organic molecule required by an enzyme for full activity. Coenzymes can be further classified as cosubstrates or prosthetic groups.

coenzyme A. A large coenyme used in transferring acyl groups.

cofactor. An inorganic ion or organic molecule required by an apoenzyme to convert it to a holoenzyme. There are two types of cofactors: essential ions and coenzymes.

column chromatography. A technique for purifying proteins. *See* affinity chromatography, gel-filtration chromatography, ion-exchange chromatography, HPLC, and affinity chromatography.

competitive inhibition. Reversible inhibition of an enzyme-catalyzed reaction by an inhibitor that prevents substrate binding.

complementary DNA (cDNA). DNA synthesized from an mRNA template by the action of reverse transcriptase.

concerted theory of cooperativity and allosteric regulation. A model of the cooperative binding of ligands to oligomeric proteins. According to the concerted theory, the change in conformation of a protein due to the binding of a substrate or an allosteric modulator shifts the equilibrium of the conformation of the protein between T (a low substrate-affinity conformation) and R (a high substrate-affinity conformation). This theory suggests that all subunits of the protein have the same conformation, either all T or all R. Also known as the symmetry-driven theory.

condensation. A reaction involving the joining of two or more molecules accompanied by the elimination of water, alcohol, or other simple substance.

configuration. A spatial arrangement of atoms that cannot be altered without breaking and re-forming covalent bonds.

conformation. Any three-dimensional structure, or spatial arrangement, of a molecule that results from rotation of functional groups around single bonds. Because there is free rotation around single bonds, a molecule can potentially assume many conformations.

conjugate acid. The product resulting from the gain of a proton by a base.

conjugate base. The product resulting from the loss of a proton by an acid.

consensus sequence. The sequence of nucleotides most commonly found at each position within a region of DNA or RNA.

cooperativity. 1. The phenomenon whereby the binding of one ligand or substrate molecule to a protein influences the affinity of the protein for additional molecules of the same substance. Cooperativity may be positive or negative. **2.** The phenomenon whereby formation

of structure in one part of a macromolecule promotes the formation of structure in the rest of the molecule.

core particle. *See* nucleosome core particle.

corepressor. A ligand that binds to a repressor of a gene causing it to bind DNA and prevent transcription.

Cori cycle. An interorgan metabolic loop that recycles carbon and transports energy from the liver to the peripheral tissues. Glucose is released from the liver and metabolized to produce ATP in other tissues. The resulting lactate is then returned to the liver for conversion back to glucose by gluconeogenesis.

cosubstrate. A coenzyme that is a substrate in an enzyme-catalyzed reaction. A cosubstrate is altered during the course of the reaction and dissociates from the active site of the enzyme. The original form of the cosubstrate can be regenerated in a subsequent enzyme-catalyzed reaction.

cotransport. The coupled transport of two different species of solutes across a membrane, in the same direction (symport) or the opposite direction (antiport), carried out by a transport protein.

coupled reactions. Two metabolic reactions that share a common intermediate.

covalent catalysis. Catalysis in which one substrate, or part of it, forms a covalent bond with the catalyst and then is transferred to a second substrate. Many enzymatic group-transfer reactions proceed by covalent catalysis.

Crassulacean acid metabolism (CAM). A modified sequence of carbon-assimilation reactions used primarily by plants in arid environments to reduce water loss during photosynthesis. In these reactions, CO_2 is taken up at night, resulting in the formation of malate. During the day, malate is decarboxylated, releasing CO_2 for use by the reductive pentose phosphate cycle.

***C*-terminus.** The amino acid residue bearing a free carboxyl group at one end of a peptide chain. Also known as the carboxyl terminus.

cyclic electron transport. A modified sequence of electron transport steps in chloroplasts that operates to provide ATP without the simultaneous formation of NADPH.

cytoplasm. The part of a cell enclosed by the plasma membrane, excluding the nucleus.

cytoskeleton. A network of proteins that contributes to the structure and organization of a eukaryotic cell.

cytosol. The aqueous portion of the cytoplasm minus the subcellular structures.

D arm. The stem-and-loop structure in a tRNA molecule that contains dihydrouridylate (D) residues.

dalton. A unit of mass equal to one atomic mass unit.

dark reactions. The photosynthetic reactions in which NADPH and ATP are used to fix CO_2 to carbohydrate. Also known as the light-independent reactions.

degeneracy. When referring to the genetic code, degeneracy refers to the fact that several different codons specify the same amino acid.

dehydrogenase. An enzyme that catalyzes the removal of hydrogen from a substrate or the oxidation of a substrate. Dehydrogenases are members of the IUBMB class of enzymes known as oxidoreductases.

denaturation. 1. A disruption in the native conformation of a biological macromolecule that results in loss of the biological activity of the macromolecule. **2.** The complete unwinding and separation of complementary strands of DNA.

detergent. An amphipathic molecule consisting of a hydrophobic portion and a hydrophilic end that may be ionic or polar. Detergent molecules can aggregate in aqueous media to form micelles. Also known as a surfactant.

dialysis. A procedure in which low-molecular-weight solutes in a sample are removed by diffusion through a semipermeable barrier and replaced by solutes from the surrounding medium.

diffusion controlled reaction. A reaction that occurs with every collision between reactant molecules. In enzyme-catalyzed reactions, the k_{cat}/K_m ratio approaches a value of $10^8 - 10^9\ M^{-1}\ s^{-1}$.

diploid. Having two sets of chromosomes or two copies of the genome.

dipole. Two equal but opposite charges, separated in space, resulting from the uneven distribution of charge within a molecule or a chemical bond.

direct repair. The removal of DNA damage by proteins that recognize damaged nucleotides and mismatched bases and repair them without cleaving the DNA or excising the base.

distributive enzyme. An enzyme that dissociates from its growing polymeric product after addition of each monomeric unit and must reassociate with the polymer for polymerization to proceed (cf., progressive enzyme).

disulfide bond. A covalent linkage formed by oxidation of the sulfhydryl groups of two cysteine residues. Disulfide bonds are important in stabilizing the three-dimensional structures of some proteins.

domain. A discrete, independent folding unit within the tertiary structure of a protein. Domains are usually combinations of several motifs forming a characteristic fold.

double helix. A nucleic acid conformation in which two antiparallel polynucleotide strands wrap around each other to form a two-stranded helical structure stabilized largely by stacking interactions between adjacent hydrogen-bonded base pairs.

double-reciprocal plot. A plot of the reciprocal of initial velocity versus the reciprocal of substrate concentration for an enzyme-catalyzed reaction. The x and y intercepts indicate the values of the reciprocals of the Michaelis constant and the maximum velocity, respectively. A double-reciprocal plot is a linear transformation of the Michaelis-Menten equation. Also known as a Lineweaver-Burk plot.

E. *See* reduction potential.

E°′. *See* standard reduction potential.

E site. Exit site. The site on a ribosome from which a deaminoacylated tRNA is released during protein synthesis.

Edman degradation. A procedure used to determine the sequence of amino acid residues from a free *N*-terminus of a polypeptide chain. The *N*-terminal residue is chemically modified, cleaved from the chain, and identified by chromatographic procedures, and the rest of the polypeptide is recovered. Multiple reaction cycles allow identification of the new *N*-terminal residue generated by each cleavage step.

effector enzyme. A membrane-associated protein that produces an intracellular second messenger in response to a signal from a transducer.

eicosanoid. An oxygenated derivative of a 20-carbon polyunsaturated fatty acid. Eicosanoids function as short-range messengers in the regulation of various physiological processes.

electromotive force (emf). A measure of the difference between the reduction potentials of the reactions on the two sides of an electrochemical cell (i.e., the voltage difference produced by the reactions).

electrolyte. A molecule such as NaCl that can dissociated to form ions.

electron transport. A set of reactions in which compounds such as NADH and reduced ubiquinone (QH_2) are aerobically oxidized and ATP is generated from ADP and P_i. Membrane-associated electron transport consists of two tightly coupled phenomena: oxidation of substrates by the respiratory electron transport chain, accompanied by the translocation of protons across the inner mitochondrial membrane to generate a proton concentration gradient; and formation of ATP, driven by the flux of protons into the matrix through a channel in ATP synthase.

electrophile. A positively charged or electron-deficient species that is attracted to chemical species that are negatively charged or contain unshared electron pairs (nucleophiles).

electrophoresis. A technique used to separate molecules by their migration in an electric field, primarily on the basis of their net charge.

electrospray mass spectrometry. A technique in mass spectrometry where the target molecule is sprayed into the detector in tiny droplets.

electrostatic interaction. A general term for the electronic interaction between particles. Electrostatic interactions include charge-charge interactions, hydrogen bonds, and van der Waals forces.

elongation factor. A protein that is involved in extending the peptide chain during protein synthesis.

enantiomers. Stereoisomers that are nonsuperimposable mirror images.

endocytosis. The process by which matter is engulfed by a plasma membrane and brought into the cell within a lipid vesicle derived from the membrane.

endonuclease. An enzyme that catalyzes the hydrolysis of phosphodiester linkages at various sites within polynucleotide chains.

endoplasmic reticulum. A membranous network of tubules and sheets continuous with the outer nuclear membrane of eukaryotic cells. Regions of the endoplasmic reticulum coated with ribosomes are called the rough endoplasmic reticulum; regions having no attached ribosomes are known as the smooth endoplasmic reticulum. The endoplasmic reticulum is involved in the sorting and transport of certain proteins and in the synthesis of lipids.

endosomes. Smooth vesicles inside the cell that are receptacles for endocytosed material.

energy-rich compound. A compound whose hydrolysis occurs with a large negative free-energy change (equal to or greater than that for ATP $\rightarrow$ ADP + P_i).

enthalpy (*H*). A thermodynamic state function that describes the heat content of a system.

entropy (*S*). A thermodynamic state function that describes the randomness or disorder of a system.

enzymatic reaction. A reaction catalyzed by a biological catalyst, an enzyme. Enzymatic reactions are 10^3 to 10^{17} times faster than the corresponding uncatalyzed reactions.

enzyme. A biological catalyst, almost always a protein. Some enzymes may require additional cofactors for activity. Virtually all biochemical reactions are catalyzed by specific enzymes.

enzyme assay. A method used to analyze the activity of a sample of an enzyme. Typically, enzymatic activity is measured under selected conditions such that the rate of conversion of substrate to product is proportional to enzyme concentration.

enzyme inhibitor. A compound that binds to an enzyme and interferes with its activity by preventing either the formation of the ES complex or its conversion to E + P.

enzyme-substrate complex (ES). A complex formed when substrate molecules bind noncovalently within the active site of an enzyme.

epimers. Isomers that differ in configuration at only one of several chiral centers.

equilibrium. The state of a system in which the rate of conversion of substrate to product is equal to the rate of conversion of product to substrate. The free-energy change for a reaction or system at equilibrium is zero.

equilibrium constant (K_{eq}). The ratio of the concentrations of products to the concentrations of reactants at equilibrium. The equilibrium constant is related to the standard Gibbs free energy change of reaction.

essential amino acid. An amino acid that cannot be synthesized by an animal and must be obtained in the diet.

essential fatty acid. A fatty acid that cannot be synthesized by an animal and must be obtained in the diet.

essential ion. An ion required as a cofactor for the catalytic activity of certain enzymes. Some essential ions, called activator ions, are reversibly bound to enzymes and often participate in the binding of substrates, whereas tightly bound metal ions frequently participate directly in catalytic reactions.

eukaryote. An organism whose cells generally possess a nucleus and internal membranes (cf., prokaryote).

excision repair. The reversal of DNA damage by excision-repair endonucleases. Gross lesions that alter the structure of the DNA helix are repaired by cleavage on each side of the lesion and removal of the damaged DNA. The resulting single-stranded gap is filled by DNA polymerase and sealed by DNA ligase.

exocytosis. The process by which material destined for secretion from a cell is enclosed in lipid vesicles that are transported to and fuse with the plasma membrane, releasing the material into the extracellular space.

exon. A nucleotide sequence that is present in the primary RNA transcript and in the mature RNA molecule. The term exon also refers to the region of the gene that corresponds to a sequence present in the mature RNA (cf., intron).

exonuclease. An enzyme that catalyzes the sequential hydrolysis of phosphodiester linkages from one end of a polynucleotide chain.

extrinsic membrane protein. *See* peripheral membrane protein.

facilitated diffusion. *See* passive transport.

facultative anaerobe. An organism that can survive in the presence or absence of oxygen.

fatty acid. A long chain aliphatic hydrocarbon with a single carboxyl group at one end. Fatty acids are the simplest type of lipid and are components of many more complex lipids, including triacylglycerols, glycerophospholipids, sphingolipids, and waxes.

feedback inhibition. Inhibition of an enzyme that catalyzes an early step in a metabolic pathway by an end product of the same pathway.

feed-forward activation. Activation of an enzyme in a metabolic pathway by a metabolite produced earlier in the pathway.

fermentation. The anaerobic catabolism of metabolites for energy production. In alcoholic fermentation, pyruvate is converted to ethanol and carbon dioxide.

fibrous proteins. A major class of water-insoluble proteins that associate to form long fibers. Many fibrous proteins are physically tough and provide mechanical support to individual cells or entire organisms.

first-order reaction. A reaction whose rate is directly proportional to the concentration of only one reactant.

