ARSENIC POISONING SHUTS DOWN METABOLISM

Disrupting the body's ability to extract energy from nutrients can drastically affect health. Arsenic is a chemical element that, if present in the body in excess, shuts down metabolism. It can do so suddenly or gradually.

Given in one large dose, arsenic poisoning causes chest pain, vomiting, diarrhea, shock, coma, and death. In contrast, many small doses cause dark skin lesions that feel as if they are burning, numb hands and feet, and eventually skin cancer. Such gradual poisoning, called arsenicosis, may occur from contact with pesticides or environmental pollutants. The world's largest outbreak of arsenicosis, however, is due to a natural exposure.

When the World Bank and UNICEF began tapping into aquifers in India and Bangladesh in the late 1960s, they were trying to supply clean water to areas ravaged by sewage and industrial waste released from rivers subject to cycles of floods and droughts. Millions of people had already perished from diarrheal diseases due to the poor sanitation. But digging wells to provide clean water backfired when workers unwittingly penetrated a layer of sediment naturally rich in arsenic. The chemical has since been leaching into the water in at least 2 million wells in Bangladesh, reaching levels fifty times the safety limit set by the World Health Organization. When effects on health began to appear years later, the people thought arsenicosis was contagious. Affected individuals not only suffered pain, but were shunned.

Arsenic damages the body by binding to bonds between sulfur atoms in proteins. It affects metabolism by impairing an enzyme that transports the breakdown products of glucose into mitochondria, where energy is extracted. The cell runs out of energy.

Today UNICEF is helping the people of India and Bangladesh to avoid arsenic poisoning. Workers are diagnosing and treating arsenicosis and providing tanks to collect and store rainwater. A vast education campaign has softened the stigma of arsenicosis. Although cases will continue to appear for a few more decades, the use of alternate water sources has finally slowed the progression of this public health problem.

4.1 INTRODUCTION

In every human cell, even in the most sedentary individual, thousands of chemical reactions essential to life take place every second. Special types of proteins called **enzymes** control the rate of each reaction. The sum total of chemical reactions in the cell constitutes **metabolism.**

Many metabolic reactions occur one after the other in a linked fashion, in which the products of one reaction are starting materials for the next. These reactions form pathways and cycles that may intersect where they share intermediate compounds, each step catalyzed by an enzyme. Metabolism in its entirety is complex. Individual pathways of metabolism reveal how cells function—in essence, how chemistry underlies biology.

Metabolic reactions and pathways can be subgrouped. *Intermediary metabolism* refers to the processes that obtain, release, and use energy. Another way to classify metabolic reactions is by their necessity. *Primary metabolites* are products of metabolism essential to survival. *Secondary metabolites* are not essential to survival, but may provide an advantage or enhancement. Secondary metabolites are best studied in plants, where they usually help to defend against predators because they are toxins. Some of our most successful drugs are plant secondary metabolites. The vinca alkaloids, for example, protect the rosy Madagascar periwinkle that produces them by sickening animals that eat the vegetation, but we use these biochemicals to treat cancer. Their effect is to destabilize microtubule formation.

This chapter covers two complex and related subjects. The first is how metabolic pathways supply energy to a cell. Then, as an illustration of how cellular energy is used, and also of how proteins such as enzymes are produced, the second major topic considers how information in the building block sequences of DNA instructs the cell to assemble amino acids into proteins.

4.2 METABOLIC PROCESSES

Metabolic reactions and pathways are of two types. In **anabolism** (ăh-nab′o-liz″-ĕm), larger molecules are constructed from smaller ones, requiring input of energy. In **catabolism** (kă-tab′o-liz″-ĕm), larger molecules are broken down into smaller ones, releasing energy.

Anabolism

Anabolism provides all the materials required for cellular growth and repair. For example, a type of anabolic process called **dehydration synthesis** (de″hi-dra′shun sin′the-sis) joins many simple sugar molecules (monosaccharides) to form larger molecules of glycogen. When a runner consumes pasta the night before a race, digestion breaks down the complex carbohydrates in the prerace meal to monosaccharides. These are absorbed into the bloodstream, which carries the energy-rich molecules to body cells. Here, dehydration synthesis joins the monosaccharides to form glycogen, which stores energy that the runner may not need until later, as the finish line nears. When monosaccharide units join, an —OH (hydroxyl group) from one monosaccharide molecule and an —H (hydrogen atom) from an —OH group of another are removed. As the —H and —OH react to produce a water molecule, the monosaccharides are joined by a shared oxygen atom, as figure 4.1 shows (read from left to right). As the process repeats, the molecular chain extends, forming a polysaccharide.

Glycerol and fatty acid molecules also join by dehydration synthesis in fat (adipose tissue) cells to form fat molecules. In

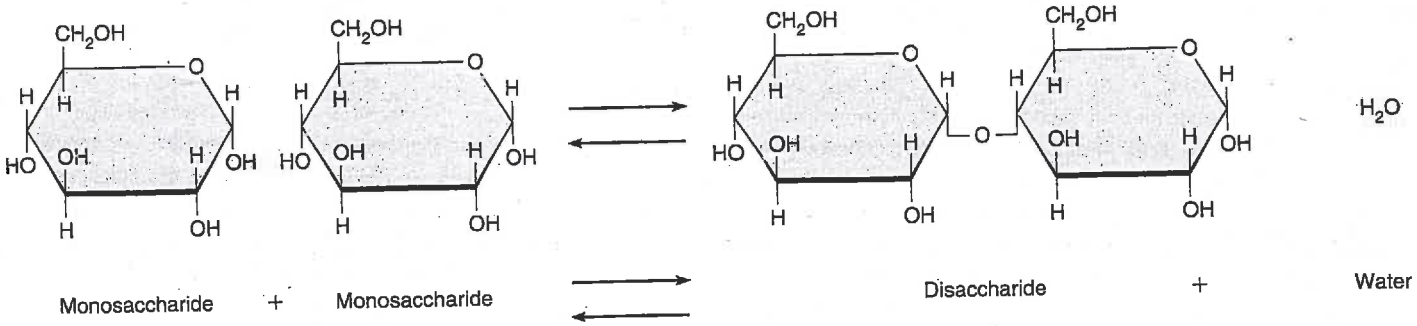


FIGURE 4.1 Building up and breaking down molecules. A disaccharide is formed from two monosaccharides in a dehydration synthesis reaction (arrows to the right). In the reverse reaction, hydrolysis, a disaccharide is broken down into two monosaccharides (arrows to the left).

this case, three hydrogen atoms are removed from a glycerol molecule, and an —OH group is removed from each of three fatty acid molecules, as figure 4.2 shows (read from left to right). The result is three water molecules and a single fat molecule whose glycerol and fatty acid portions are bound by shared oxygen atoms.

In cells, dehydration synthesis also builds protein molecules by joining amino acid molecules. When two amino acid molecules are united, an —OH from the —COOH group of one and an —H from the $—NH_2$ group of another are removed. A water molecule forms, and the amino acid molecules join by a bond between a carbon atom and a nitrogen atom (fig. 4.3; read from left to right). This type of bond, called a *peptide bond*, holds the amino acids together. Two such bound amino acids form a *dipeptide*, and many joined in a chain form a *polypeptide*. Generally, a polypeptide consisting of 100 or more amino acid molecules is called a *protein*, although the boundary between polypeptides and proteins is not precisely defined. Some proteins consist of more than one polypeptide chain.

Nucleic acids are also formed by dehydration synthesis. This process is described later in the chapter.

Catabolism

Metabolic processes that break down larger molecules into smaller ones constitute catabolism. An example of catabolism is **hydrolysis** (hi-drol'ĭ-sis), which can decompose carbohydrates, lipids, and proteins. A water molecule is used for each bond that is broken. Hydrolysis of a disaccharide, for instance, yields two monosaccharide molecules (see fig. 4.1; read from right to left). The bond between the simple sugars breaks, and the water molecule supplies a hydrogen atom to one sugar molecule and a hydroxyl group to the other. Hydrolysis is the reverse of dehydration synthesis.

Hydrolysis breaks down carbohydrates into monosaccharides; fats into glycerol and fatty acids (see fig. 4.2; read from right to left); proteins into amino acids (see fig. 4.3; read from right to left); and nucleic acids into nucleotides. It does not occur automatically, even though in the body water molecules are readily available to provide the necessary —H and —OH. For example, water-soluble substances such as the disaccharide sucrose (table sugar) *dissolve* in a glass of water but do not undergo hydrolysis. Like dehydration synthesis, hydrolysis requires specific enzymes, discussed in the next section, Control of Metabolic Reactions.

The reactions of metabolism are often reversible. However, the enzyme that speeds, or catalyzes, an anabolic reaction is often different from that which catalyzes the corresponding catabolic reaction.

Both catabolism and anabolism must be carefully controlled so that the breakdown or energy-releasing reactions occur at rates adjusted to the requirements of the building up or energy-utilizing reactions. Any disturbance in this balance is likely to damage or kill cells.

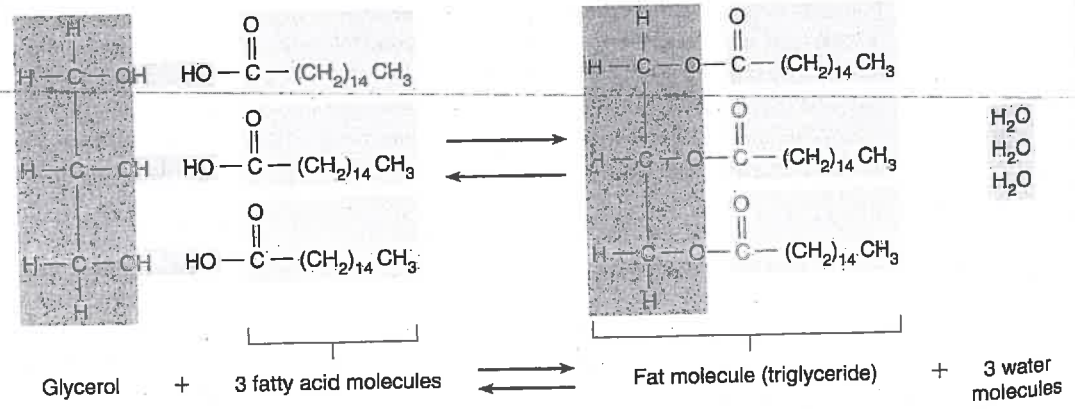


FIGURE 4.2 Forming a fat. A glycerol molecule and three fatty acid molecules react, yielding a fat molecule (triglyceride) in a dehydration synthesis reaction (arrows to the right). In the reverse reaction, hydrolysis, a triglyceride is broken down into three fatty acids and a glycerol (arrows to the left).

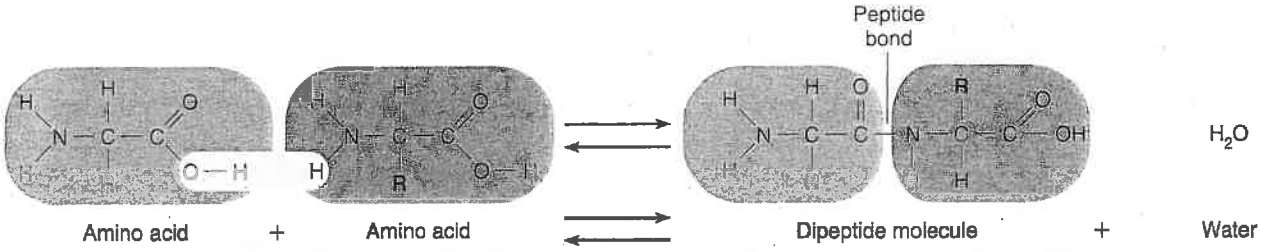


FIGURE 4.3 Peptide bonds link amino acids. When dehydration synthesis unites two amino acid molecules, a peptide bond forms between a carbon atom and a nitrogen atom, resulting in a dipeptide molecule (arrows to the right). In the reverse reaction, hydrolysis, a dipeptide molecule is broken down into two amino acids (arrows to the left).

PRACTICE

1 What are the general functions of anabolism and catabolism?

2 What type of molecule is formed by the anabolism of monosaccharides? Of glycerol and fatty acids? Of amino acids?

3 Distinguish between dehydration synthesis and hydrolysis.

4.3 CONTROL OF METABOLIC REACTIONS

Different types of cells may conduct specialized metabolic processes, but all cells perform certain basic reactions, such as the buildup and breakdown of carbohydrates, lipids, proteins, and nucleic acids. These common reactions include hundreds of very specific chemical changes that must occur in particular sequences. Enzymes control the rates of these metabolic reactions.

Enzyme Action

Like other chemical reactions, metabolic reactions require energy *(activation energy)* before they proceed. This is why in laboratory experiments heat is used to increase the rates of chemical reactions. Heat energy increases the rate at which molecules move and the frequency of molecular collisions. These collisions increase the likelihood of interactions among the electrons of the molecules that can form new chemical bonds. The temperature conditions in cells are usually too mild to adequately promote the reactions of life. Enzymes make these reactions possible.

Most enzymes are globular proteins that catalyze specific chemical reactions in cells by lowering the activation energy required to start these reactions. Enzymes can speed metabolic reactions by a factor of a million or more.

Enzymes are required in small amounts, because as they work, they are not consumed and can, therefore, function repeatedly. Each enzyme is specific, acting only on a particular molecule, called its **substrate** (sub′strāt). For example, the substrate of an enzyme called catalase (found in the peroxisomes of liver and kidney cells) is hydrogen peroxide, a toxic by-product of certain metabolic reactions. This enzyme's only function is to decompose hydrogen peroxide into water and oxygen, an action that helps prevent accumulation of hydrogen peroxide, which damages cells.

> The action of the enzyme catalase is obvious when using hydrogen peroxide to cleanse a wound. Injured cells release catalase, and when hydrogen peroxide contacts them, bubbles of oxygen are set free. The resulting foam removes debris from inaccessible parts of the wound.

Each enzyme must be able to "recognize" its specific substrate. This ability to identify a substrate depends upon the shape of an enzyme molecule. That is, each enzyme's polypeptide chain twists and coils into a unique three-dimensional conformation that fits the particular shape of its substrate molecule.

RECONNECT
To Chapter 2, Proteins, page 65.

During an enzyme-catalyzed reaction, regions of the enzyme molecule called **active sites** temporarily combine with portions of the substrate, forming an enzyme-substrate complex. This interaction strains chemical bonds in the substrate in a way that makes a particular chemical reaction more likely to occur. When it does, the enzyme is released in its original form, able to bind another substrate molecule (fig. 4.4). Many enzyme-catalyzed reactions are reversible and in some cases the same enzyme catalyzes both directions.

