ADVANCED BIOLOGY

WITH EMPHASIS ON BIOCHEMISTRY

Camp Hill, PA
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Advanced Biology with Emphasis on Biochemistry

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CHAPTER 1

Biology—The Study of Life

An artist's depiction of a neuron.

In 1906, Spanish scientist Santiago Ramón y Cajal won the Nobel Prize for showing that the nervous system is composed of individual cells, called neurons, rather than continuous nervous tissue. Just one of many diverse cell types that make up all living things, neurons have the amazing ability to transmit electrical impulses, enabling you to think, feel, and learn.

A neuron consists of three parts: the cell body (soma), the axon, and the dendrites. The soma is the central spherical part of the cell, containing the nucleus. The long arm protruding from the cell body is called the axon. Measuring as long as a meter, the axon sends a signal to stimulate another neuron. Dendrites protrude from the cell body and are able to receive signals from other neurons. A single neuron can make thousands of connections with other neurons through its axon and dendrites. Throughout your study of biology, your neurons will actually form new connections. As you review and master each concept, connections between the corresponding neurons are strengthened. How amazing is it that studying the details of life (or any body of knowledge) actually alters the neural structure of your brain?

OBJECTIVES

After studying this chapter and completing the exercises, you should be able to do each of the following tasks, using supporting terms and principles as necessary.

SECTION 1.1

1. Compare and contrast truth and facts.
2. Define hypothesis, experiment, and theory.
3. Describe each step of the Cycle of Scientific Enterprise.
4. Explain the roles played by magnification, resolution, and contrast in an image produced by a microscope.
5. Compare and contrast the images produced by light microscopes, scanning electron microscopes, and transmission electron microscopes.

SECTION 1.2

6. Describe in paragraphs the six characteristics of life, showing by each one how life exhibits purpose.
7. Explain how cells function as the fundamental building blocks of life.
8. Describe each level of biological organization and how each one incorporates all those beneath it.
9. Define metabolism.
10. Explain the roles of producers, consumers, and decomposers in the process of cycling matter and energy.
11. Describe the general process by which an organism grows and develops over its lifespan.
12. Give a specific example of an organism growing and developing.
13. Compare and contrast asexual reproduction and sexual reproduction.
14. Define DNA and macromolecule.
15. Briefly explain how organisms use and transmit genetic information.
16. Describe an example of an organism responding to a stimulus.
17. Describe homeostasis and give an example of an organism maintaining it.
18. Describe the process by which populations adapt to changing environmental conditions.

SECTION 1.3

19. Distinguish between spontaneous generation, archebiosis, and abiogenesis.
20. Describe Aristotle's thoughts on spontaneous generation and how Redi's experiments contradicted this line of thinking.
21. Explain how Needham's and Spallanzani's experiments supported or weakened the theory of spontaneous generation.
22. Describe Pasteur's experimental setup and his conclusions in detail.
23. Name the four scientists who debated the nature of spontaneous generation during the 1860s and describe their arguments and/or experiments.
24. Using the Cycle of Scientific Enterprise as a guide, trace the development of biogenic theory, explaining how each subsequent experiment either supported or modified the currently accepted theory.

1.1 The Science of Biology

You are about to undertake a great adventure—the study of *biology*. Biology is the science in which the nature of life is studied. However, defining life is not easy. In fact, it is one of the great questions that has been debated throughout scientific history. In order to define life, we first need some scientific skills in our toolboxes. Because biology is a science, it is important that we understand what science is and how science works. In the following sections, we review the nature of science and scientific knowledge.

1.1.1 *Truth and Scientific Facts*

The word science comes from the Latin *scientia*, meaning "knowledge" or "way of knowing." It is important to understand that there are different kinds of knowledge and that scientific knowledge is one kind. Another kind of knowledge deals with truth, which we address first.

Truth can be defined as the way things really are. You can know truth either by direct experience or by revelation from God.[1] God's revelation can be further divided into Special Revelation (the Bible) and General Revelation (creation).

As an example, I can say that the following statement is true: "I have a husband and five children." This is a true statement about me, the author. From my own direct experience, I know that my husband and five children, pictured in Figure 1.1, are the other members of my family. I can count them, I see them every day, and I plainly remember the day I got married and the day each of my children was born. Those who know me personally can also testify to the truth of this statement.

FIGURE 1.1. *One way to know truth is through direct experience, such as my direct knowledge of my family, shown here at the beach.*

A second example of a true statement is: "God made the world." This statement is true because God reveals it to us in the Bible, shown artfully in Figure 1.2. Genesis 1:1 tells us that "In the beginning, God created the heavens and the earth." Scores of other passages in Scripture reinforce the truth that creation is

1 According to classic philosophy, a third way to know truth is by valid reasoning or logic from true premises, but we do not explore this further here.