Fischer projection. A two-dimensional representation of the three-dimensional structures of sugars and related compounds. In a Fischer projection, the carbon skeleton is drawn vertically, with C-1 at the top. At a chiral center, horizontal bonds extend toward the viewer and vertical bonds extend away from the viewer.

fluid mosaic model. A model proposed for the structure of biological membranes. In this model, the membrane is depicted as a dynamic structure in which lipids and membrane proteins (both integral and peripheral) rotate and undergo lateral diffusion.

fluorescence. A form of luminescence in which visible radiation is emitted from a molecule as it passes from a higher to a lower electronic state.

flux. The flow of material through a metabolic pathway. Flux depends on the supply of substrates, the removal of products, and the catalytic capabilities of the enzymes involved in the pathway.

fold. A combination of secondary structures that form the core of a protein domain. Many different folds have been characterized.

frameshift mutation. An alteration in DNA caused by the insertion or deletion of a number of nucleotides not divisible by three. A frameshift mutation changes the reading frame of the corresponding mRNA molecule and affects translation of all codons downstream of the mutation.

free energy change. *See* Gibbs free energy change.

free radical. A molecule or atom with an unpaired electron.

furanose. A monosaccharide structure that forms a five-membered ring as a result of intramolecular hemiacetal formation.

G protein. A protein that binds guanine nucleotides.

ΔG. *See* Gibbs free energy change.

$\Delta G^{\circ\prime}$. *See* standard Gibbs free energy change.

ganglioside. A glycosphingolipid in which oligosaccharide chains containing *N*-acetylneuraminic acid are attached to a ceramide. Gangliosides are present on cell surfaces and provide cells with distinguishing surface markers that may serve in cellular recognition and cell-to-cell communication.

gas chromatography. A chromatographic technique used to separate components of a mixture based on their partitioning between the gas phase and a stationary phase, which can be a liquid or solid.

gel-filtration chromatography. A chromatographic technique used to separate a mixture of proteins or other macromolecules in solution based on molecular size, using a matrix of porous beads. Also known as molecular-exclusion chromatography.

gene. Loosely defined as a segment of DNA that is transcribed. In some cases, the term gene may also be used to refer to a segment of DNA that encodes a functional protein or corresponds to a mature RNA molecule.

genetic code. The correspondence between a particular three nucleotide codon and the amino acid it specifies. The standard genetic code of 64 codons is used by almost all organisms. The genetic code is used to translate the sequence of nucleotides in mRNA into protein.

genetic recombination. The exchange or transfer of DNA from one molecule of DNA to another (cf., homologous recombination).

genome. One complete set of the genetic information in an organism. It may be a single chromosome or a set of chromosomes (haploid). Mitochondria and chloroplasts have genomes separate from that in the nucleus of eukaryotic cells.

Gibbs free energy change (ΔG). A thermodynamic quantity that defines the equilibrium condition in terms of the changes in enthalpy (H) and entropy (S) of a system at constant pressure. $\Delta G = \Delta H - T\Delta S$, where T is absolute temperature. Free energy is a measure of the energy available within a system to do work.

globular proteins. A major class of proteins, many of which are water soluble. Globular proteins are compact and roughly spherical, containing tightly folded polypeptide chains. Typically, globular proteins include indentations, or clefts that specifically recognize and transiently bind other compounds.

glucogenic compound. A compound, such as an amino acid, that can be used for gluconeogenesis in animals.

gluconeogenesis. A pathway for synthesis of glucose from a noncarbohydrate precursor. Gluconeogenesis from pyruvate involves the seven near-equilibrium reactions of glycolysis traversed in the reverse direction. The three metabolically irreversible reactions of glycolysis are bypassed by four enzymatic reactions that do not occur in glycolysis.

glucoside. A glycoside where the anomeric carbon atom is from glucose.

glycan. A general term for an oligosaccharide or a polysaccharide. A homoglycan is a polymer of identical monosaccharide residues; a heteroglycan is a polymer of different monosaccharide residues.

glycerophospholipid. A lipid consisting of two fatty acyl groups bound to C-1 and C-2 of glycerol 3-phosphate and, in most cases, a polar substituent attached to the phosphate moiety. Glycerophospholipids are major components of biological membranes.

glycoconjugate. A carbohydrate derivative in which one or more carbohydrate chains are covalently linked to a peptide chain, protein, or lipid.

glycoforms. Glycoproteins containing identical amino acid sequences but different oligosaccharide-chain compositions.

glycogen. A branched homopolymer of glucose residues joined by α-(1 → 4) linkages with α-(1 → 6) linkages at branch points. Glycogen is a storage polysaccharide in animals and bacteria.

glycolysis. A catabolic pathway consisting of 10 enzyme-catalyzed reactions by which one molecule of glucose is converted to two molecules of pyruvate. In the process, two molecules of ATP are formed from ADP + P_i, and two molecules of NAD^1 are reduced to NADH.

glycoprotein. A protein that contains covalently bound carbohydrate residues.

glycosaminoglycan. An unbranched polysaccharide of repeating disaccharide units. One component of the disaccharide is an amino sugar; the other component is usually a uronic acid.

glycoside. A molecule containing a carbohydrate in which the hydroxyl group of the anomeric carbon has been replaced through condensation with an alcohol, an amine, or a thiol.

glycosidic bond. Acetal linkage formed by condensation of the anomeric carbon atom of a saccharide with a hydroxyl, amino, or thiol group of another molecule. The most commonly encountered glycosidic bonds are formed between the anomeric carbon of one sugar and a hydroxyl group of another sugar. Nucleosidic bonds are *N*-linked glycosidic bonds.

glycosphingolipid. A lipid containing sphingosine and carbohydrate moieties.

glycosylation. *See* protein glycosylation.

glyoxylate cycle. A variation of the citric acid cycle in certain plants, bacteria, and yeast that allows net production of glucose from acetyl CoA via oxaloacetate. The glyoxylate cycle bypasses the two CO_2[2] producing steps of the citric acid cycle.

glyoxysome. An organelle that contains specialized enzymes for the glyoxylate cycle.

Golgi apparatus. A complex of flattened, fluid-filled membranous sacs in eukaryotic cells, often found in proximity to the endoplasmic reticulum. The Golgi apparatus is involved in the modification, sorting, and targeting of proteins.

granum. A stack of flattened vesicles formed from the thylakoid membrane in chloroplasts.

group transfer potential. *See* photsphoryl group transfer potential.

group transfer reaction. A reaction in which a substituent or functional group is transferred from one substrate to another.

H. *See* enthalpy.

hairpin. 1. A secondary structure adopted by single-stranded polynucleotides that arises when short regions fold back on themselves and hydrogen bonds form between complementary bases. Also known as a stem-loop. **2.** A tight turn connecting two consecutive β strands of a polypeptide.

haploid. Having one set of chromosomes or one copy of the genome (cf., diploid).

high energy molecule. *See* energy-rich compound.

Haworth projection. A representation in which a cyclic sugar molecule is depicted as a flat ring that is projected perpendicular to the plane of the page. Heavy lines represent the part of the molecule that extends toward the viewer.

HDL. *See* high density lipoprotein.

heat of vaporization. The amount of heat required to evaporate 1 gram of a liquid.

heat shock protein. A protein whose synthesis is increased in response to stresses such as high temperature. Many heat shock proteins are chaperones that are also expressed in the absence of stress.

helicase. An enzyme that is involved in unwinding DNA.

hemiacetal. The product formed when an alcohol reacts with an aldehyde.

hemiketal. The product formed when an alcohol reacts with a ketone.

Henderson-Hasselbalch equation. An equation that describes the pH of a solution of a weak acid or a weak base in terms of the pK_a and the concentrations of the proton donor and proton acceptor forms.

heterochromatin. Regions of chromatin that are highly condensed.

heterocyclic molecule. A molecule that contains a ring structure made up of more than one type of atom.

heteroglycan (heteropolysaccharide). A carbohydrate polymer whose residues consist of two or more different types of monosaccharide.

heterotroph. An organism that requires at least one organic nutrient, such as glucose, as a carbon source.

high density lipoprotein (HDL). A type of plasma lipoprotein that is enriched in protein and transports cholesterol and cholesteryl esters from tissues to the liver.

high-performance liquid chromatography (HPLC). A chromatographic technique used to separate components of a mixture by dissolving the mixture in a liquid solvent and forcing it to flow through a chromatographic column under high pressure.

histones. A class of proteins that bind to DNA to form chromatin. The nuclei of eukaryotic cells contain five histones, known as H1, H2A, H2B, H3, and H4.

Holliday junction. The region of strand crossover resulting from recombination between two molecules of homologous double-stranded DNA.

homoglycan (homopolysaccharide). A carbohydrate polymer whose residues consist of a single type of monosaccharide.

homologous. Referring to genes or proteins that descend from a common ancestor.

homologous recombination. Recombination between molecules of DNA that have closely related sequences (i.e., they are homologous). This is the standard form of recombination that occurs between chromosomes in eukaryotic cells.

homology. The similarity of genes or proteins as a result of evolution from a common ancestor.

hormone response element. A DNA sequence that binds a transcriptional activator consisting of a steroid hormone receptor complex.

housekeeping genes. Genes that encode proteins or RNA molecules that are essential for the normal activities of all living cells.

HPLC. *See* high-performance liquid chromatography.

hydration. A state in which a molecule or ion is surrounded by water.

hydrogen bond. A weak electrostatic interaction the formed when a hydrogen atom bonded covalently to a strongly electronegative atom is partially shared by interacting with electron pair of another electronegative atom.

hydrolase. An enzyme that catalyzes the hydrolytic cleavage of its substrate(s) (i.e., hydrolysis).

hydropathy. A measure of the hydrophobicity of amino acid side chains. The more positive the hydropathy value, the greater the hydrophobicity.

hydrophilic. "Water loving"—describing molecules that interact favorably with water.

hydrophilicity. The degree to which a compound or functional group interacts with water or is preferentially soluble in water.

hydrophobic. "Water fearing"—describing molecules that do not interact favorably with water and are much less soluble than hydrophilic molecules.

hydrophobic effect. The exclusion of hydrophobic groups or molecules by water. The hydrophobic effect appears to depend on the increase in entropy of solvent water molecules that are released from an ordered arrangement around the hydrophobic group.

hydrophobic interaction. A weak, noncovalent interaction between nonpolar molecules or substituents that results from the strong association of water molecules with one another. Such association leads to the shielding or exclusion of nonpolar molecules from an aqueous environment.

hydrophobicity. The degree to which a compound or functional group that is soluble in nonpolar solvents is insoluble or only sparingly soluble in water.