Enzyme catalysis can be summarized as follows:

$$\text{Substrate} + \text{Enzyme} \rightarrow \underset{\text{complex}}{\text{Enzyme-substrate}} \rightarrow \text{Product} + \underset{\text{(unchanged)}}{\text{Enzyme}}$$

The speed of an enzyme-catalyzed reaction depends partly on the number of enzyme and substrate molecules in the cell. The reaction occurs more rapidly if the concentration of the enzyme or the concentration of the substrate increases. The efficiency of different types of enzymes varies greatly. Some enzymes can process only a few substrate molecules per second, whereas others can handle as many as hundreds of thousands.

Cellular metabolism includes hundreds of different chemical reactions, each controlled by a specific type of

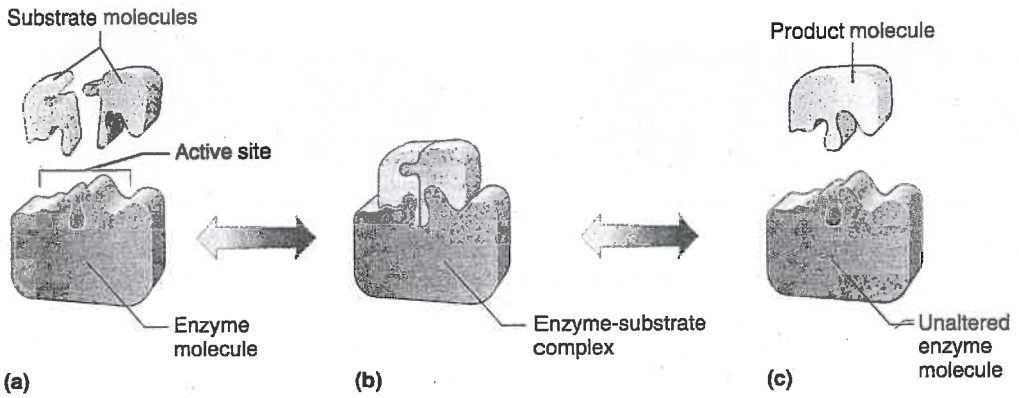


FIGURE 4.4 An enzyme-catalyzed reaction. (Many enzyme-catalyzed reactions, as depicted here, are reversible.) In the forward reaction (dark-shaded arrows), (*a*) the shapes of the substrate molecules fit the shape of the enzyme's active site. (*b*) When the substrate molecules temporarily combine with the enzyme, a chemical reaction occurs. (*c*) The result is a product molecule and an unaltered enzyme. The active site changes shape somewhat as the substrate binds, such that formation of the enzyme-substrate complex is more like a hand fitting into a glove, which has some flexibility, than a key fitting into a lock.

enzyme. Often sequences of enzyme-controlled reactions, called **metabolic pathways,** lead to synthesis or breakdown of particular biochemicals (fig. 4.5). Hundreds of different types of enzymes are present in every cell.

Enzyme names are often derived from the names of their substrates, with the suffix *-ase* added. For example, a lipid-splitting enzyme is called a *lipase,* a protein-splitting enzyme is a *protease,* and a starch (amylum)-splitting enzyme is an *amylase.* Similarly, *sucrase* is an enzyme that splits the sugar sucrose, *maltase* splits the sugar maltose, and *lactase* splits the sugar lactose.

Regulation of Metabolic Pathways

The rate at which a metabolic pathway functions is often determined by a regulatory enzyme that catalyzes one of its steps. The number of molecules of such a regulatory enzyme is limited. Consequently, these enzymes can become saturated when the substrate concentration exceeds a certain level. Once this happens, increasing the substrate concentration no longer affects the reaction rate. The enzyme becomes ineffectual at high substrate concentrations, so it is termed a **rate-limiting enzyme.**

Such an enzyme is often the first enzyme in a series (fig. 4.6). This position is important because an intermediate product of the pathway might accumulate if an enzyme occupying another position in the sequence were rate limiting.

Often the product of a metabolic pathway inhibits the rate-limiting regulatory enzyme. This type of control is an example of negative feedback. Accumulating product inhibits the pathway, and synthesis of the product falls. When the concentration of product decreases, the inhibition lifts, and more product is synthesized. In this way, a single enzyme can control a whole pathway, stabilizing the rate of production (fig. 4.6).

RECONNECT
To Chapter 1, Homeostasis, page 9.

Cofactors and Coenzymes

An enzyme may be inactive until it combines with a nonprotein component called a **cofactor** which helps the active site attain its appropriate shape or helps bind the enzyme to its

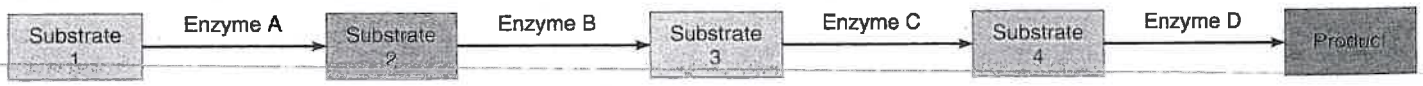


FIGURE 4.5 A metabolic pathway consists of a series of enzyme-controlled reactions leading to formation of a product.

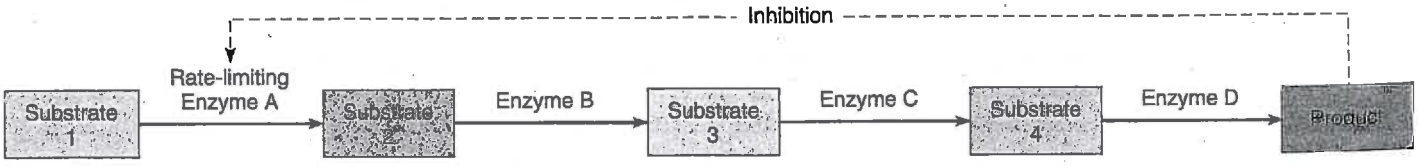


FIGURE 4.6 A negative feedback mechanism may control a rate-limiting enzyme in a metabolic pathway. The product of the pathway inhibits the enzyme.

substrate. A cofactor may be an ion of an element, such as copper, iron, or zinc, or a small organic molecule, called a **coenzyme** (ko-en′zīm). Many coenzymes are composed of vitamin molecules or incorporate altered forms of vitamin molecules into their structures.

Vitamins are essential organic molecules that human cells cannot synthesize (or may not synthesize in sufficient amounts) and therefore must come from the diet. Vitamins provide coenzymes that can, like enzymes, function repeatedly, so cells require small amounts of vitamins. An example is coenzyme A (derived from the vitamin pantothenic acid), which is necessary for one of the reactions of cellular respiration, discussed in the next section. Chapter 18 (pp. 710–716) discusses vitamins further.

Factors That Alter Enzymes

Almost all enzymes are proteins, and like other proteins, they can be denatured by exposure to excessive heat, radiation, electricity, certain chemicals, or fluids with extreme pH values. For example, many enzymes become inactive at 45°C, and nearly all of them are denatured at 55°C. Some poisons denature enzymes. Cyanide, for instance, can interfere with respiratory enzymes and damage cells by halting their energy-obtaining reactions.

Certain microorganisms, colorfully called "extremophiles," live in conditions of extremely high or low heat, salinity, or pH. Their enzymes have evolved under these conditions and are useful in industrial processes too harsh to use other enzymes.

PRACTICE

4 How can an enzyme control the rate of a metabolic reaction?
5 How does an enzyme "recognize" its substrate?
6 How can a rate-limiting enzyme be an example of negative feedback control of a metabolic pathway?
7 What is the role of a cofactor?
8 What factors can denature enzymes?

4.4 ENERGY FOR METABOLIC REACTIONS

Energy is the capacity to change something; it is the ability to do work. Therefore, we recognize energy by what it can do. Common forms of energy are heat, light, sound, electrical energy, mechanical energy, and chemical energy.

Although energy cannot be created or destroyed, it can be changed from one form to another. An ordinary incandescent light bulb changes electrical energy to heat and light, and an automobile engine changes the chemical energy in gasoline to heat and mechanical energy.

Cellular respiration is the process that transfers energy from molecules such as glucose and makes it available for cellular use. The chemical reactions of cellular respiration must occur in a particular sequence, each one controlled by a different enzyme. Some of these enzymes are in the cell's cytosol, whereas others are in the mitochondria. Such precision of activity suggests that the enzymes are physically positioned in the exact sequence as that of the reactions they control. The enzymes responsible for some of the reactions of cellular respiration are located in tiny, stalked particles on the membranes (cristae) in the mitochondria (see chapter 3, p. 84).

Changes in the human body are a characteristic of life—whenever this happens, energy is being transferred. Thus, all metabolic reactions involve energy in some form.

ATP Molecules

Adenosine triphosphate (ATP) is a molecule that carries energy in a form that the cell can use. Each ATP molecule consists of three main parts—an adenine, a ribose, and three phosphates in a chain (fig. 4.7). The second and third phosphates of ATP are attached by high-energy bonds, and the chemical energy stored in one or both high-energy bonds may be quickly transferred to another molecule in a metabolic reaction. Energy from the breakdown of ATP powers cellular work such as skeletal muscle contraction, active transport across cell membranes, secretion, and many other functions.

An ATP molecule that loses its terminal phosphate becomes an **adenosine diphosphate (ADP)** molecule, which has only two phosphates. ATP can be resynthesized from an ADP by using energy released from cellular respiration to reattach a phosphate, in a process called **phosphorylation** (fos″fōr-ĭ-la′shun). Thus, as shown in figure 4.8, ATP and ADP molecules shuttle back and forth between the energy-transferring reactions of cellular respiration and the energy-transferring reactions of the cell.

ATP is the primary energy-carrying molecule in a cell. Even though there are other energy carriers, without enough ATP, cells quickly die.

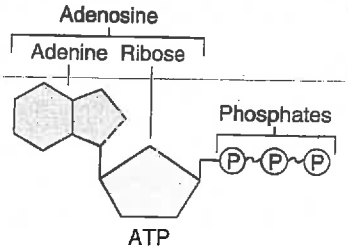


FIGURE 4.7 ATP provides cellular energy currency. An ATP (adenosine triphosphate) molecule consists of an adenine, a ribose, and three phosphates. The wavy lines connecting the last two phosphates represent high-energy chemical bonds.

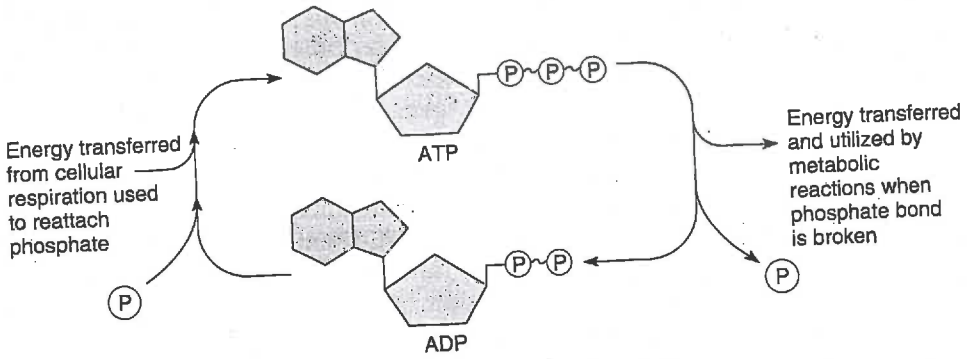


FIGURE 4.8 ATP provides energy for metabolic reactions in cells. Cellular respiration generates ATP.

Release of Chemical Energy

Most metabolic processes require chemical energy stored in ATP. This form of energy is initially held in the chemical bonds that link atoms into molecules and is released when these bonds break. Burning a marshmallow over a campfire releases the chemical energy held in the bonds of the molecules that make up the marshmallow as heat and light. Similarly, when a marshmallow is eaten, digested, and absorbed, cells "burn" glucose molecules from that marshmallow in a process called **oxidation** (ok″sĭ-da′shun). The energy released by oxidation of glucose is harnessed to promote cellular metabolism.

Oxidation of substances inside cells and the burning of substances outside them have important differences. Burning in nonliving systems (such as starting a fire in a fireplace) usually requires a great deal of energy to begin, and most of the energy released escapes as heat or light. In cells, enzymes initiate oxidation by lowering the activation energy. Also, by transferring energy to ATP, cells are able to capture almost half of the energy released in the form of chemical energy. The rest escapes as heat, which helps maintain body temperature.

PRACTICE

9 What is energy?

10 Define cellular respiration.

11 How does cellular oxidation differ from burning?

4.5 CELLULAR RESPIRATION

Cellular respiration occurs in three distinct, yet interconnected, series of reactions: **glycolysis** (glī-kol′ĭ-sis), the **citric acid cycle**, and the **electron transport chain** (oxidative phosphorylation) (fig. 4.9). The products of these reactions include carbon dioxide (CO_2), water, and energy. Although most of the energy is lost as heat, almost half is captured as ATP.

Cellular respiration includes **aerobic** (a″er-ōb′ik), reactions which require oxygen, and **anaerobic** (an-a″er-ōb′ik) reactions, which do not require oxygen. For each glucose molecule decomposed completely by cellular respiration, up to thirty-eight molecules of ATP can be produced. All but two ATP molecules are formed by the aerobic reactions.

Glycolysis

Both aerobic and anaerobic pathways begin with glycolysis. Literally "the breaking of glucose," glycolysis is a series of ten enzyme-catalyzed reactions that break down the 6-carbon glucose molecule into two 3-carbon pyruvic acid molecules. Glycolysis occurs in the cytosol (see fig. 4.9), and because it does not require oxygen, it is sometimes referred to as the *anaerobic phase of cellular respiration.*

Three main events occur during glycolysis (fig. 4.10):

1. First, glucose is phosphorylated by the addition of two phosphate groups, one at each end of the molecule. Although this step requires ATP, it "primes" the molecule for some of the energy-releasing reactions that occur later.
2. Second, the 6-carbon glucose molecule is split into two 3-carbon molecules.
3. Third, the electron carrier NADH is produced, ATP is synthesized, and two 3-carbon pyruvic acid molecules result. Some of the reactions of glycolysis release hydrogen atoms. The electrons of these hydrogen atoms contain much of the energy associated with the chemical bonds of the original glucose molecule. To keep this energy in a form the cell can use, these hydrogen atoms are passed in pairs to molecules of the hydrogen carrier NAD^+ (nicotinamide adenine dinucleotide). In this reaction, two of the electrons and one hydrogen nucleus bind to NAD^+ to form NADH. The remaining hydrogen nucleus (a hydrogen ion) is released as follows:

$$NAD^+ + 2H \rightarrow NADH + H^+$$

NADH delivers these high-energy electrons to the electron transport chain elsewhere in the mitochondria, where most of the ATP will be synthesized.

ATP is also synthesized directly in glycolysis. After subtracting the two ATP used in the priming step, this gives a net yield of two ATP per molecule of glucose.

Glycolysis

1 The 6-carbon sugar glucose is broken down in the cytosol into two 3-carbon pyruvic acid molecules with a net gain of 2 ATP and the release of high-energy electrons.

Citric Acid Cycle

2 The 3-carbon pyruvic acids generated by glycolysis enter the mitochondria. Each loses a carbon (generating CO_2) and is combined with a coenzyme to form a 2-carbon acetyl coenzyme A (acetyl CoA). More high-energy electrons are released.

3 Each acetyl CoA combines with a 4-carbon oxaloacetic acid to form the 6-carbon citric acid, for which the cycle is named. For each citric acid, a series of reactions removes 2 carbons (generating 2 CO_2's), synthesizes 1 ATP, and releases more high-energy electrons. The figure shows 2 ATP, resulting directly from 2 turns of the cycle per glucose molecule that enters glycolysis.

Electron Transport Chain

4 The high-energy electrons still contain most of the chemical energy of the original glucose molecule. Special carrier molecules bring the high-energy electrons to a series of enzymes that convert much of the remaining energy to more ATP molecules. The other products are heat and water. The function of oxygen as the final electron acceptor in this last step is why the overall process is called aerobic respiration.

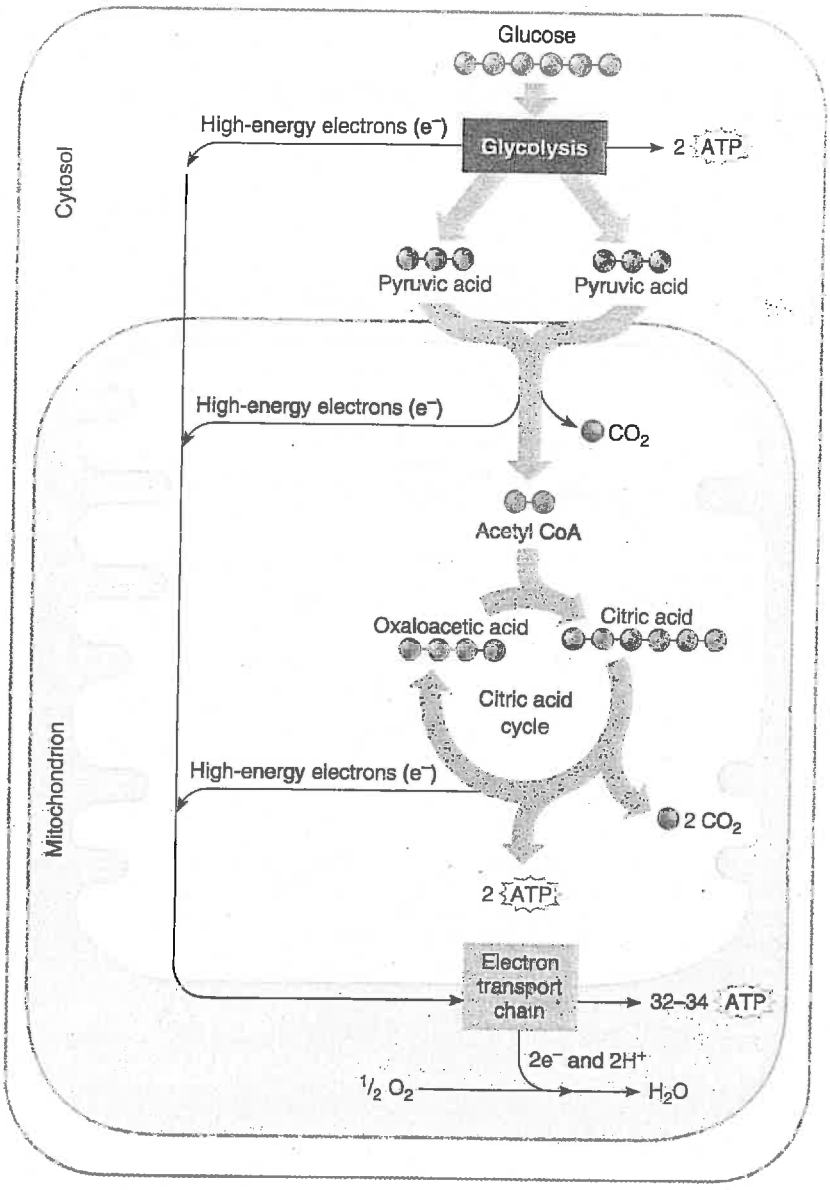


FIGURE 4.9 Glycolysis occurs in the cytosol and does not require oxygen. Aerobic respiration occurs in the mitochondria and only in the presence of oxygen. The products include ATP, heat, carbon dioxide, and water. Two ATP are generated by glycolysis, 2 result directly from the citric acid cycle, and 32–34 are generated by the electron transport chain. Thus, the total yield of ATP molecules per glucose molecule is 36–38, depending on the type of cell.

PRACTICE

12 What are the final products of cellular respiration?

13 What are aerobic and anaerobic reactions?

14 What is the result of glycolysis?

Anaerobic Reactions

For glycolysis to continue, NADH + H^+ must be able to deliver electrons to the electron transport chain, replenishing the cellular supply of NAD^+. In the presence of oxygen, this is exactly what happens. Oxygen acts as the final electron acceptor at the end of the electron transport chain, enabling the chain to continue processing electrons and recycling NAD^+.

Under anaerobic conditions, however, the electron transport chain has nowhere to unload its electrons, and it can no longer accept new electrons from NADH. As an alternative, NADH + H^+ can give its electrons and hydrogens back to pyruvic acid in a reaction that forms **lactic acid.** Although this regenerates NAD^+, the buildup of lactic acid eventually inhibits glycolysis, and ATP production declines. The lactic acid diffuses into the blood, and when oxygen levels return to normal the liver converts the lactic acid back into pyruvic acid, which can finally enter the aerobic pathway.

PRACTICE

15 What is the role of oxygen in cellular respiration?

16 Under what conditions does a cell produce lactic acid?

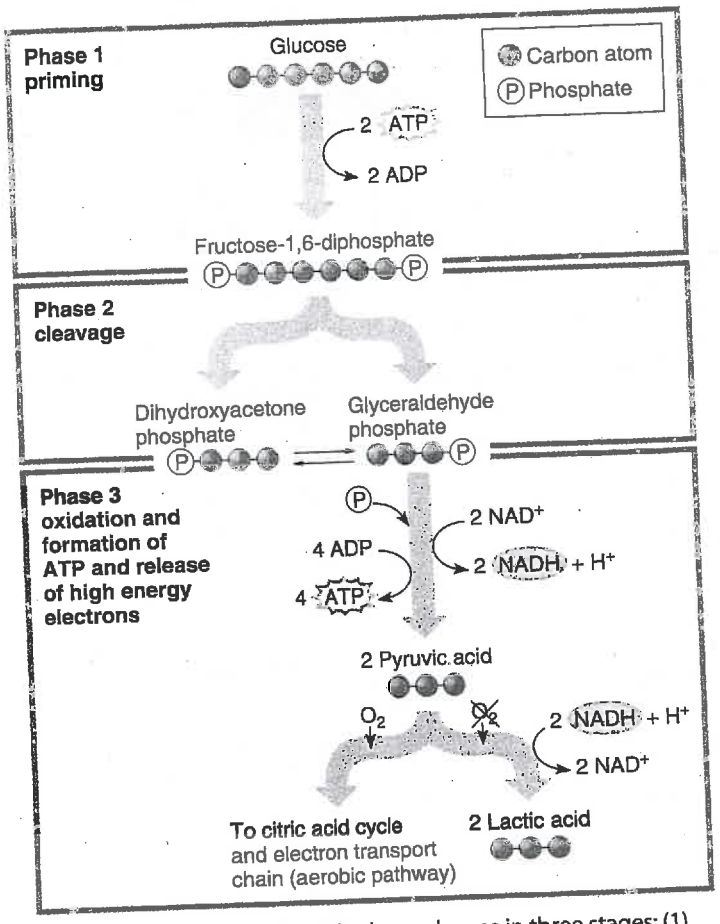


FIGURE 4.10 Glycolysis breaks down glucose in three stages: (1) phosphorylation, (2) splitting, and (3) production of NADH and ATP. Each glucose molecule broken down by glycolysis yields a net gain of 2 ATP.

Human muscle cells working so strenuously that their production of pyruvic acid exceeds the oxygen supply produce lactic acid. In this "oxygen debt," the muscle cells use solely the anaerobic pathway, which provides fewer ATPs per glucose molecule than do the aerobic reactions. The accumulation of lactic acid contributes to muscle fatigue and cramps. Walking after cramping can increase bloodflow that hastens depletion of lactic acid, easing the pain.

Aerobic Reactions

If enough oxygen is available, the pyruvic acid generated by glycolysis can continue through the aerobic pathways (see fig. 4.9). These reactions include the synthesis of **acetyl coenzyme A** (as'ĕ-til ko-en'zīm A) or acetyl CoA, the citric acid cycle, and the electron transport chain. In addition to carbon dioxide and water, the aerobic reactions yield up to thirty-six ATP molecules per glucose.

This book presents the theoretical yield of the aerobic reactions—up to 36 ATP per glucose molecule. In fact, more energy may be required to complete these reactions than once thought. Estimates taking this into account indicate a yield of ATP less than the theoretical maximum.

The aerobic reactions begin with pyruvic acid produced in glycolysis moving from the cytosol into the mitochondria (fig. 4.11). From each pyruvic acid molecule, enzymes inside the mitochondria remove two hydrogen atoms, a carbon atom, and two oxygen atoms, generating NADH and a CO_2 and leaving a 2-carbon acetic acid. The acetic acid then combines with a molecule of coenzyme A to form acetyl CoA. CoA "carries" the acetic acid into the citric acid cycle.

Citric Acid Cycle

The citric acid cycle begins when a 2-carbon acetyl CoA molecule combines with a 4-carbon oxaloacetic acid molecule to form the 6-carbon citric acid and CoA (fig. 4.11). The citric acid is changed through a series of reactions back into oxaloacetic acid. The CoA can be used again to combine with acetic acid to form acetyl CoA. The cycle repeats as long as the mitochondrion receives oxygen and pyruvic acid.

The citric acid cycle has three important consequences:

1. One ATP is produced directly for each citric acid molecule that goes through the cycle.
2. For each citric acid molecule, eight hydrogen atoms with high-energy electrons are transferred to the hydrogen carriers NAD^+ and the related FAD (flavine adenine dinucleotide):

$$NAD^+ + 2H \rightarrow NADH + H^+$$

$$FAD + 2H \rightarrow FADH_2$$

3. As the 6-carbon citric acid reacts to form the 4-carbon oxaloacetic acid, two carbon dioxide molecules are produced.

The carbon dioxide produced by the formation of acetyl CoA and in the citric acid cycle dissolves in the cytoplasm, diffuses from the cell, and enters the bloodstream. Eventually, the respiratory system excretes the carbon dioxide.

Electron Transport Chain

The hydrogen and high-energy electron carriers (NADH and $FADH_2$) generated by glycolysis and the citric acid cycle now hold most of the energy contained in the original glucose molecule. To couple this energy to ATP synthesis, the high-energy electrons are handed off to the electron transport chain, a series of enzyme complexes that carry and pass electrons along from one to another. These complexes dot the folds of the inner mitochondrial membranes (see chapter 3, p. 84), which, if stretched out, may be forty-five times as long as the cell membrane in some cells. The electron transport chain passes each electron along, gradually

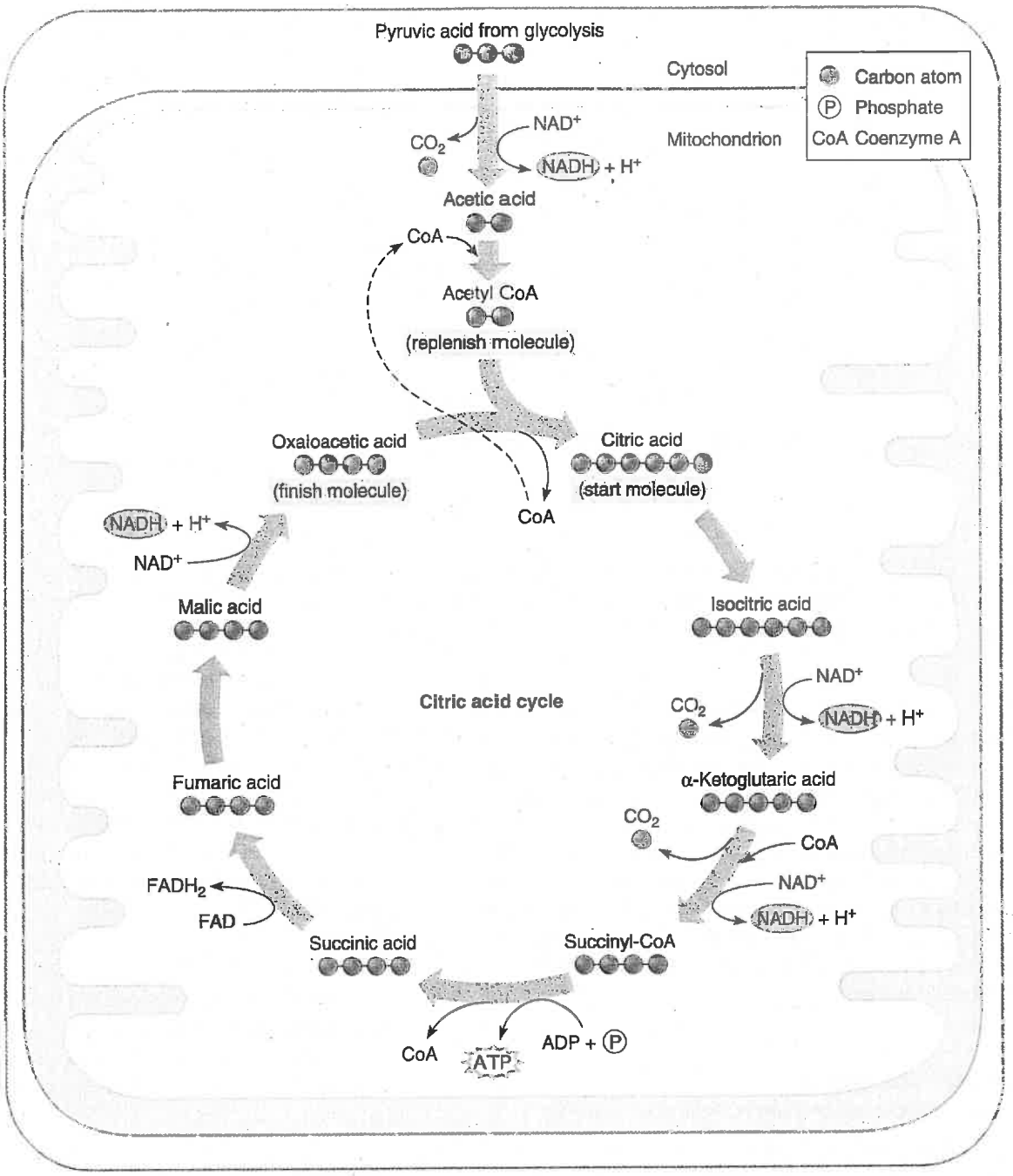


FIGURE 4.11 Each turn of the citric acid cycle (two "turns" or citric acids per glucose) produces one ATP directly, and two CO_2 molecules. Eight hydrogens with high-energy electrons are released.

lowering the electron's energy level and transferring that energy to ATP synthase, an enzyme complex that uses this energy to phosphorylate ADP to form ATP (fig. 4.12). These reactions, known as oxidation/reduction reactions, are described further in Appendix C, pages 944–947.