IDL. *See* intermediate density lipoprotein.

induced fit. Activation of an enzyme by a substrate-initiated conformational change.

inducer. A ligand that binds to and inactivates a repressor thereby increasing the transcription of the gene controlled by the repressor.

inhibition constant (K_i). The equilibrium constant for the dissociation of an inhibitor from an enzyme-inhibitor complex.

inhibitor. A compound that binds to an enzyme and inhibits its activity.

initial velocity (v_0). The rate of conversion of substrate to product in the early stages of an enzymatic reaction, before appreciable product has been formed.

initiation codon. A codon that specifies the initiation site for protein synthesis. The methionine codon (AUG) is the most common initiation codon.

initiation factor. *See* translation initiation factor.

initiator tRNA. The tRNA molecule that is used exclusively at initiation codons. The initiator tRNA is usually a specific methionyl-tRNA.

integral membrane protein. A membrane protein that penetrates the hydrophobic core of the lipid bilayer and usually spans the bilayer completely. Also known as an intrinsic membrane protein.

intercalating agent. A compound containing a planar ring structure that can fit between the stacked base pairs of DNA. Intercalating agents distort the DNA structure, partially unwinding the double helix.

intermediary metabolism. The metabolic reactions by which the small molecules of cells are interconverted.

intermediate density lipoprotein (IDL). A type of plasma lipoprotein that is formed during the breakdown of VLDLs.

intermediate filament. A structure composed of different protein subunits, found in the cytoplasm of most eukaryotic cells. Intermediate filaments are components of the cytoskeletal network.

intron. An internal nucleotide sequence that is removed from the primary RNA transcript during processing. The term intron also refers to the region of the gene that corresponds to the corresponding RNA intron (cf., exon).

inverted repeat. A sequence of nucleotides that is repeated in the opposite orientation within the same polynucleotide strand. An inverted repeat in double-stranded DNA can give rise to a cruciform structure.

ion pair. An electrostatic interaction between ionic groups of opposite charge within the interior of a macromolecule such as a globular protein.

ion product for water (K_w). The product of the concentrations of hydronium ions and hydroxide ions in an aqueous solution, equal to $1.0 \times 10^{-14}\ M^2$.

ion-exchange chromatography. A chromatographic technique used to separate a mixture of ionic species in solution, using a charged matrix. In anion-exchange chromatography, a positively charged matrix binds negatively charged solutes, and in cation-exchange chromatography, a negatively charged matrix binds positively charged solutes. The bound species can be serially eluted from the matrix by gradually changing the pH or increasing the salt concentration in the solvent.

ionophore. A compound that facilitates the diffusion of ions across bilayers and membranes by serving as a mobile ion carrier or by forming a channel for ion passage.

irreversible enzyme inhibition. A form of enzyme inhibition where the inhibitor binds covalently to the enzyme.

isoacceptor tRNA molecules. Different tRNA molecules that bind the same amino acid.

isoelectric focusing. A modified form of electrophoresis that uses buffers to create a pH gradient within a polyacrylamide gel. Each protein migrates to its isoelectric point (pI), that is, the pH in the gradient at which it no longer carries a net positive or negative charge.

isoelectric point (pI). The pH at which a zwitterionic molecule does not migrate in an electric field because its net charge is zero.

isoenzymes. *See* isozymes.

isomerase. An enzyme that catalyzes an isomerization reaction, a change in geometry or structure within one molecule.

isoprene. A branched, unsaturated five-carbon molecule that forms the basic structural unit of all isoprenoids, including the steroids and lipid vitamins.

isoprenoid. A lipid that is structurally related to isoprene.

isozymes. Different proteins from a single biological species that catalyze the same reaction. Also known as isoenzymes.

junk DNA. Regions of the genome with no known function.

K_a. *See* acid dissociation constant.

kb. *See* kilobase pair.

k_{cat}. *See* catalytic constant.

k_{cat}/K_m. The second-order rate constant for conversion of enzyme and substrate to enzyme and product at low substrate concentrations. The ratio of k_{cat} to K_m, when used to compare several substrates, is called the specificity constant.

K_{eq}. *See* equilibrium constant.

ketogenesis. The pathway that synthesizes ketone bodies from acetyl CoA in the mitochondrial matrix in mammals.

ketogenic compound. A compound, such as an amino acid, that can be degraded to form acetyl CoA and can thereby contribute to the synthesis of fatty acids or ketone bodies.

ketone bodies. Small molecules that are synthesized in the liver from acetyl CoA. During starvation, the ketone bodies β-hydroxybutyrate and acetoacetate become major metabolic fuels.

ketoses. A class of monosaccharides in which the most oxidized carbon atom, usually C-2, is ketonic.

K_j. *See* inhibition constant.

kilobase pair (kb). A unit of length of double-stranded DNA, equivalent to 1000 base pairs.

kinase. An enzyme that catalyzes transfer of a phosphoryl group to an acceptor molecule. A protein kinase catalyzes the phosphorylation of protein substrates. Kinases are also known as phosphotransferases.

kinetic mechanism. A scheme used to describe the sequence of steps in a multisubstrate enzyme-catalyzed reaction.

kinetic order. The sum of the exponents in a rate equation, which reflects how many molecules are reacting in the slowest step of the reaction. Also known as reaction order.

K_m. *See* Michaelis constant.

Krebs cycle. *See* citric acid cycle.

K_w. *See* ion product of water.

lagging strand. The newly synthesized DNA strand formed by discontinuous $5' \rightarrow 3'$ polymerization in the direction opposite replication fork movement.

lateral diffusion. The rapid motion of lipid or protein molecules within the plane of one leaflet of a lipid bilayer.

LDL. *See* low density lipoprotein.

leader peptide. The peptide encoded by a portion of the leader region of certain regulated operons. Synthesis of a leader peptide is the basis for regulating transcription of the entire operon by the mechanism of attenuation.

leader region. The sequence of nucleotides that lie between the transcription start site and the first coding region of an operon.

leading strand. The newly synthesized DNA strand formed by continuous $5' \rightarrow 3'$ polymerization in the same direction as replication fork movement.

leaflet. One layer of a lipid bilayer.

lectin. A plant protein that binds specific saccharides in glycoproteins.

leucine zipper. A structural motif found in DNA-binding proteins and other proteins. The zipper is formed when the hydrophobic faces (frequently containing leucine residues) of two amphipathic α-helices from the same or different polypeptide chains interact to form a coiled-coil structure.

LHC. *See* light-harvesting complex.

ligand. A molecule, group, or ion that binds noncovalently to another molecule or atom.

ligand-gated ion channel. A membrane ion channel that opens or closes in response to binding of a specific ligand.

ligase. An enzyme that catalyzes the joining, or ligation, of two substrates. Ligation reactions require the input of the chemical potential energy of a nucleoside triphosphate such as ATP. Ligases are commonly referred to as synthetases.

light reactions. The photosynthetic reactions in which protons derived from water are used in the chemiosmotic synthesis of ATP from ADP + P_i and a hydride ion from water reduces to NADPH. Also known as the light-dependent reactions.

light-harvesting complex (LHC). A large pigment complex in the thylakoid membrane that aids a photosystem in gathering light.

limit dextrin. A branched oligosaccharide derived from a glucose polysaccharide by the hydrolytic action of amylase or the phosphorolytic action of glycogen phosphorylase or starch phosphorylase. Limit dextrins are resistant to further degradation catalyzed by amylase or phosphorylase. Limit dextrins can be further degraded only after hydrolysis of the α-$(1 \rightarrow 6)$ linkages.

Lineweaver-Burk plot. *See* double-reciprocal plot.

linker DNA. The stretch of DNA (approximately 54 base pairs) between two adjacent nucleosome core particles.

lipase. An enzyme that catalyzes the hydrolysis of triacylglycerols.

lipid. A water-insoluble (or sparingly soluble) organic compound found in biological systems, which can be extracted by using relatively nonpolar organic solvents.

lipid bilayer. A double layer of lipids in which the hydrophobic tails associate with one another in the interior of the bilayer and the polar head groups face outward into the aqueous environment. Lipid bilayers are the structural basis of biological membranes.

lipid raft. A patch of membrane rich in cholesterol and sphingolipid.

lipid vitamin. A polyprenyl compound composed primarily of a long hydrocarbon chain or fused ring. Unlike water-soluble vitamins, lipid vitamins can be stored by animals. Lipid vitamins include vitamins A, D, E, and K.

lipid anchored membrane protein. A membrane protein that is tethered to a membrane through covalent linkage to a lipid molecule.

lipopolysaccharide. A macromolecule composed of lipid A (a disaccharide of phosphorylated glucosamine residues with attached fatty acids) and a polysaccharide. Lipopolysaccharides are found in the outer membrane of gram-negative bacteria. These compounds are released from bacteria undergoing lysis and are toxic to humans and other animals. Also known as an endotoxin.

lipoprotein. A macromolecular assembly of lipid and protein molecules with a hydrophobic core and a hydrophilic surface. Lipids are transported via lipoproteins.

liposome. A synthetic vesicle composed of a phospholipid bilayer that encloses an aqueous compartment.

loop. A nonrepetitive polypeptide region that connects secondary structures within a protein molecule and provides directional changes necessary for a globular protein to attain its compact shape. Loops contain from 2 to 16 residues. Short loops of up to 5 residues are often called turns.

low density lipoprotein (LDL). A type of plasma lipoprotein that is formed during the breakdown of IDLs and is enriched in cholesterol and cholesteryl esters.

lumen. The aqueous space enclosed by a biological membrane, such as the membrane of the endoplasmic reticulum or the thylakoid membrane.

lyase. An enzyme that catalyzes a nonhydrolytic or nonoxidative elimination reaction, or lysis, of a substrate, with the generation of a double bond. In the reverse direction, a lyase catalyzes addition of one substrate to a double bond of a second substrate.

lysophosphoglyceride. An amphipathic lipid that is produced when one of the two fatty acyl moieties of a glycerophospholipid is hydrolytically removed. Low concentrations of lysophosphoglycerides are metabolic intermediates, whereas high concentrations disrupt membranes, causing cells to lyse.

lysosome. A specialized digestive organelle in eukaryotic cells. Lysosomes contain a variety of enzymes that catalyze the breakdown of cellular biopolymers, such as proteins, nucleic acids, and polysaccharides, and the digestion of large particles, such as some bacteria ingested by the cell.

major groove. The wide groove on the surface of a DNA double helix created by the stacking of base pairs and the resulting twist in the sugar-phosphate backbones.

MALDI. *See* matrix-assisted laser desorption ionization.

mass action ratio (Q). The ratio of the concentrations of products to the concentrations of reactants of a reaction.

mass spectrometry. A technique that determines the mass of a molecule.

matrix. *See* mitochondrial matrix.

matrix-assisted laser desorption ionization (MALDI). A technique in mass spectrometry where the target molecule is released from a solid matrix by a laser beam.

maximum velocity (V_{max}). The initial velocity of a reaction when the enzyme is saturated with substrate, that is, when all the enzyme is in the form of an enzyme-substrate complex.

melting curve. A plot of the change in absorbance versus temperature for a DNA molecule. The change in absorbance indicates unfolding of the double helix.

melting point (T_m). The midpoint of the temperature range in which double-stranded DNA is converted to single-stranded DNA or a protein is converted from its native form to the denatured state.

membrane. A lipid bilayer containing associated proteins that serves to delineate and compartmentalize cells or organelles. Biological membranes are also the site of many important

biochemical processes related to energy transduction and intracellular signaling.

membrane-associated electron transport. *See* electron transport.

membrane potential ($\Delta\psi$). The charge separation across a membrane that results from differences in ionic concentrations on the two sides of the membrane.

messenger ribonucleic acid. *See* mRNA.

metabolic fuel. A small compound that can be catabolized to release energy. In multicellular organisms, metabolic fuels may be transported between tissues.

metabolically irreversible reaction. A reaction in which the value of the mass action ratio is two or more orders of magnitude smaller than the value of the equilibrium constant. The Gibbs free energy change for such a reaction is a large negative number; thus, the reaction is essentially irreversible.

metabolism. The sum total of biochemical reactions carried out by an organism.

metabolite. An intermediate in the synthesis or degradation of biopolymers and their component units.

metabolite channeling. Transfer of the product of one reaction of a multifunctional enzyme or a multienzyme complex directly to the next active site or enzyme without entering the bulk solvent. Channeling increases the rate of a reaction pathway by decreasing the transit time for an intermediate to reach the next enzyme and by producing high local concentrations of the intermediate.

metalloenzyme. An enzyme that contains one or more firmly bound metal ions. In some cases, such metal ions constitute part of the active site of the enzyme and are active participants in catalysis.

micelle. An aggregation of amphipathic molecules in which the hydrophilic portions of the molecules project into the aqueous environment and the hydrophobic portions associated with one another in the interior of the structure to minimize contact with water molecules.

Michaelis constant (K_m). The concentration of substrate that results in an initial velocity (v_0) equal to one-half the maximum velocity (V_{max}) for a given reaction.