Neither glycolysis nor the citric acid cycle uses oxygen directly, although they are part of the aerobic metabolism of glucose. Instead, the final enzyme of the electron transport chain gives up a pair of electrons that combine with two hydrogen ions (provided by the hydrogen carriers) and an atom of oxygen to form a water molecule:

$$2e^- + 2H^+ + 1/2\ O_2 \rightarrow H_2O$$

Thus, oxygen is the final electron "carrier." In the absence of oxygen, electrons cannot continue to pass through the electron transport chain, and the aerobic reactions of cellular respiration grind to a halt.

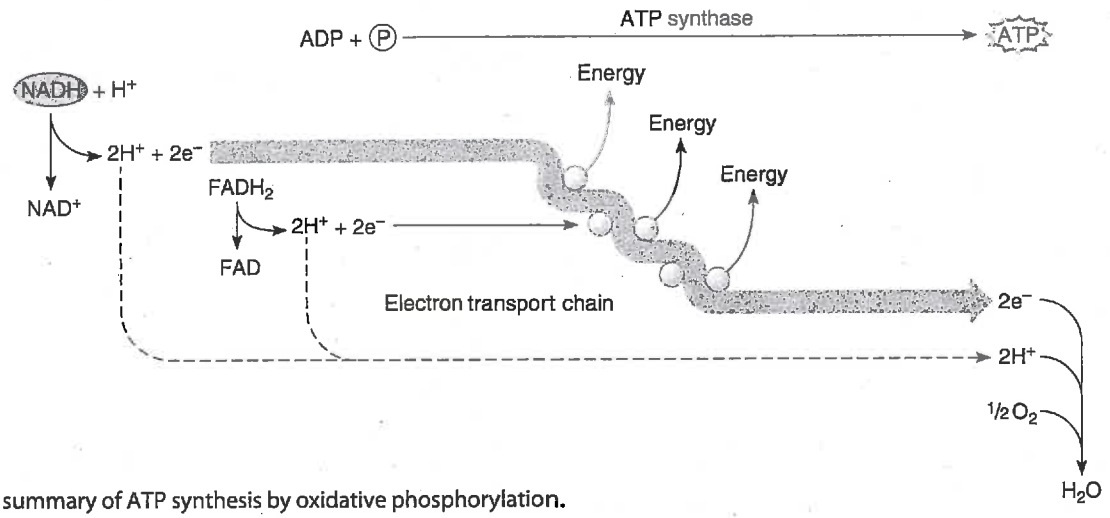


FIGURE 4.12 A summary of ATP synthesis by oxidative phosphorylation.

Figure 4.13 summarizes the steps in glucose metabolism. More detailed descriptions of the reactions of cellular respiration are in Appendix C, pages 944–947.

Cyanide is a deadly poison that halts ATP production in cells. It binds to an iron atom that is part of the enzyme that enables NADH from the citric acid cycle to transfer electrons to oxygen. Cyanide is absorbed through the skin, gastrointestinal tract, and respiratory tract, and exposure can kill in minutes. One source of cyanide is bitter almonds (not the sweet type that people prefer), which produce a compound called amygdalin that an enzyme in the human small intestine breaks down, releasing the poison. Cyanide is encountered in certain industrial processes, including metal plating, gold extraction, and in the raw materials for plastics. Rat poison and fumigants also contain cyanide.

Carbohydrate Storage

Metabolic pathways are usually interconnected in ways that enable certain molecules to enter more than one pathway. For example, carbohydrate molecules from foods may enter catabolic pathways and be used to supply energy, or they may enter anabolic pathways and be stored or react to form some of the twenty different amino acids (fig. 4.14).

Excess glucose in cells may enter anabolic carbohydrate pathways and be linked into storage forms such as glycogen. Most cells can produce glycogen; liver and muscle cells store the greatest amounts. Following a meal, when blood glucose concentration is relatively high, liver cells obtain glucose from the blood and synthesize glycogen. Between meals, when blood glucose concentration is lower, the reaction reverses, and glucose is released into the blood. This mechanism ensures that cells throughout the body have a continual supply of glucose to support cellular respiration.

Glucose can also react to form fat molecules, later deposited in adipose tissue. This happens when a person takes in more carbohydrates than can be stored as glycogen or are required for normal activities. The body has an almost unlimited capacity to perform this type of anabolism, so overeating carbohydrates can cause accumulation of body fat.

This section has considered the metabolism of glucose, although lipids and proteins can also be broken down to release energy for ATP synthesis. In all three cases, the final process is aerobic respiration, and the most common entry point is into the citric acid cycle as acetyl CoA (fig. 4.15). These pathways are described in detail in chapter 18 (pp. 702–704).

PRACTICE

17 State the products of the aerobic reactions.
18 List the products of the citric acid cycle.
19 Explain the function of the electron transport chain.
20 Discuss fates of glucose other than cellular respiration.

4.6 NUCLEIC ACIDS AND PROTEIN SYNTHESIS

Enzymes control the metabolic pathways that enable cells to survive, so cells must have information for producing these specialized proteins. Many other proteins are important in physiology as well, such as blood proteins, the proteins that form muscle and connective tissues, and the antibodies that protect against infection. The information that instructs a cell to synthesize a particular protein is held in the sequence of building blocks of **deoxyribonucleic acid (DNA)**, the genetic material. As we will see later in this chapter, the correspondence between a unit of DNA information and a particular amino acid constitutes the **genetic code** (jĕ-net'ik kōd).

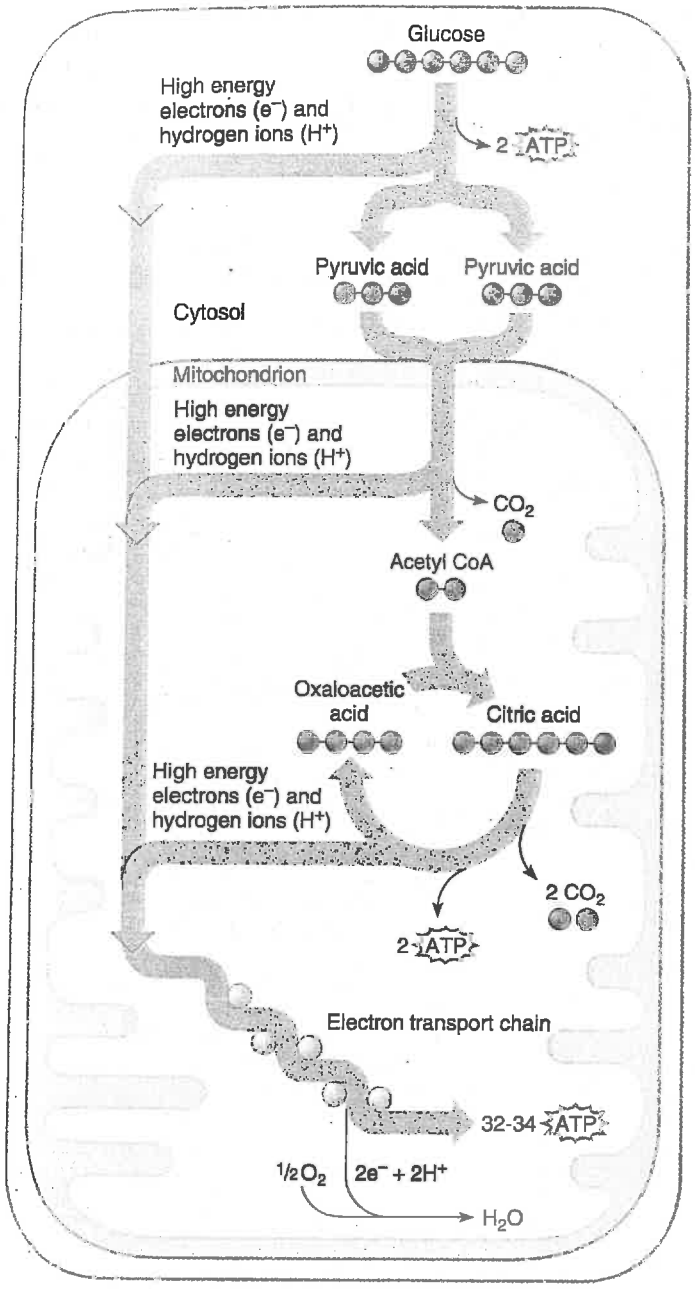


FIGURE 4.13 An overview of aerobic respiration, including the net yield of ATP at each step per molecule of glucose.

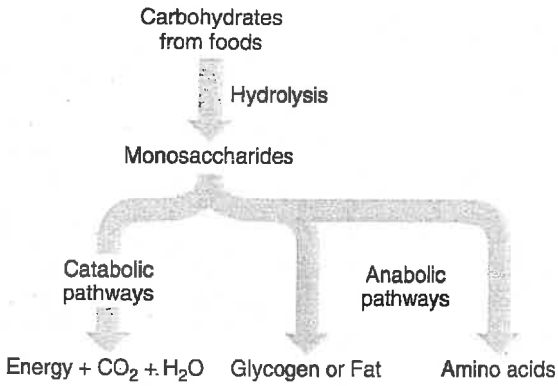


FIGURE 4.14 Hydrolysis breaks down carbohydrates from foods into monosaccharides. The resulting molecules may enter catabolic pathways and be used as energy sources, or they may enter anabolic pathways and be stored as glycogen or fat, or react to yield amino acids.

Genetic Information

Children resemble their parents because of inherited traits, but what passes from parents to a child is genetic information, in the form of DNA molecules from the parents' sex cells. Chromosomes are long molecules of DNA and associated proteins. As an offspring develops, mitosis passes the information in the DNA sequences of the chromosomes to new cells. Genetic information "tells" cells how to construct a great variety of protein molecules, each with a specific function. The portion of a DNA molecule that contains the genetic information for making a particular protein is called a **gene** (jēn). Enzymes control synthesis reactions, so all four groups of organic molecules—proteins, carbohydrates, lipids, and nucleic acids—depend on proteins, and thus require genetic instructions.

RECONNECT
To Chapter 3, Cell Nucleus, page 90.

The complete set of genetic instructions in a cell constitutes the **genome.** The "first draft" of the human genome sequence was announced in June 2000, following nearly fifteen years of discussion and work by thousands of researchers worldwide. Only a small part of the human genome encodes protein. The rest includes many controls over which proteins are produced in a particular cell under particular circumstances, called *gene expression.* Chapter 24 (p. 917) discusses the human genome.

Recall from chapter 2 (p. 68) that nucleotides are the building blocks of nucleic acids. A nucleotide consists of a 5-carbon sugar (ribose or deoxyribose), a phosphate group, and one of several nitrogenous bases (fig. 4.16). DNA and RNA nucleotides form long strands (polynucleotide chains) by alternately joining their sugar and phosphate portions by dehydration synthesis, which provides a "backbone" structure (fig. 4.17).

A DNA molecule consists of two polynucleotide chains, making it double-stranded. The nitrogenous bases project from the sugar-phosphate backbone of one strand and bind, or pair, by hydrogen bonds to the nitrogenous bases of the second strand (fig. 4.18). The resulting structure is somewhat like a ladder, in which the rails represent the sugar and phosphate backbones of the two strands and the rungs represent the paired nitrogenous bases. The sugars forming the two backbones point in opposite directions. For this reason, the two strands are called *antiparallel.*

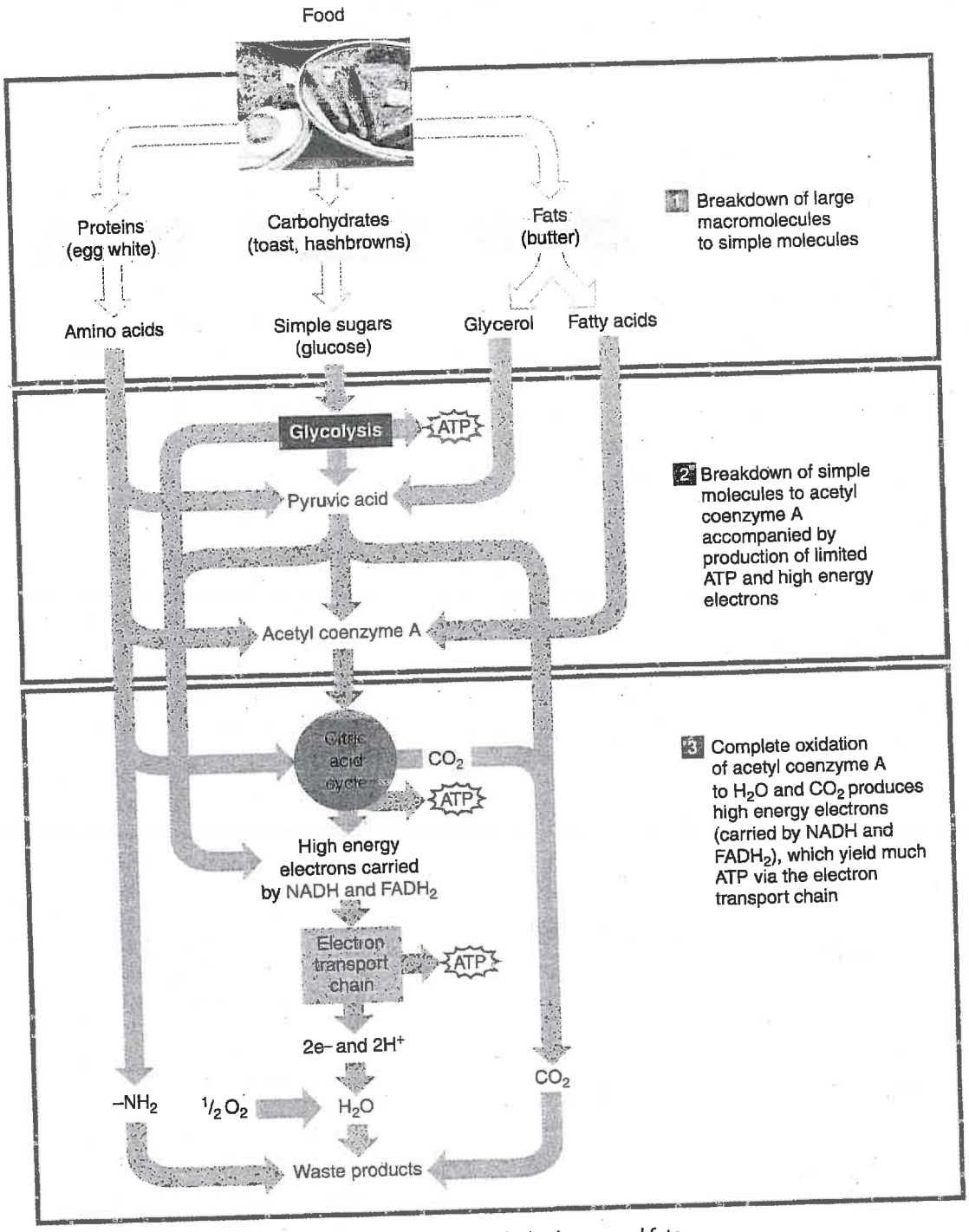


FIGURE 4.15 A summary of the breakdown (catabolism) of proteins, carbohydrates, and fats.

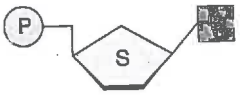


FIGURE 4.16 Each nucleotide of a nucleic acid consists of a 5-carbon sugar (S); a phosphate group (P); and an organic, nitrogenous base (B).

A DNA molecule is sleek and symmetrical because the bases pair in only two combinations, which maintains a constant width of the overall structure. In a DNA nucleotide, the base may be one of four types: adenine (A), thymine (T), cytosine (C), or guanine (G). A and G are **purines** (pu'rēnz), and they consist of two organic ring structures. T and C are **pyrimidines** (pe-rimi-denz), and they have a

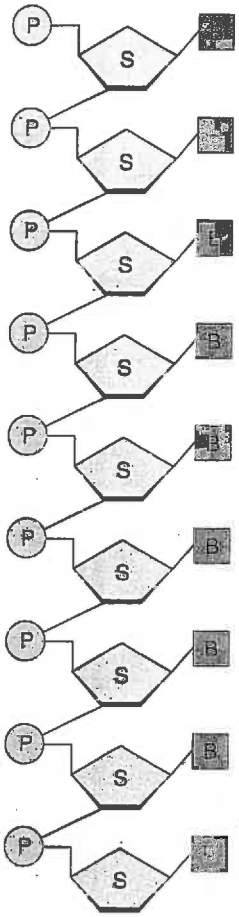


FIGURE 4.17 A polynucleotide chain consists of nucleotides connected by a sugar-phosphate backbone.

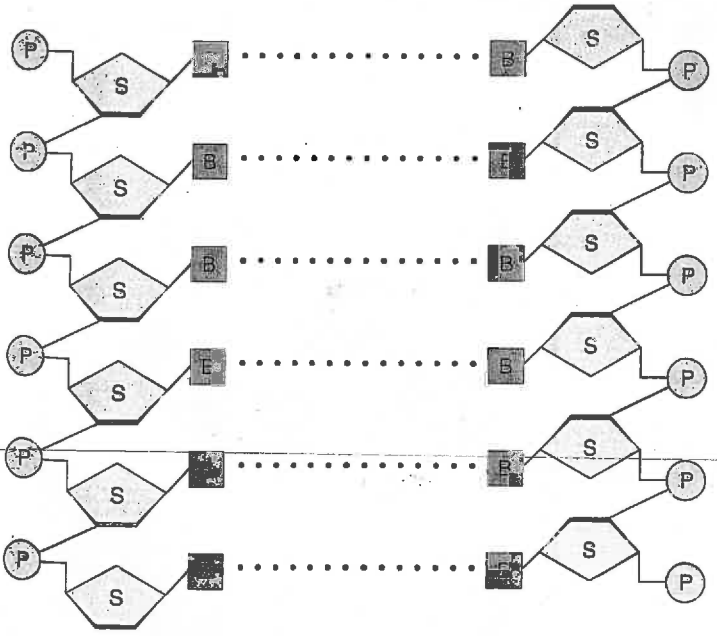


FIGURE 4.18 DNA is double-stranded, consisting of two polynucleotide chains. Hydrogen bonds (dotted lines) hold the nitrogenous bases of one strand to their partners on the other strand. The sugars point in opposite directions—that is, the strands are antiparallel.

single organic ring structure. A binds to T and G binds to C—that is, a purine always binds to a pyrimidine, and this is what establishes the constant width of the DNA molecule. These pairs—A with T, and G with C—are called **complementary base pairs** (fig. 4.19*a*). The sequence of one DNA strand can always be derived from the other by following the "base-pairing rules." If the sequence of one strand of the DNA molecule is G, A, C, T, then the complementary strand's sequence is C, T, G, A.

The double-stranded DNA molecule twists, forming a double helix, (fig. 4.19*b*). The human genome is 3.2 billion DNA bases long, dispersed over the 24 types of chromosomes. A single gene may be thousands or even millions of bases long. In the nucleus, DNA is wound around octets of proteins called *histones* to form chromatin (fig. 4.19*b*). Histones and other molecules come on and off different parts of the genome as some genes are accessed for their information to make proteins and others are silenced. During mitosis chromatin condenses to form chromosomes visible under the microscope (fig. 4.19*c*). Investigators can use DNA sequences to identify individuals (From Science to Technology 4.1). Appendix D, pages 948–949, has more detailed DNA structures.

DNA Replication

When a cell divides, each newly formed cell must have a copy of the original cell's genetic information (DNA) so it will be able to synthesize the proteins necessary to build cellular parts and metabolize. DNA **replication** (re″plĭ-ka′shun) is the process that creates an exact copy of a DNA molecule. It happens during interphase of the cell cycle.

RECONNECT
To Chapter 3, The Cell Cycle, page 100.

As DNA replication begins, hydrogen bonds break between the complementary base pairs of the double strands. Then the strands unwind and separate, exposing unpaired bases. New nucleotides pair with the exposed bases, forming hydrogen bonds. An enzyme, DNA polymerase, catalyzes this base pairing. Enzymes then knit together the new sugar-phosphate backbone. In this way, a new strand of complementary nucleotides extends along each of the old (original) strands. Two complete DNA molecules result, each with one new and one original strand (fig. 4.20). During mitosis, the two DNA molecules that form the two chromatids of each of the chromosomes separate so that one of these DNA molecules passes to each of the new cells.

From Science to Technology 4.2 discusses the polymerase chain reaction (PCR), a method for mass-producing, or amplifying, DNA. PCR has revolutionized biomedical science.

PRACTICE

21 What is the function of DNA?

22 What is the structure of DNA?

23 How does DNA replicate?

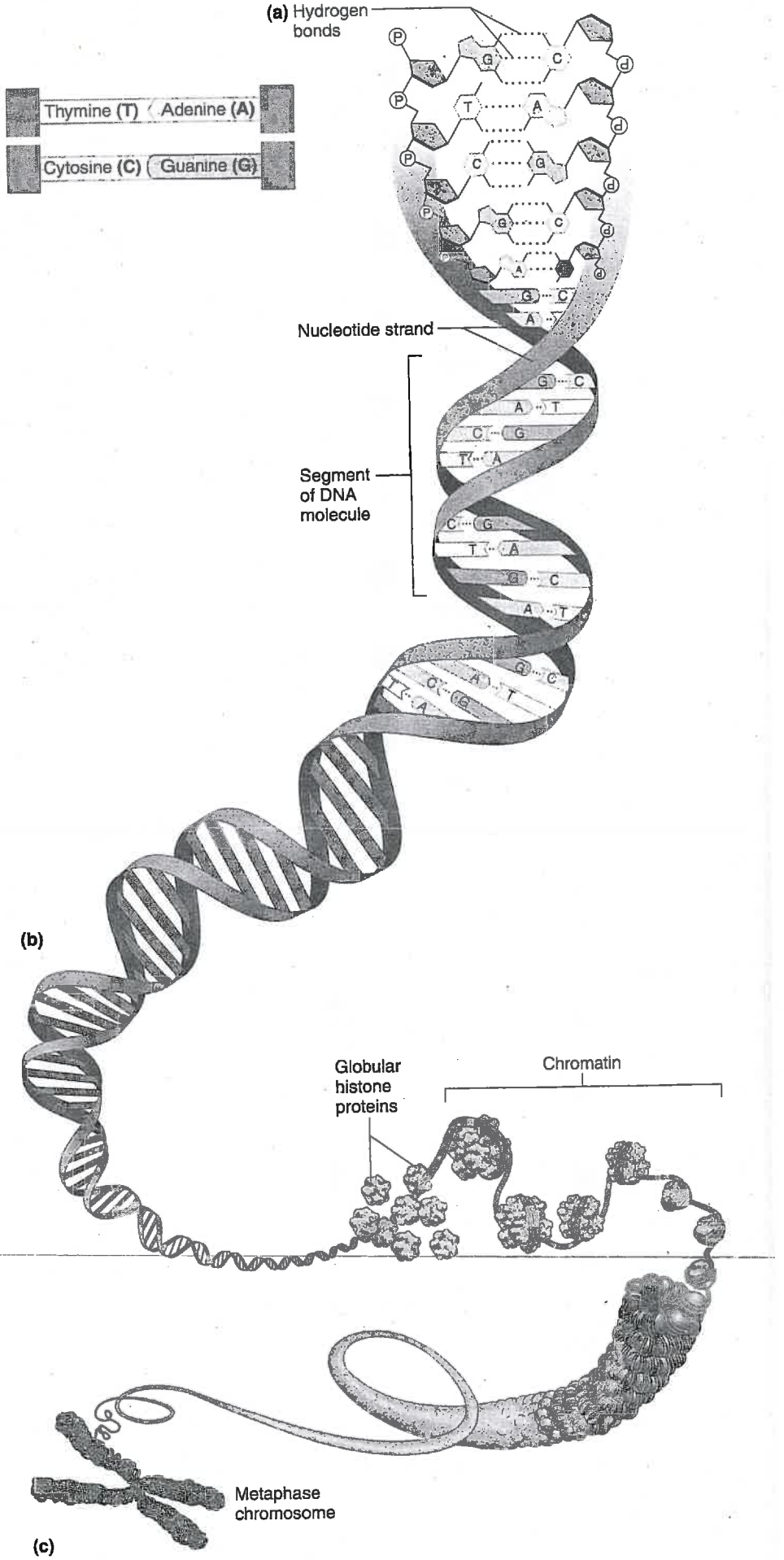
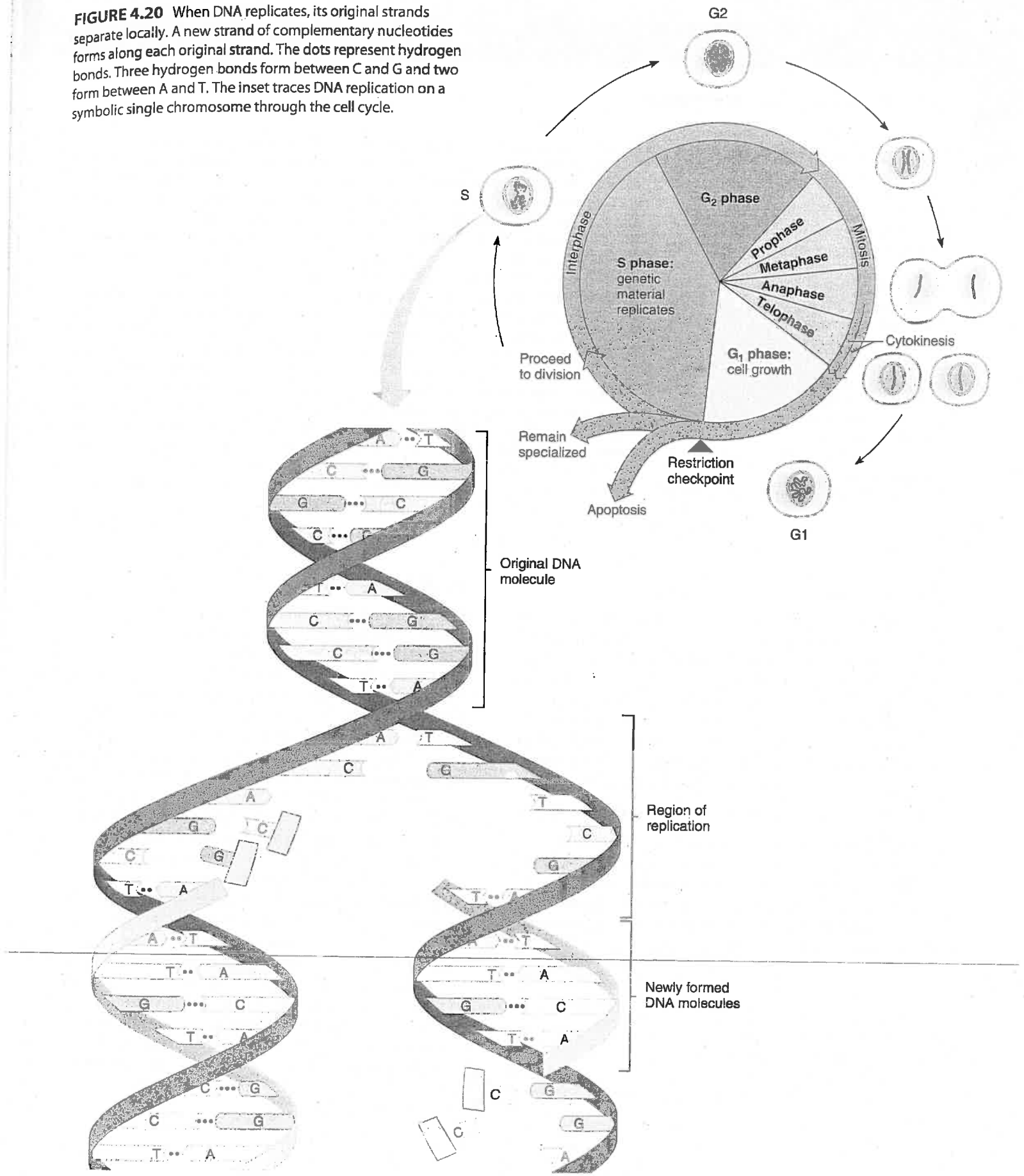


FIGURE 4.19 DNA and chromosome structure. (*a*) The two polynucleotide chains of a DNA molecule point in opposite directions (antiparallel) and are held together by hydrogen bonds between complementary base pairs. (*b*) The molecular "ladder" of a DNA molecule twists into a double helix. (*c*) Histone proteins enable the long double helix to assume a compact form (chromosome) and move when sections of the DNA are accessed for gene expression.