Michaelis-Menten equation. A rate equation relating the initial velocity (v_0) of an enzymatic reaction to the substrate concentration ([S]), the maximum velocity (V_{max}), and the Michaelis constant (K_m).

microfilament. *See* actin filament.

microtubule. A protein filament composed of a and b tubulin heterodimers. Microtubules are components of the cytoskeletal network and can form structures capable of directed movement.

minor groove. The narrow groove on the surface of a DNA double helix created by the stacking of base pairs and the resulting twist in the sugar-phosphate backbones.

mismatch repair. Restoration of the normal nucleotide sequence in a DNA molecule containing mismatched bases. In mismatch repair, the correct strand is recognized, a portion of the incorrect strand is excised, and correctly base-paired, double-stranded DNA is synthesized by the actions of DNA polymerase and DNA ligase.

missense mutation. An alteration in DNA that involves the substitution of one nucleotide for another, resulting in a change in the amino acid specified by that codon.

mitochondrial matrix. The gel-like phase enclosed by the inner membrane of the mitochondrion. The mitochondrial matrix contains many enzymes involved in aerobic energy metabolism.

mitochondrion. An organelle that is the main site of oxidative energy metabolism in most eukaryotic cells. Mitochondria contain an outer and an inner membrane, the latter characteristically folded into cristae.

mixed inhibition. A form of enzyme inhibition where both K_m and V_{max} are affected.

molar mass. The weight in grams of one mole of a compound.

molecular chaperone. *See* chaperone.

molecular crowding. The decrease in diffusion rate that occurs when molecules collide with each other.

molecular weight. *See* relative molecular mass.

monocistronic mRNA. An mRNA molecule that encodes only a single polypeptide. Most eukaryotic mRNA molecules are monocistronic.

monomer. 1. A small compound that becomes a residue when polymerized with other monomers. **2.** A single subunit of a multisubunit protein.

monosaccharide. A simple sugar of three or more carbon atoms with the empirical formula $(CH_2O)_n$.

monounsaturated fatty acid. An unsaturated fatty acid with a single carbon-carbon double bond.

motif. A combination of secondary structure that appears in a number of different proteins. Also known as supersecondary structure.

M_r. *See* relative molecular mass.

mRNA. A class of RNA molecules that serve as templates for protein synthesis.

mRNA precursor. A class of RNA molecules synthesized by eukaryotic RNA polymerase II. mRNA precursors are processed posttranscriptionally to produce mature messenger RNA.

mucin. A high-molecular-weight *O*-linked glycoprotein containing as much as 80% carbohydrate by mass. Mucins are extended, negatively charged molecules that contribute to the viscosity of mucus, the fluid found on the surfaces of the gastrointestinal, genitourinary, and respiratory tracts.

multienzyme complex. An oligomeric protein that catalyzes several metabolic reactions.

mutagen. An agent that can cause DNA damage.

mutation. A heritable change in the sequence of nucleotides in DNA that causes a permanent alteration of genetic information.

near-equilibrium reaction. A reaction in which the value of the mass action ratio is close to the value of the equilibrium constant. The Gibbs free energy change for such a reaction is small; thus, the reaction is reversible.

Nernst equation. An equation that relates the observed change in reduction potential (ΔE) to the change in standard reduction potential ($\Delta E^{\circ\prime}$) of a reaction.

neutral phospholipids. Glycerophospholipids, such as phosphatidyl choline, having no net charge.

neutral solution. An aqueous solution that has a pH value of 7.0.

nick translation. The process in which DNA polymerase binds to a gap between the 3′ end of a nascent DNA chain and the 5′ end of the next RNA primer, catalyzes hydrolytic removal of ribonucleotides using $5' \rightarrow 3'$ exonuclease activity, and replaces them with deoxyribonucleotides using $5' \rightarrow 3'$ polymerase activity.

nitrogen cycle. The flow of nitrogen from N_2 to nitrogen oxides ($NO_2^{\ominus}$ and $NO_3^{\ominus}$) ammonia, nitrogenous biomolecules, and back to N_2.

nitrogen fixation. The reduction of atmospheric nitrogen to ammonia. Biological nitrogen fixation occurs in only a few species of bacteria and algae.

***N*-linked oligosaccharide.** An oligosaccharide chain attached to a protein through covalent bonds to the amide nitrogen atom of side chain of asparagine residues. The oligosaccharide chains of *N*-linked glycoproteins contain a core pentasaccharide of two *N*-acetylglucosamine residues and three mannose residues.

NMR spectroscopy. *See* nuclear magnetic resonance spectroscopy.

noncompetitive inhibition. Inhibition of an enzyme-catalyzed reaction by a reversible inhibitor that binds to either the enzyme or the enzyme- substrate complex.

nonessential amino acid. An amino acid that an animal can produce in sufficient quantity to meet metabolic needs.

nonhomologous recombination. Recombination between unrelated sequences that do not share significant sequence similarity.

nonrepetitive structure. An element of protein structure in which consecutive residues do not have a single repeating conformation.

nonsense mutation. An alteration in DNA that involves the substitution of one nucleotide for another, changing a codon that specifies an amino acid to a termination

codon. A nonsense mutation results in premature termination of a protein's synthesis.

***N*-terminus.** The amino acid residue bearing a free α-amino group at one end of a peptide chain. In some proteins, the *N*-terminus is blocked by acylation. The *N*-terminal residue is usually assigned the residue number 1. Also known as the amino terminus.

nuclear envelope. The double membrane that surrounds the nucleus and contains protein-lined nuclear pore complexes that regulate the import and export of material to and from the nucleus. The outer membrane of the nuclear envelope is continuous with the endoplasmic reticulum; the inner membrane is lined with filamentous proteins, constituting the nuclear lamina.

nuclear magnetic resonance spectroscopy (NMR spectroscopy). A technique used to study the structures of molecules in solution. In nuclear magnetic resonance spectroscopy, the absorption of electromagnetic radiation by molecules in magnetic fields of varying frequencies is used to determine the spin states of certain atomic nuclei.

nuclease. An enzyme that catalyzes hydrolysis of the phosphodiester linkages of a polynucleotide chain. Nucleases can be classified as endonucleases and exonucleases.

nucleic acid. A polymer composed of nucleotide residues linked in a linear sequence by 3′–5′ phosphodiester linkages. DNA and RNA are nucleic acids composed of deoxyribonucleotide residues and ribonucleotide residues, respectively.

nucleoid region. The region within a prokaryotic cell that contains the chromosome.

nucleolus. The region of the eukaryotic nucleus where rRNA transcripts are processed and ribosomes are assembled.

nucleophile. An electron-rich species that is negatively charged or contains unshared electron pairs and is attracted to chemical species that are positively charged or electron-deficient (electrophiles).

nucleophilic substitution. A reaction in which one nucleophile (e.g., $Y^{\ominus}$) displaces another (e.g., $X^{\ominus}$).

nucleoside. A purine or pyrimidine *N*-glycoside of ribose or deoxyribose.

nucleosome. A DNA-protein complex that forms the fundamental unit of chromatin. A nucleosome consists of a nucleosome core particle (approximately 146 base pairs of DNA plus a histone octamer), linker DNA (approximately 54 base pairs), and histone H1 (which binds the core particle and linker DNA).

nucleosome core particle. A DNA-protein complex composed of approximately 146 base pairs of DNA wrapped around an octamer of histones (two each of H2A, H2B, H3, and H4).

nucleotide. The phosphate ester of a nucleoside, consisting of a nitrogenous base linked to a pentose phosphate. Nucleotides are the monomeric units of nucleic acids.

nucleus. An organelle that contains the principal genetic material of eukaryotic cells and functions as the major site of RNA synthesis and processing.

obligate aerobe. An organism that requires the presence of oxygen for survival.

obligate anaerobe. An organism that requires an oxygen-free environment for survival.

Okazaki fragments. Relatively short strands of DNA that are produced during discontinuous synthesis of the lagging strand of DNA.

oligomer. A multisubunit molecule whose arrangement of subunits always has a defined stoichiometry and almost always displays symmetry.

oligonucleotide. A polymer of several (up to about 20) nucleotide residues linked by phosphodiester bonds.

oligopeptide. A polymer of several (up to about 20) amino acid residues linked by peptide bonds.

oligosaccharide. A polymer of 2 to about 20 monosaccharide residues linked by glycosidic bonds.

oligosaccharide processing. The enzyme-catalyzed addition and removal of saccharide residues during the maturation of a glycoprotein.

***O*-linked oligosaccharide.** An oligosaccharide attached to a protein through a covalent bond to the hydroxyl oxygen atom of a serine or threonine residue.

open reading frame. A stretch of nucleotide triplets that contains no termination codons. Protein-encoding regions are examples of open reading frames.

operator. A DNA sequence to which a specific repressor protein binds, thereby blocking transcription of a gene or operon.

operon. A bacterial transcriptional unit consisting of several different coding regions cotranscribed from one promoter.

ordered sequential reaction. A reaction in which both the binding of substrates to an enzyme and the release of products from the enzyme follow an obligatory order.

organelle. Any specialized membrane-bounded structure within a eukaryotic cell. Organelles are uniquely organized to perform specific functions.

origin of replication. A DNA sequence at which replication is initiated.

osmosis. The movement of solvent molecules from a less concentrated solution to an adjacent, more concentrated solution.

osmotic pressure. The pressure required to prevent the flow of solvent from a less concentrated solution to a more concentrated solution.

oxidase. An enzyme that catalyzes an oxidation-reduction reaction in which O_2 is the electron acceptor. Oxidases are members of the IUBMB class of enzymes known as oxidoreductases.

oxidation. The loss of electrons from a substance through transfer to another substance (the oxidizing agent). Oxidations can take several forms, including the addition of oxygen to a compound, the removal of hydrogen from a compound to create a double bond, or an increase in the valence of a metal ion.

oxidative phosphorylation. *See* electron transport.

oxidizing agent. A substance that accepts electrons in an oxidation-reduction reaction and thereby becomes reduced.

oxidoreductase. An enzyme that catalyzes an oxidation-reduction reaction. Some oxidoreductases are known as dehydrogenases, oxidases, peroxidases, oxygenases, or reductases.

oxygenation. The reversible binding of oxygen to a macromolecule.

Δp. *See* protonmotive force.

PAGE. *See* polyacrylamide gel electrophoresis.

passive transport. The process by which a solute specifically binds to a transport protein and is transported across a membrane, moving with the solute concentration gradient. Passive transport occurs without the expenditure of energy. Also known as facilitated diffusion.

Pasteur effect. The slowing of glycolysis in the presence of oxygen.

pathway. A sequence of metabolic reactions.

pause site. A region of a gene where transcription slows. Pausing is exaggerated at palindromic sequences, where newly synthesized RNA can form a hairpin structure.

PCR. *See* polymerase chain reaction.

pentose phosphate pathway. A pathway by which glucose 6-phosphate is metabolized to generate NADPH and ribose 5-phosphate. In the oxidative stage of the pathway, glucose 6-phosphate is converted to ribulose 5-phosphate and CO_2 rating two molecules of NADPH. In the nonoxidative stage, ribulose 5-phosphate can be isomerized to ribose 5-phosphate or converted to intermediates of glycolysis. Also known as the hexose monophosphate shunt.

peptide. Two or more amino acids covalently joined in a linear sequence by peptide bonds.

peptide bond. The covalent secondary amide linkage that joins the carbonyl group of one amino acid residue to the amino nitrogen of another in peptides and proteins.

peptide group. The nitrogen and carbon atoms involved in a peptide bond and their four substituents: the carbonyl oxygen atom, the amide hydrogen atom, and the two adjacent α-carbon atoms.

peptidoglycan. A macromolecule containing a heteroglycan chain of alternating *N*-acetylglucosamine and *N*-acetylmuramic acid cross-linked to peptides of varied composition. Peptidoglycans are the major components of the cell walls of many bacteria.

peptidyl site. *See* P site.

peptidyl transferase. The enzymatic activity responsible for the formation of a peptide bond during protein synthesis.

peptidyl-tRNA. The tRNA molecule to which the growing peptide chain is attached during protein synthesis.

peripheral membrane protein. A membrane protein that is weakly bound to the interior or exterior surface of a membrane through ionic interactions and hydrogen bonding with the polar heads of the membrane lipids or with an integral membrane protein. Also known as an extrinsic membrane protein.

periplasmic space. The region between the plasma membrane and the cell wall in bacteria.

permeability coefficient. A measure of the ability of an ion or small molecule to diffuse across a lipid bilayer.

peroxisome. An organelle in all animal and many plant cells that carries out oxidation reactions, some of which produce the toxic compound hydrogen peroxide (H_2O_2). Peroxisomes contain the enzyme catalase, which catalyzes the breakdown of toxic H_2O_2 to water and O_2.

pH. A logarithmic quantity that indicates the acidity of a solution, that is, the concentration of hydronium ions in solution. pH is defined as the negative logarithm of the hydronium ion concentration.

pH optimum. In an enzyme-catalyzed reaction, the pH at the point of maximum catalytic activity.

phage. *See* bacteriophage.

phase-transition temperature (T_m). The midpoint of the temperature range in which lipids or other macromolecular aggregates are converted from a highly ordered phase or state (such as a gel) to a less-ordered state (such as a liquid crystal).

ϕ (phi). The angle of rotation around the bond between the α-carbon and the nitrogen of a peptide group.

phosphagen. A "high energy" phosphate storage molecule found in animal muscle cells. Phosphagens are phosphoamides and have a higher phosphoryl-group-transfer potential than ATP.

phosphatase. An enzyme that catalyzes the hydrolytic removal of a phosphoryl group.

phosphatidate. A glycerophospholipid that consists of two fatty acyl groups esterified to C-1 and C-2 of glycerol 3-phosphate. Phosphatidates are metabolic intermediates in the biosynthesis or breakdown of more complex glycerophospholipids.

phosphoanhydride. A compound formed by condensation of two phosphate groups.

phosphodiester linkage. A linkage in nucleic acids and other molecules in which two alcoholic hydroxyl groups are joined through a phosphate group.

phosphoester linkage. The bond by which a phosphoryl group is attached to an alcoholic or phenolic oxygen.

phospholipid. A lipid containing a phosphate moiety.

phosphorolysis. Cleavage of a bond within a molecule by group transfer to an oxygen atom of phosphate.

phosphorylase. An enzyme that catalyzes the cleavage of its substrate(s) via nucleophilic attack by inorganic phosphate (P_i) (i.e., via phosphorolysis).

phosphorylation. A reaction involving the addition of a phosphoryl group to a molecule.

phosphoryl group transfer potential. A measure of the ability of a compound to transfer a phosphoryl group to another compound. Under standard conditions, group transfer potentials have the same values as the standard free energies of hydrolysis but are opposite in sign.

photoautotroph. A photosynthetic organism that can utilize CO_2 as its main carbon source.

photon. A quantum of light energy.

photophosphorylation. The light-dependent formation of ATP from ADP and P_i catalyzed by chloroplast ATP synthase.

photoheterotroph. Photosynthetic organism that requires organic molecules as a carbon source.

photoreactivation. The direct repair of damaged DNA by an enzyme that is activated by visible light.

photorespiration. The light-dependent uptake of O_2 and the subsequent metabolism of phosphoglycolate that occurs primarily in C_3 photosynthetic plants. Photorespiration can occur because O_2 competes with CO_2 for the active site of ribulose 1,5-*bis*phosphate carboxylase-oxygenase, the enzyme that catalyzes the first step of the reductive pentose phosphate cycle.

photosynthesis. The conversion of light energy (photons) to chemical energy in the form of ATP and/or NADPH.

photosystem. A functional unit of the light-dependent electron-transfer reactions of photosynthesis. Each membrane-embedded photosystem contains a reaction center, which forms the core of the photosystem, and a pool of light-absorbing antenna pigments.

phototroph. An organism that can convert light energy into chemical potential energy (i.e., an organism capable of photosynthesis).

physiological pH. The normal pH of human blood, which is 7.4.

pI. *See* isoelectric point.

ping-pong reaction. A reaction in which an enzyme binds one substrate and releases a product, leaving a substituted enzyme that then binds a second substrate and releases a second product, thereby restoring the enzyme to its original form.

pitch. The axial distance for one complete turn of a helical structure.

pK_a. A logarithmic value that indicates the strength of an acid. pK_a is defined as the negative logarithm of the acid dissociation constant, K_a.

plasma membrane. The membrane that surrounds the cytoplasm of a cell and thus defines the perimeter of the cell.

plasmalogen. A glycerophospholipid that has a hydrocarbon chain linked to C-1 of glycerol 3-phosphate through a vinyl ether linkage. Plasmalogens are found in the central nervous system and in peripheral nerve and muscle tissue.

plasmid. A relatively small, extrachromosomal DNA molecule that is capable of autonomous replication. Plasmids are usually closed, circular, double-stranded DNA molecules.

P:O ratio. The ratio of molecules of ADP phosphorylated to atoms of oxygen reduced during oxidative phosphorylation.

polar. Having uneven distribution of charge. A molecule or functional group is polar if its center of negative charge does not coincide with its center of positive charge.

poly A tail. A stretch of polyadenylate, up to 250 nucleotide residues long, that is added to the 3′ end of a eukaryotic mRNA molecule following transcription.

polyacrylamide gel electrophoresis (PAGE). A technique used to separate molecules of different net charge and/or size based on their migration through a highly cross-linked gel matrix in an electric field.

polycistronic mRNA. An mRNA molecule that contains multiple coding regions. Many prokaryotic mRNA molecules are polycistronic.

polymerase chain reaction (PCR). A method for amplifying the amount of DNA in a sample and for enriching a particular DNA sequence in a population of DNA molecules. In the polymerase chain reaction, oligonucleotides complementary to the ends of the desired DNA sequence are used as primers for multiple rounds of DNA synthesis.

polynucleotide. A polymer of many (usually more than 20) nucleotide residues linked by phosphodiester bonds.

polypeptide. A polymer of many (usually more than 20) amino acid residues linked by peptide bonds.

polyribosome. *See* polysome.

polysaccharide. A polymer of many (usually more than 20) monosaccharide residues linked by glycosidic bonds. Polysaccharide chains can be linear or branched.

polysome. The structure formed by the binding of many translation complexes to a large mRNA molecule. Also known as a polyribosome.

polyunsaturated fatty acid. An unsaturated fatty acid with two or more carbon-carbon double bonds.

pore. *See* channel.

posttranscriptional processing. RNA processing that occurs after transcription is complete.

posttranslational modification. Covalent modification of a protein that occurs after synthesis of the polypeptide is complete.

prenylated protein. A lipid-anchored protein that is covalently linked to an isoprenoid moiety via the sulfur atom of a cysteine residue at the *C*-terminus of the protein.

primary structure. The sequence in which residues are covalently linked to form a polymeric chain.

primary transcript. A newly synthesized RNA molecule before processing.

primase. An enzyme in the primosome that catalyzes the synthesis of short pieces of RNA about 10 residues long. These oligonucleotides are the primers for synthesis of Okazaki fragments.

primosome. A multiprotein complex, including primase and helicase in *E. coli*, that catalyzes the synthesis of the short RNA primers needed for discontinuous DNA synthesis of the lagging strand.

processive enzyme. An enzyme that remains bound to its growing polymeric product through many polymerization steps (cf., distributive enzyme).

prochiral atom. An atom with multiple substituents, two of which are identical. A prochiral atom can become chiral when one of the identical substituents is replaced.

prokaryote. An organism, usually a single cell, which contains no nucleus or internal membranes (cf., eukaryote).

promoter. The region of DNA where RNA polymerase binds during transcription initiation.

prostaglandin. An eicosanoid that has a cyclopentane ring. Prostaglandins are metabolic regulators that act in the immediate neighborhood of the cells in which they are produced.

prosthetic group. A coenzyme that is tightly bound to an enzyme. A prosthetic group, unlike a cosubstrate, remains bound to a specific site of the enzyme throughout the catalytic cycle of the enzyme.

protease. An enzyme that catalyzes hydrolysis of peptide bonds. The physiological substrates of proteases are proteins.

protein. A biopolymer consisting of one or more polypeptide chains. The biological function of each protein molecule depends not only on the sequence of covalently linked amino acid residues, but also on its three-dimensional structure (conformation).

protein coenzyme. A protein that does not itself catalyze reactions but is required for the action of certain enzymes.

protein glycosylation. The covalent addition of carbohydrate to proteins. In *N*-glycosylation, the carbohydrate is attached to the amide group of the side chain of an asparagine residue. In *O*-glycosylation, the carbohydrate is attached to the hydroxyl group of the side chain of a serine or threonine residue.

protein kinase. *See* kinase.

protein phosphatase. *See* phosphatase.

proteoglycan. A complex of protein with glycosaminoglycan chains covalently bound through their anomeric carbon atoms. Up to 95% of the mass of a proteoglycan may be glycosaminoglycan.

proteomics. The study of all proteins produced in a certain cell type, tissue, organ, or organism.

protonmotive force (Δp). The energy stored in a proton concentration gradient across a membrane.

proximity effect. The increase in the rate of a nonenzymatic or enzymatic reaction attributable to high effective concentrations of reactants, which result in more frequent formation of transition states.

pseudo first-order reaction. A multi-reactant reaction carried out under conditions where the rate depends on the concentration of only one reactant.

pseudogene. A nonexpressed sequence of DNA that evolved from a protein-encoding gene. Pseudogenes often contain mutations in their coding regions and cannot produce functional proteins.

ψ (psi). The angle of rotation around the bond between the α-carbon and the carbonyl carbon of a peptide group.

$\Delta\psi$. *See* membrane potential.

P site. Peptidyl site. The site on a ribosome that is occupied during protein synthesis by a tRNA molecule attached to the growing polypeptide chain (peptidyl tRNA).

purine. A nitrogenous base having a two-ring structure in which a pyrimidine is fused to imidazole. Adenine and guanine are substituted purines found in both DNA and RNA.

pyranose. A monosaccharide structure that forms a six-membered ring as a result of intramolecular hemiacetal formation.

pyrimidine. A nitrogenous base having a heterocyclic ring that consists of four carbon atoms and two nitrogen atoms. Cytosine, thymine, and uracil are substituted pyrimidines found in nucleic acids (cytosine in DNA and RNA, uracil in RNA, and thymine principally in DNA).

***Q*.** *See* mass action ratio.

Q cycle. A cyclic pathway proposed to explain the sequence of electron transfers and proton movements within Complex III of mitochondria or the cytochrome *bf* complex in chloroplasts. The net result of the two steps of the Q cycle is oxidation of two molecules of QH_2 or plastoquinol (PQH_2); formation of one molecule of QH_2 or PQH_2; transfer of two electrons; and net translocation of four protons across the inner mitochondrial membrane to the intermembrane space or across the thylakoid membrane to the lumen.

quaternary structure. The organization of two or more polypeptide chains within a multisubunit protein.

R state. The more active conformation of an allosteric protein; opposite of T state.

Ramachandran plot. A plot of c versus f values for amino acid residues in a polypeptide chain. Certain f and c values are characteristic of different conformations.

random sequential reaction. A reaction in which neither the binding of substrates to an enzyme nor the release of products from the enzyme follows an obligatory order.

rate acceleration. The ratio of the rate constant for a reaction in the presence of enzyme (k_{cat}) divided by the rate constant for that reaction in the absence of enzyme (k_n). The rate acceleration value is a measure of the efficiency of an enzyme.

rate equation. An expression of the observed relationship between the velocity of a reaction and the concentration of each reactant.

rate determining step. The slowest step in a chemical reaction. The rate determining step has the highest activation energy among the steps leading to formation of a product from the substrate.

reaction center. A complex of proteins, electron transport cofactors, and a special pair of chlorophyll molecules that forms the core of a photosystem. The reaction center is the site of conversion of photochemical energy to electrochemical energy during photosynthesis.

reaction mechanism. The step-by-step atomic or molecular events that occur during chemical reactions.

reaction order. *See* kinetic order.

reaction specificity. The lack of formation of wasteful by-products by an enzyme. Reaction specificity results in essentially 100% product yields.

reactive center. The part of a coenzyme to which mobile metabolic groups are attached.

reading frame. The sequence of nonoverlapping codons of an mRNA molecule that specifies the amino acid sequence. The reading frame of an mRNA molecule is determined by the position where translation begins; usually an AUG codon.

receptor. A protein that binds a specific ligand, such as a hormone, leading to some cellular response.

recombinant DNA. A DNA molecule that includes DNA from different sources.

recombination. *See* genetic recombination.

reducing agent. A substance that loses electrons in an oxidation-reduction reaction and thereby becomes oxidized.

reducing end. The residue containing a free anomeric carbon in a polysaccharide. A polysaccharide usually contains no more than one reducing end.

reduction. The gain of electrons by a substance through transfer from another substance (the reducing agent). Reductions can take several forms, including the loss of oxygen from a compound, the addition of

hydrogen to a double bond of a compound, or a decrease in the valence of a metal ion.

reduction potential (*E*). A measure of the tendency of a substance to reduce other substances. The more negative the reduction potential, the greater the tendency to donate electrons.

regulated enzyme. An enzyme located at a critical point within one or more metabolic pathways, whose activity may be increased or decreased based on metabolic demand. Most regulated enzymes are oligomeric.

regulatory protein. A protein that is involved in the regulation of gene expression, usually at the point of transcription initiation. Repressors and activators are examples of regulatory proteins.