FIGURE 4.20 When DNA replicates, its original strands separate locally. A new strand of complementary nucleotides forms along each original strand. The dots represent hydrogen bonds. Three hydrogen bonds form between C and G and two form between A and T. The inset traces DNA replication on a symbolic single chromosome through the cell cycle.



4.1 FROM SCIENCE TO TECHNOLOGY

DNA Profiling Frees A Prisoner

The human genome sequence differs from person to person because it includes 3.2 billion bits of information. Techniques called DNA profiling (or fingerprinting) compare the most variable parts of the genome among individuals for several purposes—to identify remains at crime scenes or after natural disasters; to confirm or rule out "blood" relationships; and, increasingly, to establish innocence when other types of evidence are questionable. The Innocence Project is a national litigation and public policy organization that provides DNA testing to people who claim that they have been wrongfully convicted. So far the Innocence Project has exonerated more than 200 people. Among them is Josiah Sutton.

Sutton had served four and a half years of a twenty-five-year sentence for rape when DNA profiling established his innocence. He and a friend had become suspects after a woman in Houston identified them as the men who had raped and threatened her with a gun, leaving her in a field. The two young men supplied saliva and blood samples, from which DNA profiles were done and compared to DNA profiles from semen found in the victim and in her car. At the trial, an employee of the crime lab doing the DNA analysis testified that the probability that Sutton's DNA matched that of the evidence by chance was 1 in 694,000—a number so compelling that it led jurors to convict him, even though Sutton did not fit the victim's description of her assailant.

A DNA profile analyzes only 13 parts of the genome, known to vary in most populations. Usually this is sufficient information to rule out a suspect. Using these criteria, Sutton's DNA at first seemed to match the evidence. The problem, though, wasn't in the DNA, but in the population to which it was compared. Although Sutton's pattern may indeed have been very rare in the large population to which it was compared, among black men, it wasn't rare at all—1 in 16 black men have the exact same pattern!

Proclaiming his innocence all along, Sutton had asked right away for an independent DNA test, but was told he couldn't afford one. So while he was in prison, he read voraciously about DNA profiling and again, in a handwritten note, requested retesting. Then he got lucky. Two journalists began investigating the Houston crime laboratory. They sent information on a few cases to a professor of criminology, who immediately saw the errors made in Sutton's DNA analysis, claiming that the test wasn't even of the quality of a middle school science project. Retesting Sutton's DNA, and comparing it to a relevant population, proved his innocence. ■

Genetic Code

Genetic information specifies the correct sequence of amino acids in a polypeptide chain. Each of the twenty different types of amino acids is represented in a DNA molecule by a triplet code, consisting of sequences of three nucleotides. That is, the sequence C, G, T in a DNA strand represents one type of amino acid; the sequence G, C, A represents another type. Other sequences encode instructions for beginning or ending the synthesis of a protein molecule, and for determining which genes are accessed for their information.

The genetic code is said to be universal because all species use the same DNA base triplets to specify the same amino acids. Researchers deciphered the code in the 1960s. When the media mentions an individual's genetic code, they really are referring to the sequence of DNA bases comprising a certain gene or genome—not the genetic code (the correspondence between DNA triplet and amino acid).

DNA molecules are in the nucleus and protein synthesis occurs in the cytoplasm. Because the cell must keep a permanent copy of the genetic instructions, genetic information must get from the nucleus into the cytoplasm for the cell to use it. RNA molecules accomplish this transfer of information.

RNA Molecules

RNA (ribonucleic acid) molecules differ from DNA molecules in several ways. RNA molecules are single-stranded, and their nucleotides have ribose rather than deoxyribose sugar. Like DNA, RNA nucleotides each have one of four nitrogenous bases, but whereas adenine, cytosine, and guanine nucleotides are part of both DNA and RNA, thymine nucleotides are only in DNA. In place of thymine nucleotides, RNA molecules have uracil (U) nucleotides (fig. 4.21 and Appendix D, p. 949). In RNA U pairs with A (fig. 4.22). Different types of RNA have different size ranges and functions. The process of copying DNA information into an RNA sequence is called **transcription** (trans-krip'-shun).

The first step in delivering information from the nucleus to the cytoplasm is the synthesis of **messenger RNA (mRNA)**. RNA nucleotides form complementary base pairs with one of the two strands of DNA that encodes a particular protein. However, just as the words in a sentence must be read in the correct order to make sense, the base sequence of a strand of DNA must be "read" in the correct direction and from the correct starting point. Furthermore, only one of the two antiparallel strands of DNA contains the genetic message. An enzyme called RNA polymerase recognizes the correct DNA strand and the right direction for RNA synthesis. The "sentence" always begins with the mRNA base sequence AUG (fig. 4.23).

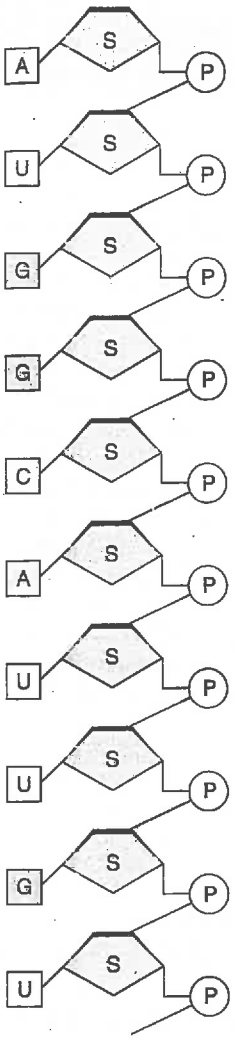


FIGURE 4.21 RNA differs from DNA in that it is single-stranded, contains ribose rather than deoxyribose, and has uracil (U) rather than thymine (T) as one of its four bases.

In mRNA synthesis, RNA polymerase binds to a promoter, a DNA base sequence that begins a gene. Then a section of the double-stranded DNA unwinds and pulls apart, exposing a portion of the gene. RNA polymerase moves along the strand, exposing other portions of the gene. At the same time, a molecule of mRNA forms as RNA nucleotides complementary to those along the DNA strand are strung together. For example, if the sequence of DNA bases is TACCCGAGG, the complementary bases in the developing mRNA molecule will be AUGGGCUCC, as figure 4.23 shows. For different genes, different strands of the DNA molecule may be used to manufacture RNA.

RNA polymerase continues to move along the DNA strand, exposing portions of the gene, until it reaches a special DNA base sequence (termination signal) that signals the end of the gene. At this point, the RNA polymerase releases the newly formed mRNA molecule and leaves the DNA. The DNA then rewinds and assumes its previous double helix structure.

Each amino acid in the protein to be synthesized was originally represented by a series of three bases in DNA.

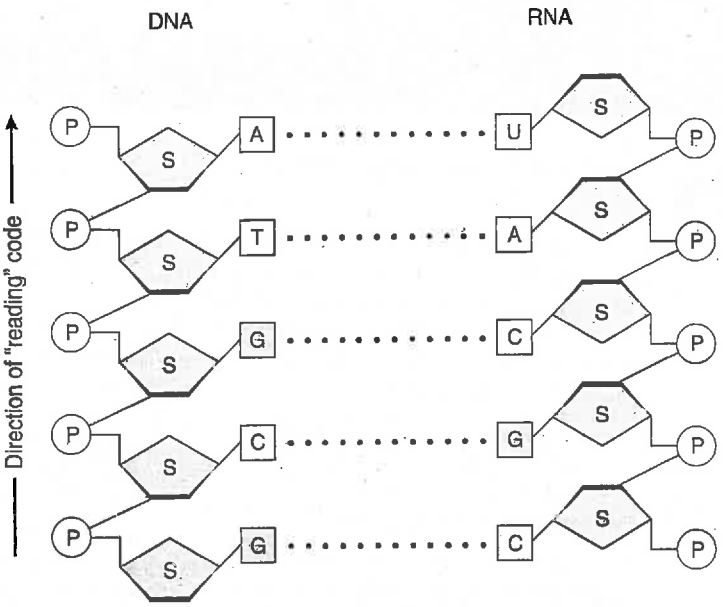


FIGURE 4.22 Transcription of RNA from DNA. When an RNA molecule is synthesized beside a strand of DNA, complementary nucleotides bond as in a double-stranded DNA molecule, with one exception: RNA contains uracil nucleotides (U) in place of thymine nucleotides (T).

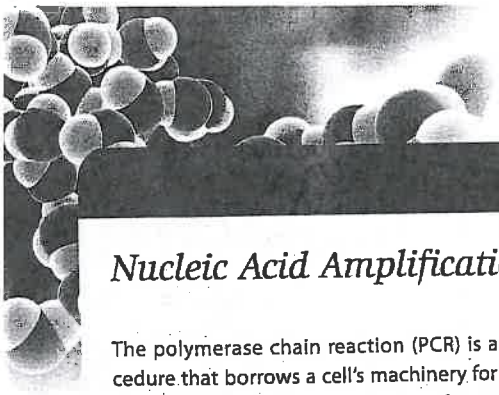
Those amino acids, in the proper order, are now represented by a series of three base sequences, called **codons,** (ko'donz) in mRNA. To complete protein synthesis, mRNA must leave the nucleus and associate with a ribosome. There, the series of codons of the mRNA is translated from the "language" of nucleic acids to the "language" of amino acids. This process is fittingly called **translation** (see fig. 4.23). Table 4.1 compares DNA and RNA molecules.

Protein Synthesis

Synthesizing a protein molecule requires that the specified amino acid building blocks in the cytoplasm align in the proper sequence along an mRNA. A second type of RNA molecule, transcribed in the nucleus and called **transfer RNA (tRNA),** aligns amino acids in a way that enables them to bond to each other. A tRNA molecule consists of seventy to eighty nucleotides and has a complex three-dimensional shape, somewhat like a cloverleaf. The two ends of the tRNA molecule are important for the "connector" function (see fig. 4.23).

At one end, each tRNA molecule is a binding site for a particular amino acid. At least one type of tRNA specifies each of the twenty amino acids. An amino acid must be activated for a tRNA to pick it up. Special enzymes catalyze this step. ATP provides the energy for an amino acid and its tRNA to bond (fig. 4.24).

The other end of each transfer RNA molecule includes a specific three nucleotide sequence, called the **anticodon,** unique to that type of tRNA. An anticodon bonds only to the complementary mRNA codon. In this way, the appropriate tRNA carries its amino acid to the correct place in the mRNA sequence (fig. 4.24).



4.2 FROM SCIENCE TO TECHNOLOGY

Nucleic Acid Amplification

The polymerase chain reaction (PCR) is a procedure that borrows a cell's machinery for DNA replication, making many copies of a gene of interest. Developed in 1983, PCR was the first of several technologies called nucleic acid amplification. Starting materials for PCR include:

- two types of short DNA pieces known to bracket the gene of interest, called primers
- a large supply of DNA bases
- the enzymes that replicate DNA

Here's how it works. First, heat is used to separate the two strands of the target DNA—such as bacterial DNA in a body fluid sample from a person who has symptoms of an infection. Next, the temperature is lowered, and the two short DNA primers are added. The primers complementary base pair to the separated target strands. The third step adds DNA polymerase and bases. The DNA polymerase adds bases to the primers and builds a sequence complementary to the target sequence. The newly synthesized strands then act as templates in the next round of replication, which begins by raising the temperature. All of this is done in an automated device called a thermal cycler that controls the key temperature changes. The DNA polymerase can withstand the temperature shifts because it comes from a bacterium that lives in hot springs.

The pieces of DNA exponentially accumulate. The number of amplified pieces of DNA equals 2^n where n equals the number of temperature cycles. After just twenty cycles, 1 million copies of the original sequence are in the test tube. PCR has had many diverse applications, from detecting moose meat in hamburger to analysis of insect larvae in decomposing human corpses.

PCR's greatest strength is that it works on crude samples of rare and short DNA sequences, such as a bit of brain tissue on the bumper of a car, which in one criminal case led to identification of a missing person. PCR's greatest weakness, ironically, is its exquisite sensitivity. A blood sample submitted for diagnosis of an infection contaminated by leftover DNA from a previous run, or a stray eyelash dropped from the person running the reaction, can yield a false positive result. The technique is also limited in that a user must know the sequence to be amplified and that mutations can sometimes occur in the amplified DNA not present in the source DNA.

The invention of PCR inspired other nucleic acid amplification technologies. One, which copies DNA into RNA and then amplifies the RNA, does not require temperature shifts and produces 100 to 1,000 copies per cycle, compared to PCR's doubling. ■

The genetic code specifies more than enough information. Although only twenty types of amino acids need be encoded, the four types of bases can form sixty-four different mRNA codons. Therefore, some amino acids correspond to more than one codon (table 4.2). Three of the codons do not have a corresponding tRNA. They provide a "stop" signal, indicating the end of protein synthesis, much like the period at the end of this sentence. Sixty-one different tRNAs are specific for the remaining sixty-one codons, which means that more than one type of tRNA can correspond to the same amino acid type.

The binding of tRNA and mRNA occurs in close association with a ribosome. A ribosome is a tiny particle of two unequal-sized subunits composed of **ribosomal RNA (rRNA)** and protein molecules. The smaller subunit of a ribosome binds to a molecule of mRNA near the first codon. A tRNA with the complementary anticodon brings its attached amino acid into position, temporarily joining to the ribosome. A second tRNA, complementary to the second mRNA codon, then binds (with its activated amino acid) to an adjacent site on the ribosome. The first tRNA molecule releases its amino acid, providing the energy for a peptide bond to form between the two amino acids (see fig. 4.24). This process repeats as the ribosome moves along the mRNA, adding amino acids one at a time to the extending polypeptide chain. The enzymatic activity necessary for bonding of the amino acids comes from ribosomal proteins and some RNA molecules (ribozymes) in the larger subunit of the ribosome. This subunit also holds the growing chain of amino acids.