regulatory site. A ligand-binding site in a regulatory enzyme distinct from the active site. Allosteric modulators alter enzyme activity by binding to the regulatory site. Also known as an allosteric site.

relative molecular mass (M_r). The mass of a molecule relative to 1/12th the mass of ^{12}C. There are no units associated with the values for relative molecular mass.

release factor. A protein involved in terminating protein synthesis.

renaturation. The restoration of the native conformation of a biological macromolecule, usually resulting in restoration of biological activity.

replication. The duplication of double-stranded DNA, during which parental strands separate and serve as templates for synthesis of new strands. Replication is carried out by DNA polymerase and associated factors.

replication fork. The Y-shaped junction where double-stranded, template DNA is unwound and new DNA strands are synthesized during replication.

replisome. A multiprotein complex that includes DNA polymerase, primase, helicase, single-strand binding protein, and additional components. The replisomes, located at each of the replication forks, carry out the polymerization reactions of bacterial chromosomal DNA replication.

repressor. A regulatory DNA-binding protein that prevents transcription by RNA polymerase.

residue. A single component within a polymer. The chemical formula of a residue is that of the corresponding monomer minus the elements of water.

resonance energy transfer. A form of excitation energy transfer between molecules that does not involve transfer of an electron.

respiratory electron transport chain. A series of enzyme complexes and associated cofactors that are electron carriers, passing electrons from reduced coenzymes or substrates to molecular oxygen (O_2), the terminal electron acceptor of aerobic metabolism.

restriction endonuclease. An endonuclease that catalyzes the hydrolysis of double-stranded DNA at a specific nucleotide sequence. Type I restriction endonucleases catalyze both the methylation of host DNA and the cleavage of nonmethylated DNA, whereas type II restriction endonucleases catalyze only the cleavage of nonmethylated DNA.

restriction map. A diagram showing the size and arrangement of fragments produced from a DNA molecule by the action of various restriction endonucleases.

reverse transcriptase. A type of DNA polymerase that catalyzes the synthesis of a strand of DNA from an RNA template.

reverse turn. *See* turn.

ribonucleic acid (RNA). A polymer consisting of ribonucleotide residues joined by 3′–5′ phosphodiester bonds. The sugar moiety in RNA is ribose. Genetic information contained in DNA is transcribed in the synthesis of RNA, some of which (mRNA) is translated in the synthesis of protein.

ribonucleoprotein. A complex containing both ribonucleic acid and protein.

ribosome. A large ribonucleoprotein complex composed of multiple ribosomal RNA molecules and proteins. Ribosomes are the site of protein synthesis.

ribozyme. An RNA molecule with enzymatic activity.

rise. The distance between one residue and the next along the axis of a helical macromolecule.

RNA processing. The reactions that transform a primary RNA transcript into a mature RNA molecule. The three general types of RNA processing include the removal of RNA nucleotides from primary transcripts, the addition of RNA nucleotides not encoded by the gene, and the covalent modification of bases.

rRNA. *See* ribosomal ribonucleic acid.

S. *See* Svedberg unit.

S. *See* entropy.

salt bridge. *See* charge-charge interactions.

salvage pathway. A pathway in which a major metabolite, such as a purine or pyrimidine nucleotide, can be synthesized from a preformed molecular entity, such as a purine or pyrimidine.

saturated fatty acid. A fatty acid that does not contain a carbon-carbon double bond.

Schiff base. A complex formed by the reversible condensation of a primary amine with an aldehyde (to form an aldimine) or a ketone (to form a ketimine).

SDS-PAGE. *See* sodium dodecyl sulfate–polyacrylamide gel electrophoresis.

second messenger. A compound that acts intracellularly in response to an extracellular signal.

secondary structure. The regularities in local conformations within macromolecules. In proteins, secondary structure is maintained by hydrogen bonds between carbonyl and amide groups of the backbone. In nucleic acids, secondary structure is maintained by hydrogen bonds and stacking interactions between the bases.

second-order reaction. A reaction whose rate depends on the concentrations of two reactants.

self-splicing intron. An intron that is excised in a reaction mediated by the RNA precursor itself.

sense strand. In double-stranded DNA the sense strand is the strand that contains codons. Also called the coding strand. The opposite strand is called the antisense strand or the template strand.

sequential reaction. An enzymatic reaction in which all the substrates must be bound to the enzyme before any product is released.

sequential theory of cooperativity and allosteric regulation. A model of the cooperative binding of identical ligands to oligomeric proteins. According to the simplest form of the sequential theory, the binding of a ligand may induce a change in the tertiary structure of the subunit to which it binds and may alter the conformations of neighboring subunits to varying extents. Only one subunit conformation has a high affinity for the ligand. Also known as the ligand-induced theory.

Shine-Dalgarno sequence. A purine-rich region just upstream of the initiation codon in prokaryotic mRNA molecules. The Shine-Dalgarno sequence binds to a pyrimidine-rich sequence in the ribosomal RNA, thereby positioning the ribosome at the initiation codon.

σ factor. *See* σ subunit.

σ subunit (sigma subunit). A subunit of prokaryotic RNA polymerase, which acts as a transcription initiation factor by binding to the promoter. Different σ subunits are specific for different promoters. Also known as a σ factor.

signal peptidase. An integral membrane protein of the endoplasmic reticulum that catalyzes cleavage of the signal peptide of proteins translocated to the lumen.

signal peptide. The *N*-terminal sequence of residues in a newly synthesized polypeptide that targets the protein for translocation across a membrane.

signal transduction. The process whereby an extracellular signal is converted to an intracellular signal by the action of a membrane-associated receptor, a transducer, and an effector enzyme.

signal recognition particle (SRP). A eukaryotic protein-RNA complex that binds a newly synthesized peptide as it is extruded from the ribosome. The signal-recognition particle is involved in anchoring the ribosome to the cytosolic face of the endoplasmic

reticulum so that protein translocation to the lumen can occur.

single-strand binding protein (SSB). A protein that binds tightly to single-stranded DNA, preventing the DNA from folding back on itself to form double-stranded regions.

site-directed mutagenesis. An in vitro procedure by which one particular nucleotide residue in a gene is replaced by another, resulting in production of an altered protein sequence.

site-specific recombination. An example of recombination that occurs at specific sites in the genome.

small nuclear ribonucleoprotein (snRNP). An RNA-protein complex composed of one or two specific snRNA molecules plus a number of proteins. snRNPs are involved in splicing mRNA precursors and in other cellular events.

small RNA. A class of RNA molecules. Some small RNA molecules have catalytic activity. Some small nuclear RNA molecules (snRNA) are components of small nuclear ribonucleoproteins (snRNPs).

snRNA. *See* small nuclear RNA.

snRNP. *See* small nuclear ribonucleoprotein.

sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE). Polyacrylamide gel electrophoresis performed in the presence of the detergent sodium dodecyl sulfate. SDS-PAGE allows separation of proteins on the basis of size only rather than charge and size.

solvation. A state in which a molecule or ion is surrounded by solvent molecules.

solvation sphere. The shell of solvent molecules that surrounds an ion or solute.

special pair. A specialized pair of chlorophyll molecules in reaction centers that is the primary electron donor during the light-dependent reactions of photosynthesis.

specific heat. The amount of heat required to raise the temperature of 1 gram of a substance by 1°C.

specificity constant. *See* k_{cat}/K_m.

sphingolipid. An amphipathic lipid with a sphingosine (trans-4-sphingenine) backbone. Sphingolipids, which include sphingomyelins, cerebrosides, and gangliosides, are present in plant and animal membranes and are particularly abundant in the tissues of the central nervous system.

sphingomyelin. A sphingolipid that consists of phosphocholine attached to the C-1 hydroxyl group of a ceramide. Sphingomyelins are present in the plasma membranes of most mammalian cells and are a major component of myelin sheaths.

splice site. The conserved nucleotide sequence surrounding an exon-intron junction. It includes the site where the RNA molecule is cleaved during intron excision.

spliceosome. The large protein-RNA complex that catalyzes the removal of introns from mRNA precursors. The spliceosome is composed of small nuclear ribonucleoproteins.

splicing. The process of removing introns and joining exons to form a continuous RNA molecule.

SRP. *See* signal recognition particle.

SSB. *See* single-strand binding protein.

stacking interactions. The weak noncovalent forces between adjacent bases or base pairs in single-stranded or double-stranded nucleic acids, respectively. Stacking interactions contribute to the helical shape of nucleic acids.

standard Gibbs free energy change ($\Delta G^{\circ\prime}$). The free energy change for a reaction under biochemical standard state conditions.

standard reduction potential ($E^{\circ\prime}$). A measure of the tendency of a substance to reduce other substances under biochemical standard state conditions.

standard state. A set of reference conditions for a chemical reaction. In biochemistry, the standard state is defined as a temperature of 298 K (25°C), a pressure of 1 atmosphere, a solute concentration of 1.0 M, and a pH of 7.0.

starch. A homopolymer of glucose residues that is a storage polysaccharide in plants. There are two forms of starch: amylose, an unbranched polymer of glucose residues joined by α-(1 → 4) linkages; and amylopectin, a branched polymer of glucose residues joined by α-(1 → 4) linkages with α-(1 → 6) linkages at branch points.

steady state. A state in which the rate of synthesis of a compound is equal to its rate of utilization or degradation.

stem-loop. *See* hairpin.

stereoisomers. Compounds with the same molecular formula but different spatial arrangements of their atoms.

stereospecificity. The ability of an enzyme to recognize and act upon only a single stereoisomer of a substrate.

steroid. A lipid containing a fused, four-ring isoprenoid structure.

sterol. A steroid containing a hydroxyl group.

stomata. Structures on the surface of a leaf through which carbon dioxide diffuses directly into photosynthetic cells.

stop codon. *See* termination codon.

strand invasion. The exchange of single strands of DNA from two nicked molecules having homologous nucleotide sequences.

stroma. The interior of a chloroplast corresponding to the cytoplasm of the ancestral cyanobacterium.

stromal lamellae. Regions of the thylakoid membrane that are in contact with the stroma.

substrate. A reactant in a chemical reaction. In enzymatic reactions, substrates are specifically acted upon by enzymes, which catalyze the conversion of substrates to products.

substrate cycle. A pair of opposing reactions that catalyzes a cycle between two pathway intermediates.

substrate level phosphorylation. Phosphorylation of a nucleoside diphosphate by transfer of a phosphoryl group from a nonnucleotide substrate.

supercoil. A topological arrangement assumed by over- or underwound double-stranded DNA. Underwinding gives rise to negative supercoils; overwinding produces positive supercoils.

supersecondary structure. *See* motif.

Svedberg unit (S). A unit of 10^{-13} second used for expressing the sedimentation coefficient, a measure of the rate at which a large molecule or particle sediments in an ultracentrifuge. Large S values usually indicate large masses.

symport. The cotransport of two different species of ions or molecules in the same direction across a membrane by a transport protein.

synonymous codons. Different codons that specify the same amino acid.

synthase. A common name for an enzyme, often a transferase, that catalyzes a synthetic reaction.

synthetase. An enzyme that catalyzes the joining of two substrates and requires the input of the chemical potential energy of a nucleoside triphosphate. Synthetases are members of the IUBMB class of enzymes known as ligases.

T state. The less active conformation of an allosteric protein; opposite of R state.

TATA box. An A/T-rich DNA sequence found within the promoter of both prokaryotic and eukaryotic genes.

template strand. The strand of DNA within a gene whose nucleotide sequence is complementary to that of the transcribed RNA. During transcription, RNA polymerase binds to and moves along the template strand in the 3′ → 5′ direction, catalyzing the synthesis of RNA in the 5′ → 3′ direction.

termination codon. A codon that is recognized by specific proteins that cause newly synthesized peptides to be released from the translation machinery thus terminating translation. The three termination codons (UAG, UAA, and UGA) are also known as stop codons.

termination sequence. A sequence at the 3′ end of a gene that mediates transcription termination.

tertiary structure. The compacting of polymeric chains into one or more domains within a macromolecule. In proteins, tertiary structure is stabilized mainly by hydrophobic interactions between side chains.

thermodynamics. The branch of physical science that studies transformations of heat and energy.

30 nm fiber. A chromatin structure in which nucleosomes are coiled into a solenoid 30 nm in diameter.

−35 region. A sequence found within the promoter of some prokaryotic genes about 30 to 35 base pairs upstream of the transcription initiation site.

3_{10} helix. A secondary structure of proteins, consisting of a helix in which the carbonyl oxygen of each amino acid residue (residue *n*) forms a hydrogen bond with the amide hydrogen of the third residue further toward the *C*-terminus of the polypeptide chain (residue $n + 3$).

thylakoid lamella. *See* thylakoid membrane.

thylakoid membrane. A highly folded, continuous membrane network suspended in the aqueous matrix of the chloroplast. The thylakoid membrane is the site of the light-dependent reactions of photosynthesis, which lead to the formation of NADPH and ATP. Also known as the thylakoid lamella.

T_m. *See* melting point and phase-transition temperature.

topoisomerase. An enzyme that alters the supercoiling of a DNA molecule by cleaving a phosphodiester linkage in either one or both strands, rewinding the DNA, and resealing the break. Some topoisomerases are also known as DNA gyrases.

topology. 1. The arrangement of membrane-spanning segments and connecting loops in an integral membrane protein. **2.** The overall morphology of a nucleic acid molecule.

TψC arm. The stem-and-loop structure in a tRNA molecule that contains the sequence ribothymidylate–pseudouridylate–cytidylate (TψC).

trace element. An element required in very small quantities by living organisms. Examples include copper, iron, and zinc.

transaminase. An enzyme that catalyzes the transfer of an amino group from an α-amino acid to an α-keto acid. Transaminases require the coenzyme pyridoxal phosphate. They are also called aminotransferases.

transcription. The copying of biological information from a double-stranded DNA molecule to a single-stranded RNA molecule, catalyzed by a transcription complex consisting of RNA polymerase and associated factors.

transcription bubble. A short region of double-stranded DNA that is unwound by RNA polymerase during transcription.

transcription factor. A protein that binds to the promoter region, to RNA polymerase, or to both during assembly of the transcription initiation complex. Some transcription factors remain bound during RNA chain elongation.

transcription initiation complex. The complex of RNA polymerase and other factors that assembles at the promoter at the start of transcription.

transcriptional activator. A regulatory DNA-binding protein that enhances the rate of transcription by increasing the activity of RNA polymerase at specific promoters.

transducer. The component of a signal-transduction pathway that couples receptor-ligand binding with generation of a second messenger catalyzed by an effector enzyme.

transfer ribonucleic acid. *See* tRNA.

transferase. An enzyme that catalyzes a group-transfer reaction. Transferases often require a coenzyme.

transition state. An unstable, high-energy arrangement of atoms in which chemical bonds are being formed or broken. Transition states have structures between those of the substrates and the products of a reaction.

transition-state analog. A compound that resembles a transition state. Transition-state analogs characteristically bind extremely tightly to the active sites of appropriate enzymes and thus act as potent inhibitors.

transition-state stabilization. The increased binding of transition states to enzymes relative to the binding of substrates or products. Transition-state stabilization lowers the activation energy and thus contributes to catalysis.

translation. The synthesis of a polypeptide whose sequence reflects the nucleotide sequence of an mRNA molecule. Amino acids are donated by activated tRNA molecules, and peptide bond synthesis is catalyzed by the translation complex, which includes the ribosome and other factors.

translation complex. The complex of a ribosome and protein factors that carries out the translation of mRNA in vivo.

translation initiation complex. The complex of ribosomal subunits, an mRNA template, an initiator tRNA molecule, and initiation factors that assembles at the start of protein synthesis.

translation initiation factor. A protein involved in the formation of the initiation complex at the start of protein synthesis.

translocation. 1. The movement of the ribosome by one codon along an mRNA molecule. **2.** The movement of a polypeptide through a membrane.

transposon. A mobile genetic element that jumps between chromosomes or parts of a chromosome by taking advantage of recombination mechanisms. Also known as a transposable element.

transverse diffusion. The passage of lipid or protein molecules from one leaflet of a lipid bilayer to the other leaflet. Unlike lateral diffusion within one leaflet of a bilayer, transverse diffusion is extremely slow.

triacylglycerol. A lipid containing three fatty acyl residues esterified to glycerol. Fats and oils are mixtures of triacylglycerols. Formerly known as a triglyceride.

tricarboxylic acid cycle. *See* citric acid cycle.

triglyceride. *See* triacylglycerol.

triose. A three-carbon sugar.

tRNA. A class of RNA molecules that carry activated amino acids to the site of protein synthesis for incorporation into growing peptide chains. tRNA molecules contain an anticodon that recognizes a complementary codon in mRNA.

turn (in proteins). A protein loop of 4-5 residues that causes a change in the direction of a polypeptide chain in a folded protein.

turnover. The dynamic metabolic steady state in which molecules are degraded and replaced by newly synthesized molecules.

turnover number. *See* catalytic constant.

twist. The angle of rotation between adjacent residues within a helical macromolecule.

type I reaction center. The special pair of chlorophyll molecules and associated electron transfer chain found in photosystem I.

type II reaction center. The reaction center found in photosystem II.

uncompetitive inhibition. Inhibition of an enzyme-catalyzed reaction by a reversible inhibitor that binds only to the enzyme-substrate complex, not to the free enzyme.

uncouplers. *See* uncoupling agent.

uncoupling agent. A compound that disrupts the usual tight coupling between electron transport and phosphorylation of ADP.

uniport. The transport of a single type of solute across a membrane by a transport protein.

unsaturated fatty acid. A fatty acid with at least one carbon-carbon double bond. An unsaturated fatty acid with only one carbon-carbon double bond is called a monounsaturated fatty acid. A fatty acid with two or more carbon-carbon double bonds is called a polyunsaturated fatty acid. In general, the double bonds of unsaturated fatty acids are of the *cis* configuration and are separated from each other by methylene ($—CH_2—$) groups.

urea cycle. A metabolic cycle consisting of four enzyme-catalyzed reactions that converts nitrogen from ammonia and aspartate to urea. Four ATP equivalents are consumed during formation of one molecule of urea.

***v*.** *See* velocity.

v_0. *See* initial velocity.

vacuole. A fluid-filled organelle in plant cells that is a storage site for water, ions, or nutrients.

van der Waals force. A weak intermolecular force produced between neutral atoms by transient electrostatic interactions. Van der Waals attraction is strongest when atoms are separated by the sum of their van der Waals radii; strong van der Waals repulsion precludes closer approach.

van der Waals radius. The effective size of an atom. The distance between the nuclei of two nonbonded atoms at the point of maximal attraction is the sum of their van der Waals radii.

variable arm. The arm of a tRNA molecule that is located between the anticodon arm and the TψC arm. The variable arm can range in length from about 3 to 21 nucleotides.

velocity (V). The rate of a chemical reaction, expressed as amount of product formed per unit time.

very low density lipoprotein (VLDL). A type of plasma lipoprotein that transports endogenous triacylglycerols, cholesterol, and cholesteryl esters from the liver to the tissues.

vitamin. An organic micronutrient that cannot be synthesized by an animal and must be obtained in the diet. Many coenzymes are derived from vitamins.

VLDL. *See* very low density lipoprotein.

V_{max}. *See* maximum velocity.

wax. A nonpolar ester that consists of a long chain monohydroxylic alcohol and a long chain fatty acid.

wobble position. The 5′ position of an anticodon, where non-Watson-Crick base pairing with a nucleotide in mRNA is permitted. The wobble position makes it possible for a tRNA molecule to recognize more than one codon.

X-ray crystallography. A technique used to determine secondary, tertiary, and quaternary structures of biological macromolecules. In X-ray crystallography, a crystal of the macromolecule is bombarded with X rays, which are diffracted and then detected electronically or on a film. The atomic structure is deduced by mathematical analysis of the diffraction pattern.

Z-DNA. A conformation of oligonucleotide sequences containing alternating deoxycytidylate and deoxyguanylate residues. Z-DNA is a left-handed double helix containing approximately 12 base pairs per turn.

zero-order reaction. A reaction whose rate is independent of reactant concentration.

Z-scheme. A zigzag scheme that illustrates the reduction potentials associated with electron flow through photosynthetic electron carriers.

zwitterion. A molecule containing negatively and positively charged groups.

Photo and Illustration Credits

Chapter 1 Page 2 top, Science Photo Library/Photo Researchers, Inc.; 2 middle, Photos 12/Alamy; 2 bottom, Science Photo Library/Photo Researchers, Inc.; 3 top, Corbis; 3 bottom, Shutterstock; 11, Shutterstock; 12, Manuscripts & Archives—Yale University Library; 15 top, SSPL/The Image Works; 15 bottom, Richard Bizley/Photo Researchers, Inc.; 18 top, Lee D. Simon/Photo Researchers, Inc.; 18 bottom, National Library of Medicine Profiles in Science; 20, Matthew Daniels, Wellcome Images; 22, Dr. Torsten Wittmann/Photo Researchers, Inc.; and 23, David S. Goodsell, the RCSB Protein Data Bank. Coordinates from PDB entry 1atn.

Chapter 2 Page 28 top, NASA; 28 bottom, Michael Charters; 31, iStockphoto; 32, NOAA; 33, Valley Vet Supply; 37, Travel Ink/Getty Images; 41, Elemental-Imaging/iStockphoto; 44 top, Edgar Fahs Smith Memorial Collection; 44 bottom, Fotolia; and 48, Library of Congress.

Chapter 3 Page 56, Thomas Deerinck, NCMIR/Photo Researchers, Inc.; 57, Argonne National Laboratory; 58, Pascal Goetgheluck/Photo Researchers, Inc.; 60, iStockphoto; 69, iStockphoto; 70, MARKA/Alamy; 71, Bio-Rad Laboratories, Inc.; 73 top, REUTERS/William Philpott WP/HB; 73 bottom, AFP Photo/Newscom; and 78, Bettmann/CORBIS.

Chapter 4 Page 85, Shutterstock; 86, Swiss Institute of Bioinformatics; 88, Lisa A. Shoemaker; 89 top, Bror Strandberg; 89 bottom, Hulton Archive/Getty Images; 93, Custom Life Science Images/Alamy; 94, Bettmann/Corbis; 95, Julian Voss-Andreae; 108, From Kühner et al., "Proteome Organization in a Genome-Reduced Bacterium" Science 27 Nov 2009 Vol. 326 no. 5957 pp. 1235–1240. American Association for the Advancement of Science.; 109, Howard Ochman; 111, From Butland et al., "Interaction network containing conserved and essential protein complexes in Escherichia coli," Nature 433 (2005), 531–537; 113, National Library of Medicine; 117, Laurence A. Moran; 119, Easawara Subramanian, http://www.nature.com/nsmb/journal/v8/n6/full/nsb0601_489.html; 121, Danielle Anthony; 122, SSPL/The Image Works; 123, Janice Carr/Centers for Disease Control; 126, Ed Uthman, licensed via Creative Commons http:// creativecommons.org/licenses/by/2.0/; and 127, Julian Voss-Andreae.

Chapter 5 Page 135, Dorling Kindersley; 136, Jonathan Elegheert; 137, Michael P. Walsh/IUBMB; 138, Leonardo DaVinci; 142 top, Rockefeller Archives Center; 142 bottom left, University of Pittsburgh, Archives Service Center; 142 bottom right, Laurence A. Moran; and 149, AP Photo/Paul Sakuma.

Chapter 6 Page 167, Ronsdale Press, photo copyright Dina Goldstein; 174, Bettmann/CORBIS; 183, Paramount/Photofest; and 186, Shutterstock.

Chapter 7 Page 198, Shutterstock; 200, Library of Congress; 204, Heath Folp/Industry & Investment NSW; 209, History Press; 212, Christian Heintzen, University of Manchester; 214, iStockphoto; 215, John Olive; 216, Stephanie Schuller/Photo Researchers, Inc.; 219 left, Meg and Raul via Flickr/CC-BY-2.0 http://creativecommons.org/licenses/by/2.0/deed.en 219 right, and 220, Shutterstock; and 223, both, ©® The Nobel Foundation.

Chapter 8 Pages 227, 239, 240, Shutterstock; 244 top, Image Source/Alamy; 244 bottom, Jack Griffith; 245, Jakob Jeske/Fotolia; 246, Jens Stougaard; 247 top, Eric Erbe, Christopher Pooley, Beltsville Agricultural Research/USDA; 247 bottom, Robert Hubert, Microbiology Program, Iowa State University; and 252, Christine Ortlepp.