RECONNECT
To Chapter 3, A Composite Cell, page 82.

Protein synthesis is economical. A molecule of mRNA usually associates with several ribosomes at the same time. Thus, several copies of that protein, each in a different stage of formation, may be present at any given moment. As the polypeptide forms, proteins called *chaperones* fold it into its unique shape, and when the process is completed, the polypeptide is released as a separate functional molecule. The tRNA molecules, ribosomes, mRNA, and the enzymes can function repeatedly in protein synthesis.

ATP molecules provide the energy for protein synthesis. A protein may consist of many hundreds of amino acids and the energy from three ATP molecules is required to link each amino acid to the growing chain. This means that a large fraction of a cell's energy supply supports protein synthesis. Table 4.3 summarizes protein synthesis.

The number of molecules of a particular protein that a cell synthesizes is generally proportional to the number of corresponding mRNA molecules. The rate at which mRNA is transcribed from DNA in the nucleus and the rate at which enzymes (ribonucleases) destroy the mRNA in the cytoplasm therefore control protein synthesis.

Proteins called transcription factors activate certain genes, moving aside the surrounding histone proteins to expose the promoter DNA sequences that represent the start of a gene. These actions are called "chromatin remodeling,"

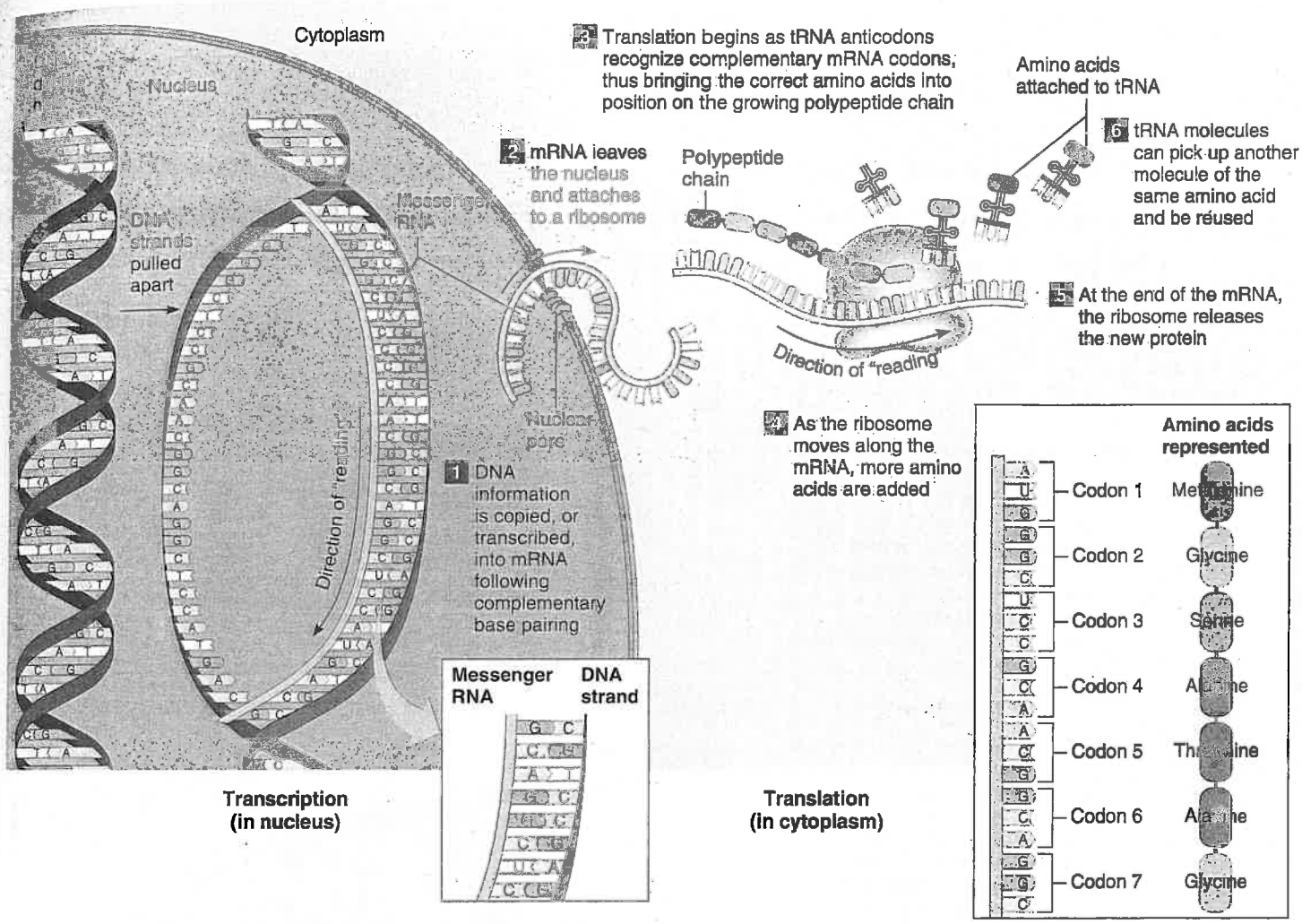


FIGURE 4.23 Protein synthesis. DNA information is transcribed into mRNA, which in turn is translated into a sequence of amino acids. The inset shows some examples of the correspondence between mRNA codons and the specific amino acids that they encode.

TABLE 4.1 | A Comparison of DNA and RNA Molecules

	DNA	RNA
Main location	Part of chromosomes, in nucleus	Cytoplasm
5-carbon sugar	Deoxyribose	Ribose
Basic molecular structure	Double-stranded	Single-stranded
Nitrogenous bases included	Cytosine, guanine, adenine, thymine	Cytosine, guanine, adenine, uracil
Major functions	Contains genetic code for protein synthesis; replicates prior to mitosis	Messenger RNA carries transcribed DNA information to cytoplasm and acts as template for synthesis of protein molecules; transfer RNA carries amino acids to messenger RNA; ribosomal RNA provides structure for ribosomes

and they control which proteins a cell produces and how many copies form under particular conditions. A connective tissue cell might have many mRNAs representing genes that encode the proteins collagen and elastin; a muscle cell would have abundant mRNAs encoding contractile proteins, such as actin and myosin. Extracellular signals such as hormones and growth factors activate transcription factors.

From Science to Technology 4.3 describes another type of transcriptional control—microRNAs.

PRACTICE

24 How is genetic information carried from the nucleus to the cytoplasm?

25 How are protein molecules synthesized?

26 How is gene expression controlled?

1 The transfer RNA molecule for the last amino acid added holds the growing polypeptide chain and is attached to its complementary codon on mRNA.

2 A second tRNA binds complementarily to the next codon, and in doing so brings the next amino acid into position on the ribosome. A peptide bond forms, linking the new amino acid to the growing polypeptide chain.

3 The tRNA molecule that brought the last amino acid to the ribosome is released to the cytoplasm, and will be used again. The ribosome moves to a new position at the next codon on mRNA.

4 A new tRNA complementary to the next codon on mRNA brings the next amino acid to be added to the growing polypeptide chain.

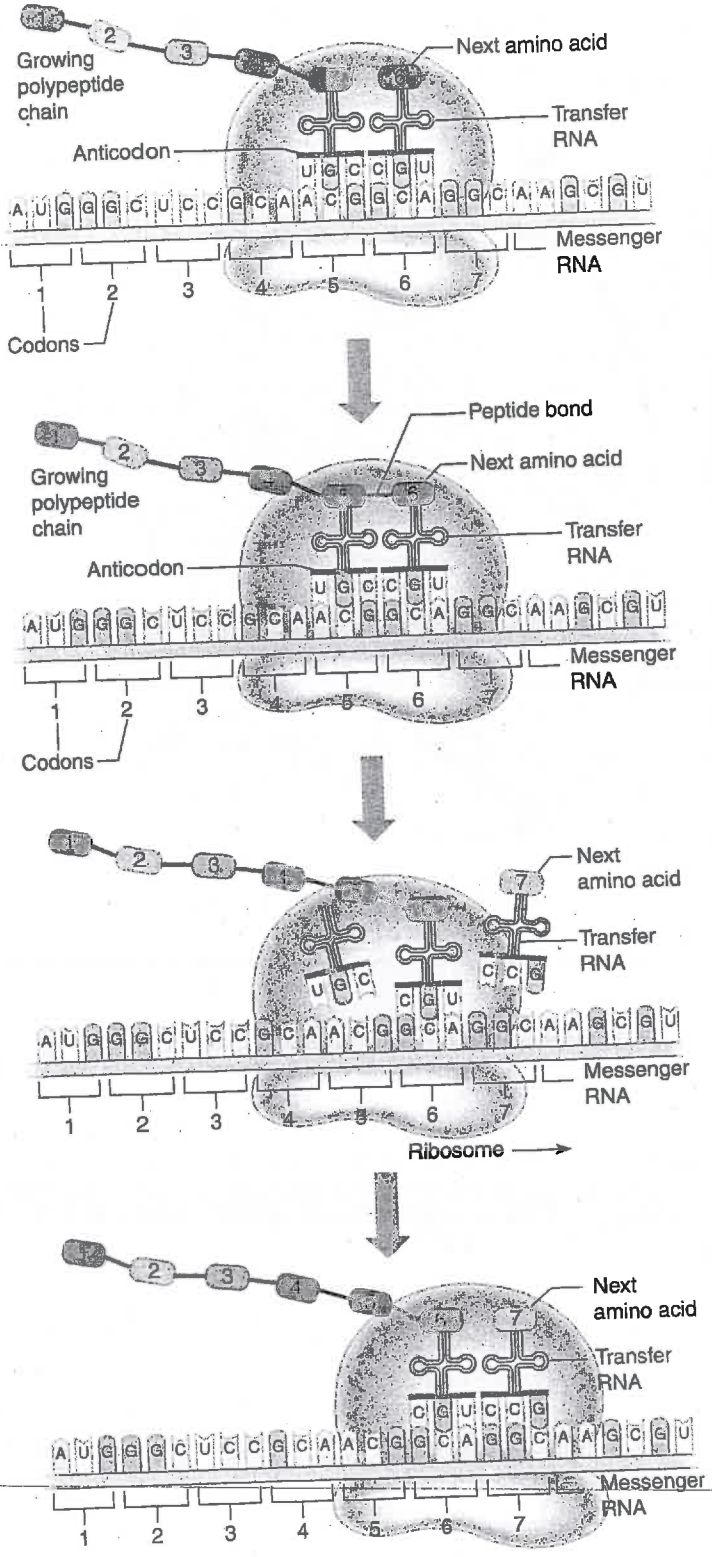


FIGURE 4.24 A closer look at protein synthesis. Molecules of transfer RNA (tRNA) attach to and carry specific amino acids, aligning them in the sequence determined by the codons of mRNA. These amino acids, connected by peptide bonds, form a polypeptide chain of a protein molecule. Protein synthesis occurs on ribosomes.

TABLE **4.2** | Codons (mRNA Three Base Sequences)

First Letter												Third Letter
U	UUU	phenylalanine (phe)	UCU	serine (ser)	UAU	tyrosine (tyr)	UGU	cysteine (cys)	U
	UUC		UCC		UAC		UGC		C
	UUA	leucine (leu)	UCA		(UAA)	STOP	(UGA)	STOP	A
	UUG		UCG		(UAG)	STOP	UGG	tryptophan (trp)	G
C	CUU	leucine (leu)	CCU	proline (pro)	CAU	histidine (his)	CGU	arginine (arg)	U
	CUC		CCC		CAC		CGC		C
	CUA		CCA		CAA	glutamine (gln)	CGA		A
	CUG		CCG		CAG		CGG		G
A	AUU	isoleucine (ile)	ACU	threonine (thr)	AAU	asparagine (asn)	AGU	serine (ser)	U
	AUC		ACC		AAC		AGC		C
	AUA		ACA		AAA	lysine (lys)	AGA	arginine (arg)	A
	(AUG) START methionine (met)		ACG		AAG		AGG		G
G	GUU	valine (val)	GCU	alanine (ala)	GAU	aspartic acid (asp)	GGU	glycine (gly)	U
	GUC		GCC		GAC		GGC		C
	GUA		GCA		GAA	glutamic acid (glu)	GGA		A
	GUG		GCG		GAG		GGG		G

TABLE **4.3** | Protein Synthesis

Transcription (in the Nucleus)

1. RNA polymerase binds to the DNA base sequence of a gene.
2. This enzyme unwinds a portion of the DNA molecule, exposing part of the gene.
3. RNA polymerase moves along one strand of the exposed gene and catalyzes synthesis of an mRNA, whose nucleotides are complementary to those of the strand of the gene.
4. When RNA polymerase reaches the end of the gene, the newly formed mRNA is released.
5. The DNA rewinds and closes the double helix.
6. The mRNA passes through a pore in the nuclear envelope and enters the cytoplasm.

Translation (in the Cytoplasm)

1. A ribosome binds to the mRNA near the codon at the beginning of the messenger strand.
2. A tRNA molecule that has the complementary anticodon brings its amino acid to the ribosome.
3. A second tRNA brings the next amino acid to the ribosome.
4. A peptide bond forms between the two amino acids, and the first tRNA is released.
5. This process is repeated for each codon in the mRNA sequence as the ribosome moves along its length, forming a chain of amino acids.
6. As the chain of amino acids grows, it folds, with the help of chaperone proteins, into the unique conformation of a functional protein molecule.
7. The completed protein molecule (polypeptide) is released. The mRNA molecule, ribosome, and tRNA molecules are recycled.

4.7 CHANGES IN GENETIC INFORMATION

Remarkably, we are more alike than different—human genome sequences are 99.9 percent the same among individuals. The tenth of a percent of the human genome that can vary from person-to-person includes rare DNA sequences that affect health or appearance, as well as common DNA base variations that do not exert any observable effects.

Nature of Mutations

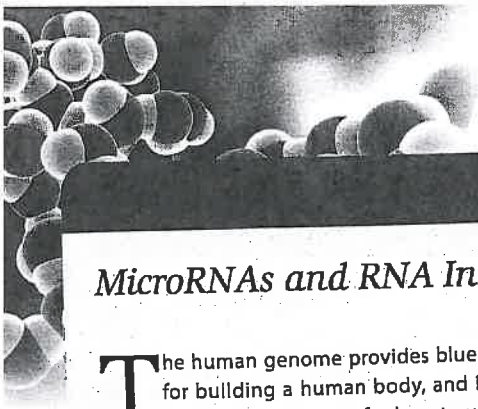
The rare distinctions in DNA sequence that affect how we look or feel are called **mutations** (mu-ta'shunz) More common genetic variants with no detectable effects are called **single nucleotide polymorphisms,** abbreviated SNPs (pronounced "snips"). "Polymorphism" means "many forms."

To visualize the concept of genetic variability, imagine a simplified DNA sequence that is part of a particular genome region:

A A A A A A A A A A A A

A person with a mutation or polymorphism at the fourth base might have any of the following sequences for this portion of the genome, with the differences highlighted:

A A A C A A A A A A A A
A A A T A A A A A A A A
A A A G A A A A A A A A



4.3 FROM SCIENCE TO TECHNOLOGY

MicroRNAs and RNA Interference

The human genome provides blueprints for building a human body, and it also includes instructions for how to use the blueprints. Those instructions are so small—RNA molecules 21 or 22 bases long—that for many years researchers unwittingly threw them out. Today an entire industry is forming to adapt these natural controllers of gene expression, called microRNAs, into diagnostic tests and even new types of treatments for disease.

MicroRNAs belong to a class of RNA molecules called noncoding RNAs, so-named because they were not among the first three major classes of RNA described (mRNA, tRNA, and rRNA). The human genome probably has close to 1,000 microRNAs, about half of which have been discovered. The DNA sequences that encode microRNAs are found in parts of the genome accessed to produce proteins and also in the vast regions that do not encode protein and are less well understood.

Each microRNA binds to parts of the initial control regions (corresponding to DNA promoters) of a particular set of mRNAs, by complementary base pairing. When a microRNA binds a "target" mRNA, it turns off transcription. In this way, a single type of microRNA controls specific sets of genes. In turn, a single type of mRNA can bind several different microRNAs. To analyze these complex interactions, researchers use experiments as well as computational tools (bioinformatics).

Within the patterns of microRNA function may lie clues to developing new ways to fight disease, because these controls of gene expression have stood the test of evolutionary time. The first applications are in cancer, as certain microRNAs are either more or less abundant in cancer cells than in healthy cells of the same type from which the cancer cells formed. Restoring the levels of microRNAs that normally suppress the too-rapid cell cycling of cancer, or blocking production of microRNAs too abundant in cancer, could help to return cells to normal. The first microRNA-based diagnostic tests became available in 2008 and are used to distinguish types of lung cancer and for cancer that has spread and the original tumor cannot be identified by other means.

In a related technology called RNA interference (RNAi), small, synthetic RNA molecules are introduced into cells. They block gene expression in the same manner as the naturally occurring microRNAs. Many companies are developing RNAi-based drugs. The technological challenge is in directing where they affect the genome. ■

If the change affects the person in a noticeable or detectable way and occurs in less than one percent of the population, it is considered a mutation. If there is no detectable change from what is considered normal and the change is seen in more than one percent of the population, it is considered a SNP. These designations, however, are subjective. They depend upon what we can identify and what we consider normal. A more general and traditional use of the term "mutation" is as the mechanism of change in a DNA sequence.

The human genome has millions of SNPs. Association studies look at SNP combinations in populations and attempt to identify patterns found almost exclusively in people with a particular disorder. The correlations between SNP patterns and elevated disease risks can be used to guide clinical decision-making—for example, suggesting which patients might respond to one drug but not another. However, sometimes the associations are statistical flukes that vanish when more data are included. Still, several companies promote SNP-based tests direct-to-consumers on the Internet. These should be approached with caution, because the accuracy of using population-level data to diagnose disease in an individual has not been well-studied.

Another way that people differ in their DNA sequences is by the number of repeats of particular sequences, called copy number variants. Such a repeated sequence may range from only a few DNA bases to millions.

Mutations occur in two general ways—spontaneously or induced. They may happen spontaneously due to the chemical tendency of free nitrogenous bases to exist in two slightly different structures. For extremely short times, a base can be in an unstable form. If, by chance, such an unstable base is inserted into newly forming DNA, an error in sequence will be generated and perpetuated when the strand replicates. Another replication error that can cause mutation is when the existing (parental) DNA strand slips, adding nucleotides to or deleting nucleotides from the sequence.

In contrast to spontaneous mutations are induced mutations, a response to exposure to certain chemicals or radiation. Anything that causes mutation is termed a **mutagen** (mu'tah-jen). A familiar mutagen is ultraviolet radiation, part of sunlight. Prolonged exposure to ultraviolet radiation can form an extra bond between two adjacent thymine DNA bases that are part of the same DNA strand in a skin cell. This extra bond kinks the double helix, causing an incorrect base to be inserted during replication. The cell harboring such a mutation may not be affected, may be so damaged that it dies and peels off, or it may become cancerous. This is how too much sun exposure can cause skin cancer. Mutagens are also found in hair dye, food additives, smoked meats, and flame retardants.

Disease may result from a mutation, whether spontaneous or induced. If the mutation alters the amino acid sequence of the encoded protein so that it malfunctions or isn't produced at all, and health is impaired. For example,

the muscle weakness of Duchenne muscular dystrophy results from a mutation in the gene encoding the protein dystrophin. This protein normally enables muscle cell membranes to withstand the force of contraction. The mutation may be a missing or changed nucleotide base or absence of part or all of the dystrophin gene. Lack of the normal protein causes muscle cells to collapse, and muscles throughout the body weaken and break down. Figure 4.25 shows how the change of one base causes another inherited illness, sickle cell disease.

Although mutations are commonly associated with diseases or otherwise considered abnormal, they also can confer an advantage. The opening vignette to chapter 3 (p. 76) describes one such helpful mutation that protects against HIV infection.

Once DNA changes, producing a mutation or a SNP or copy number variant, the change is transmitted every time the cell in which it originated divides. If that cell is an egg or sperm, then the change is passed to the next generation. We return to this point in the next section.

Protection Against Mutation

Cells detect many mutations and take action to correct the errors. Special "repair enzymes" recognize and remove mismatched nucleotides and fill the resulting gap with the accurate, complementary nucleotides. This mechanism, called the **DNA damage response,** restores the original structure of the double-stranded DNA molecule.

Disorders of the DNA damage response can make life difficult. Xeroderma pigmentosum, for example, causes extreme sun sensitivity. A child with the condition must completely cover up, swathing sunblock on any exposed skin to prevent freckles, sores, and cancer. Special camps and programs allow these children to play outdoors at night, away from the danger of the sun.

The nature of the genetic code protects against mutation, to a degree. Sixty-one codons specify the twenty types of amino acids, and therefore, some amino acids correspond to more than one codon type. Usually, two or three codons specifying the same amino acid differ only in the third base of the codon. A mutation that changes the third codon base can encode the same amino acid. For example, the DNA triplets GGA and GGG each specify the amino acid proline. If a mutation changes the third position of GGA to a G, the amino acid for that position in the encoded protein does not change—it is still proline.

If a mutation alters a base in the second position, the substituted amino acid is often similar in overall shape to the normal one, and the protein is not changed significantly enough to affect its function. This mutation, too, would go unnoticed. (An important exception is the mutation shown in fig. 4.25.) Yet another protection against mutation is that a person has two copies of each chromosome, and therefore of each gene. If one copy is mutated, the other may provide enough of the gene's normal function to maintain health. (This is more complicated for the sex chromosomes, X and Y, discussed in chapter 24, pp. 927–928.)

Timing of a mutation influences effects on health. A mutation in a sperm cell, egg cell, or fertilized ovum is repeated in every cell of the individual. A mutation in an embryo might be devastating because much of the body is still to develop, and many cells inherit the mutation. In contrast, a mutation in a body cell of an adult would most likely have no effect because it would be only one among trillions of cells that do not have the mutation. However, if such a somatic (body cell) mutation confers a faster cell cycle and therefore cells bearing the mutation have a division advantage, cancer can result.

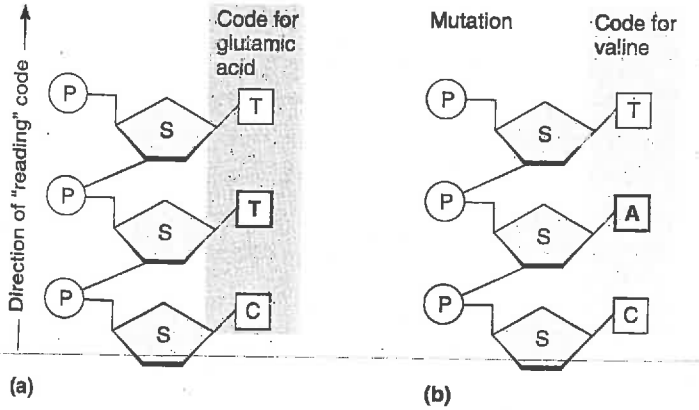


FIGURE 4.25 An example of mutation. (*a*) The DNA code for the amino acid glutamic acid is CTT. (*b*) If something happens to change the first T to A, the DNA code changes to CAT, which specifies the amino acid valine. The resulting mutation, when it occurs in the DNA that encodes the sixth amino acid in a subunit of the protein hemoglobin, causes sickle cell disease. The abnormal hemoglobin bends the red blood cells containing it into sickle shapes. The cells lodge in narrow blood vessels, blocking the circulation and causing great pain.

Inborn Errors of Metabolism

The first part of the chapter discussed enzymes that catalyze the reactions of energy metabolism. Enzymes are also essential to many other reactions and pathways.

A type of disorder called an "inborn error of metabolism" results from inheriting a mutation that alters an enzyme. Such an enzyme block in a biochemical pathway has two general effects: the biochemical that the enzyme normally acts on builds up, and the biochemical resulting from the enzyme's normal action becomes scarce. It is similar to blocking a garden hose: water pressure builds up behind the block, but no water comes out after it.

The biochemical excesses and deficiencies that an inborn error of metabolism triggers can drastically affect health. The specific symptoms depend upon which pathways and biochemicals are affected. Figure 4.26 shows how blocks of different enzymes in one biochemical pathway lead to different sets of symptoms.

Understanding the pathways of metabolism and the many steps and controls of protein synthesis (gene expression) can be daunting. Advances in computational science, however, have vastly improved our ability to tease out the meanings from these complex processes that underlie our physiology. From Science to Technology 4.4 provides a glimpse of this new "systems biology" approach to dissecting the controls of how the human body functions.

PRACTICE

27 Distinguish between a mutation and a SNP.

28 How do mutations arise?

29 How do mutations affect health or appearance?

30 Describe protections against mutation.

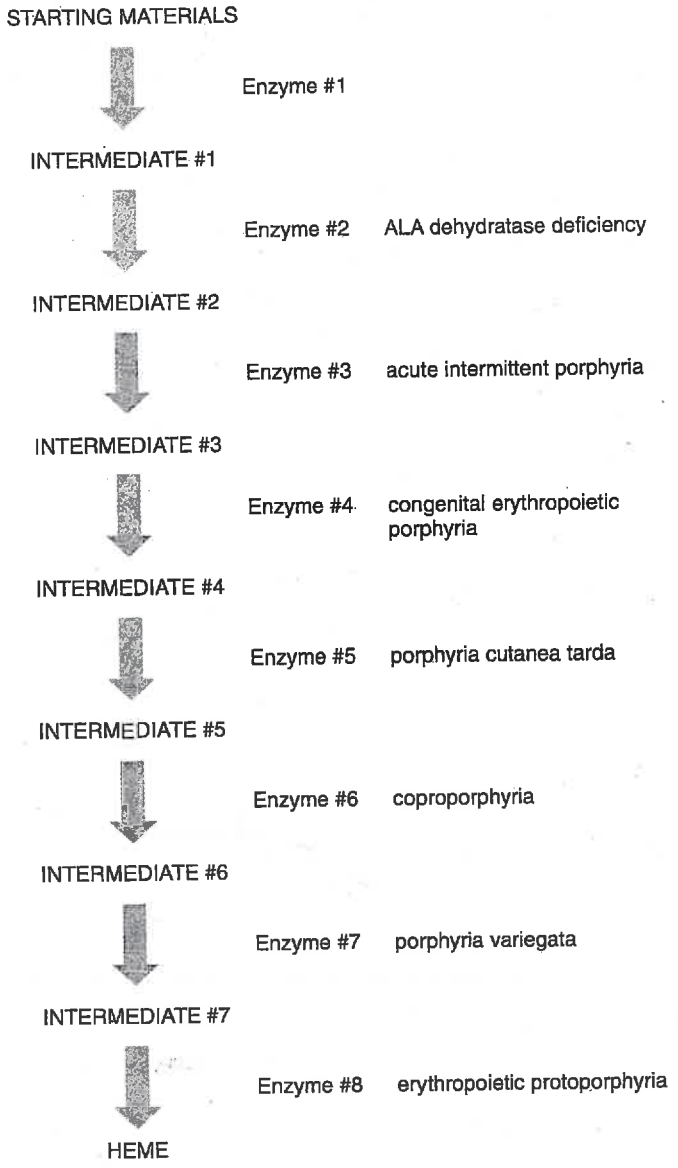


FIGURE 4.26 Seven related but distinct inborn errors of metabolism result from abnormal or missing enzymes that catalyze reactions of the pathway for the synthesis of heme, part of the hemoglobin molecule that is packed into red blood cells. In each disorder, the intermediate biochemical that a deficient enzyme would normally affect builds up. The excess is excreted in the urine or accumulates in blood, feces, or inside red blood cells. Some of the symptoms include reddish teeth, pink urine, excess hair, and photosensitivity.

CHAPTER SUMMARY

4.1 INTRODUCTION (PAGE 115)

A cell continuously carries on metabolic processes. Enzymes are critical to the reactions and pathways of metabolism.

4.2 METABOLIC PROCESSES (PAGE 115)

Metabolic processes include two types of reactions, anabolism and catabolism.

1. Anabolism
   a. Anabolism builds large molecules.
   b. In dehydration synthesis, hydrogen atoms and hydroxyl groups are removed, water forms, and smaller molecules bind by sharing atoms.
   c. Complex carbohydrates are synthesized from monosaccharides, fats are synthesized from glycerol and fatty acids, and proteins are synthesized from amino acids.
2. Catabolism
   a. Catabolism breaks down larger molecules.
   b. In hydrolysis, a water molecule supplies a hydrogen atom to one portion of a molecule and a hydroxyl group to a second portion; the bond between these two portions breaks.
   c. Complex carbohydrates are decomposed into monosaccharides, fats are decomposed into glycerol and fatty acids, and proteins are decomposed into amino acids.

4.3 CONTROL OF METABOLIC REACTIONS (PAGE 117)

Metabolic processes have many steps that occur in a specific sequence and are interconnected. A sequence of enzyme-controlled reactions is a metabolic pathway.