Chapter 9 Page 258, imagebroker/Alamy; 262 top, Steve Gschmeissner/Photo Researchers, Inc.; 262 bottom, Shutterstock; 268 bottom, Shutterstock; 270, John Ross; 273 top, Professors Pietro M. Motta & Tomonori Naguro/Photo Researchers, Inc.; 273 bottom, Biophoto Associates/Photo Researchers, Inc.; 277, Lisa A. Shoemaker; 278 bottom, Julie Marie/Fotolia; 284 top, M.M. Perry; and 284 bottom, Shutterstock.

Chapter 10 Page 294, Quade Paul, Echo Medical Media; 296, Charles Boone, From Costanzo et al. "The Genetic Landscape of a Cell" *Science 327*; (2010):425–432; 297, Roche Applied Science; 303, Shutterstock; 305 top, University of Edinburgh/Wellcome Images; 305 bottom, Biophoto Associates/Photo Researchers, Inc.; and 312, National Library of Medicine.

Chapter 11 Page 325, Barton W. Spear—Pearson Education; 331 left, Super-Stock, Inc;. 331 right, Bettmann/CORBIS; 336, Warner Bros./Photofest; 341, ChinaFotoPress/Zuma/ICON/Newscom; and 349, dreambigphotos/Fotolia.

Chapter 12 Page 359, CBS/Landov; 369, United States Postal Service; 370 top, A. Jones/Photo Researchers, Inc.; 370 bottom, Laura Van Niftrik; and 375, Tim Crosby/Getty Images.

Chapter 13 Page 386, Science Photo Library/Photo Researchers, Inc.; 387, From Zhou, Z.H. et al. (2001) Proc. Natl. Acad. Sci. USA 98, pp. 14802–14807; 390 top, From Zhou, Z.H. et al. (2001) Proc. Natl. Acad. Sci. USA 98, pp. 14802–14807; 390 bottom, NASA; and 396, 401, Shutterstock.

Chapter 14 Page 417 top and left, Shutterstock; 417 bottom, Dirk Freder/iStockphoto; 419 top, Lisa A. Shoemaker; 419 middle and bottom, Shutterstock; 420 top Roberto Danovaro; 420 left, Milton Saier; 426, Michael Radermacher; 433, Alexander Tzagoloff; and 438, NASA/Sandra Joseph and Kevin O'Connell.

Chapter 15 Page 443, Mary Ginsburg; 444, Arizona State University—Plant Bio Department; 447 top, Makoto Kusaba; 447 bottom, Shutterstock; 448 top, CHINE NOUVELLE/SIPA/Newscom; 448 bottom, Robert Lucking; 452, Niels Ulrik Frigaard; 457, Michelle Liberton, Howard Berg, and Himadri Pakrasi, of the Donald Danforth Plant Science Center and of Washington University, St. Louis; 458 top, Andrew Syred/Photo Researchers, Inc.; 458 bottom, NSF Polar Programs/NOAA; 459, Lisa A. Shoemaker; 462, Lawrence Berkeley National Laboratory; 468, Shutterstock; 469 top, From Bhattacharyya et al, "The wrinkled-seed ..." *Cell*, Vol 60, No 1, 1990, pp 115–122; 469 middle, Peter Arnold/Photolibrary; 469 bottom, Fotolia; 470, From David F. Savage et al., "Spatially Ordered Dynamics of the Bacterial Carbon Fixation Machinery," 2011. American Association for the Advancement of Science; 471 top, AP Photo/Charlie Neibergall; and 471 bottom, Shutterstock.

Chapter 16 Page 475, Kennan Ward/Corbis; 486, Shutterstock; 490 top, Bettmann/CORBIS; 490 bottom, Hulton Archive/Getty Images; 493, Environmental Justice Foundation, Ltd.; 495 top, David Leys, Toodgood et al., 2004; 495 bottom, Eric Clark/Molecular Expressions; 501 top, Shutterstock; 501 bottom, Steve Gschmeissner/SPL/Alamy; 504, Donald Nicholson/IUBMB; 506, Shutterstock; and 507, Robin Fraser.

Chapter 17 Page 515 top, NASA Visible Earth; 515 bottom, NOAA; 516, Inga Spence/Photo Researchers, Inc.; 531, Shutterstock; 532, iStockphoto.com; 534, National Library of Medicine; and 540, U.S Air Force photo/Staff Sgt Eric T. Sheler.

Chapter 18 Page 552, G. Robert Greenberg; 554, National Library of Medicine; 561, Peter Reichard; 564, Shutterstock; and 568, Fotolia.

Chapter 19 Page 574, National Cancer Institute; 581, SSPL/The Image Works; 587, Andrew Paterson/Alamy; 589, Lisa A. Shoemaker; 591 both, Ulrich K. Laemmli; 597 top left, 597 top right, Lisa A. Shoemaker; 597 middle, Stanford University School of Medicine; and 597 bottom, Steve Northup/Time&Life Images/Getty Images.

Chapter 20 Page 603 top, John Cairns; 603 bottom left, David S. Hogness; 603 bottom right, Regional Oral History Office, The Bancroft Library, University of California, Berkeley; 613 both, Timothy Lohman; 615, From *Structure,* 6, Dec. 2008 Copyright Elsevier. Original artwork by Glass Egg Design, Jessica Eichman, www.glasseggdesign.com; 618, Lisa A. Shoemaker; 619, David Bentley; 627 top, Laguna Design/Photo Researchers, Inc.; 627 bottom, Paul Sabatier/Art Life Images/Superstock; 628 top, James Kezer/Stanley Sessions; 628 bottom, Dr. L. Caro/Photo Researchers, Inc; 630, Institute of Molecular-biology and Biophysics, From Yamada et al., *Molecular Cell* Vol 10 p 671 (2002). Figure 4b (right), with permission from Elsevier.; and 630, Vanderbilt University, Genes and Development. From Wang et al. *BASC, a super complex of BRCA1-associated proteins involved in the recognition and repair of aberrant DNA structures.* Vol. 14, No. 8, pp. 927–939, April 15, 2000 Fig 3M.

Chapter 21 Page 634, Marc Gantier/Getty Images; 636, From Murakami.et al., Science 296: 1285–1290 (2002) Fig5A (left) American Association for the Advancement of Science; 638, Oscar L. Miller, Jr.; and 651, Lisa A. Shoemaker.

Chapter 22 Page 666, National Security Agency; 667, US Navy Office of Information; 675, David Goodsell; 677, Stanford University School of Medicine; 681, Oscar L. Miller, Jr.; and 692, H. H. Mollenhauer/USDA.

This page intentionally left blank

Index

In this index, the page numbers listed indicate tables (with a T added to that page number) and figures (with an F added to that page number).

Common Abbreviations in Biochemistry

ACP	acyl carrier protein
ADP	adenosine 5′-diphosphate
AMP	adenosine 5′-monophosphate (adenylate)
cAMP	3′,5′-cyclic adenosine monophosphate
ATP	adenosine 5′-triphosphate
bp	base pair
1,3BPG	1,3-*bis*phosphoglycerate
2,3BPG	2,3-*bis*phosphoglycerate
CDP	cytidine 5′-diphosphate
CMP	cytidine 5′-monophosphate (cytidylate)
CoA	coenzyme A
CTP	cytidine 5′-triphosphate
DHAP	dihydroxyacetone phosphate
DNA	deoxyribonucleic acid
cDNA	complementary DNA
DNase	deoxyribonuclease
$E°$	reduction potential
$E°'$	standard reduction potential
EF	elongation factor
emf	electromotive force
ETF	electron-transferring flavoprotein
$\mathcal{F}$	Faraday's constant
FAD	flavin adenine dinucleotide
$FADH_2$	flavin adenine dinucleotide (reduced form)
F1,6BP	fructose 1,6-*bis*phosphate
FMN	flavin mononucleotide
$FMNH_2$	flavin mononucleotide (reduced form)
F6P	fructose 6-phosphate
ΔG	actual free-energy change
$\Delta G°'$	standard free-energy change
GDP	guanosine 5′-diphosphate
GMP	guanosine 5′-monophosphate (guanylate)
cGMP	3′,5′-cyclic guanosine monophosphate
G3P	glyceraldehyde 3-phosphate
G6P	glucose 6-phosphate
GTP	guanosine 5′-triphosphate
H	enthalpy
Hb	hemoglobin
HDL	high density lipoprotein
HETPP	hydroxyethylthiamine pyrophosphate
HPLC	high-pressure liquid chromatography
IDL	intermediate density lipoprotein
IF	initiation factor
eIF	eukaryotic initiation factor
IMP	inosine 5′-monophosphate
IP_3	inositol 1,4,5-*tris*phosphate
K_a	acid dissociation constant
k_{cat}	catalytic constant
K_{eq}	equilibrium constant
K_m	Michaelis constant
kb	kilobase pair
LDL	low density lipoprotein
LHC	light-harvesting complex
M_r	relative molecular mass
Mb	myoglobin
$NAD^{\oplus}$	nicotinamide adenine dinucleotide
NADH	nicotinamide adenine dinucleotide (reduced form)
$NADP^{\oplus}$	nicotinamide adenine dinucleotide phosphate
NADPH	nicotinamide adenine dinucleotide phosphate (reduced form)
$NMN^{\oplus}$	nicotinamide mononucleotide
NDP	nucleoside 5′-diphosphate
NMP	nucleoside 5′-monophosphate
NTP	nucleoside 5′-triphosphate
dNTP	deoxynucleoside triphosphate
P_i	inorganic phosphate (or orthophosphate)
PAGE	polyacrylamide gel electrophoresis
PCR	polymerase chain reaction
2PG	2-phosphoglycerate
3PG	3-phosphoglycerate
PEP	phosphoenolpyruvate
PFK	phosphofructokinase
pI	isoelectric point
PIP_2	phosphatidylinositol 4,5-*bis*phosphate
PLP	pyridoxal phosphate
PP_i	inorganic pyrophosphate
PQ	plastoquinone
PQH_2	plastoquinol
PRPP	5-phosphoribosyl 1-pyrophosphate
PSI	photosystem I
PSII	photosystem II
Q	ubiquinone
QH_2	ubiquinol
RF	release factor
RNA	ribonucleic acid
mRNA	messenger ribonucleic acid
rRNA	ribosomal ribonucleic acid
snRNA	small nuclear ribonucleic acid
tRNA	transfer ribonucleic acid
RNase	ribonuclease
snRNP	small nuclear ribonucleoprotein
RPP	reductive pentose phosphate
Rubisco	ribulose 1,5-*bis*phosphate carboxylase-oxygenase
S	entropy
dTDP	deoxythymidine 5′-diphosphate
TF	transcription factor
dTMP	deoxythymidine 5′-monophosphate (thymidylate)
TPP	thiamine pyrophosphate
dTTP	deoxythymidine 5′-triphosphate
UDP	uridine 5′-diphosphate
UMP	uridine 5′-monophosphate (uridylate)
UTP	uridine 5′-triphosphate
v	velocity
V_{max}	maximum velocity
v_0	initial velocity
VLDL	very low density lipoprotein
XMP	xanthosine 5′-monophosphate

Abbreviations for amino acids are given on pages 57–62, and those for major pyrimidine and purine bases are given on page 575.

First position (5′ end)	Second position U	C	A	G	Third position (3′ end)
U	Phe	Ser	Tyr	Cys	U
	Phe	Ser	Tyr	Cys	C
	Leu	Ser	STOP	STOP	A
	Leu	Ser	STOP	Trp	G
C	Leu	Pro	His	Arg	U
	Leu	Pro	His	Arg	C
	Leu	Pro	Gln	Arg	A
	Leu	Pro	Gln	Arg	G
A	Ile	Thr	Asn	Ser	U
	Ile	Thr	Asn	Ser	C
	Ile	Thr	Lys	Arg	A
	Met	Thr	Lys	Arg	G
G	Val	Ala	Asp	Gly	U
	Val	Ala	Asp	Gly	C
	Val	Ala	Glu	Gly	A
	Val	Ala	Glu	Gly	G

One- and three-letter abbreviations for amino acids

A	Ala	Alanine
B	Asx	Asparagine or aspartate
C	Cys	Cysteine
D	Asp	Aspartate
E	Glu	Glutamate
F	Phe	Phenylalanine
G	Gly	Glycine
H	His	Histidine
I	Ile	Isoleucine
K	Lys	Lysine
L	Leu	Leucine
M	Met	Methionine
N	Asn	Asparagine
P	Pro	Proline
Q	Gln	Glutamine
R	Arg	Arginine
S	Ser	Serine
T	Thr	Threonine
V	Val	Valine
W	Trp	Tryptophan
Y	Tyr	Tyrosine
Z	Glx	Glutamate or glutamine