FIGURE 1.2. *Special Revelation, or God's Word, is one way that God directly reveals truth to us.*

FIGURE 1.3. *General Revelation is another way that God directly reveals truth to us. This image shows a vast number of stars as well as the Milky Way Galaxy (on the left) as viewed from Earth.*

a work of God. In addition to this Special Revelation through Scripture, the lovely photo in Figure 1.3 reminds us that we also understand through General Revelation that God created the world. General Revelation is the way God speaks to all people through what He has made. As we look up into a clear night sky and see the vast array of stars, as in the view of the photograph, we know that a higher power, infinitely more powerful than we are, is the cause behind such beauty. Many other majestic aspects of nature and its study convey this same truth. Indeed, in Romans 1:20 the Bible itself affirms that God speaks to us through General Revelation: "For his invisible attributes, namely his eternal power and divine nature, have been clearly perceived, ever since the creation of the world, in the things that have been made..."

Scientific facts represent a type of knowledge that is different from truth. Illustrated in Figures 1.4 and 1.5, a fact is a statement based on evidence from many experiments or observations that is correct so far as we know. Experiments are carefully designed tests that are meant to give us further information about how the world works. Because we are constantly learning new things about the world, scientific facts can and do change. You may have already studied the Copernican Revolution, the paradigm shift that occurred as Copernicus and Galileo overturned the geocentric theory of the solar system. Before that, everyone accepted the fact that the Sun orbited the Earth. Today, everyone accepts the fact that the Earth orbits the Sun.

As we do research, we come closer and closer to understanding the truth about the nature of the atom, the composition of cells, or the manner by which genetic change occurs over time. Yet, the truth about these subjects is not plainly evident to our everyday experience, and so the only way we can learn about them is through experiment and observation. Only God knows the whole truth about every aspect of his creation. We can only discover scientific facts that are correct so far as we know, and seek to account for the facts by the scientific theories we develop. As time progresses, hopefully these facts—and

FIGURE 1.4. *Experiments are one way that we obtain scientific facts. This picture shows a scientist using a micropipette, an instrument designed to transfer tiny, precisely measured volumes of liquid.*

FIGURE 1.5. *Observations are another way we obtain scientific facts. Scientists carefully watch and measure organisms in their natural environments. They may also collect samples for further testing.*

the theories that explain them—move closer and closer to the truth. Yet, as limited humans, there will always be the potential for adjustments to our knowledge as we investigate the world.

1.1.2 *Theories, Hypotheses, and Experiments*

A *theory* is a mental model or representation that accounts for a large number of scientific facts in an organized way. A theory is judged to be successful when it is repeatedly tested and shown to be consistent with the current body of facts (that are correct so far as we know). If new facts are discovered that do not support a theory, the theory must be reevaluated or revised. The main goal of science is to develop robust and successful theories. To quote textbook author John D. Mays, "Theories are the glory of science." Our goal as scientists is to build successful mental models that accurately describe the way the world works.

You may have heard the word *hypothesis* defined simply as a guess as to the outcome of a test or experiment. However, a hypothesis is not a random guess. Hypotheses are informed predictions, based on a particular scientific theory. Hypotheses are tested and supported (or not supported) by observations and experiments. The results of these tests strengthen or weaken the theories on which the hypotheses are based.

1.1.3 *The Cycle of Scientific Enterprise*

The interplay between facts, theories, hypotheses, and experiments is evident in a diagram of the Cycle of Scientific Enterprise, shown in Figure 1.6. Currently known scientific facts are gathered together as part of a cohesive theory that explains most or all of these scientific facts. A widely accepted theory may be understood as our best current explanation for a body of data (scientific facts). The theoretical understanding of the natural world then enables scientists to make predictions about what would happen in as-yet untested circumstances. As noted above, these informed predictions are called hypotheses. A hypothesis is tested by an experiment. The experiment provides evidence that either supports or does not support the hypothesis. If supported, the theory is strengthened. If the hypothesis is not supported, further tests must be done, perhaps with revised experimental methods. If the experiments continue to fail to support a hypothesis, then the theory is weakened and must be reevaluated. If enough evidence challenging a theory is collected, then a revised theory may be needed. Occasionally, a theory must be thrown out altogether and replaced. As time progresses, the cycle proceeds on and on, hopefully giving us scientific facts and theories that are closer and closer to the truth about reality.

We are making a subtle point here about scientific knowledge that needs to be repeated and emphasized. Scientific theories are models. A widely accepted theory is the scientific *best explanation*. But in principle, theories are provisional; they are always subject to change as new information becomes known.

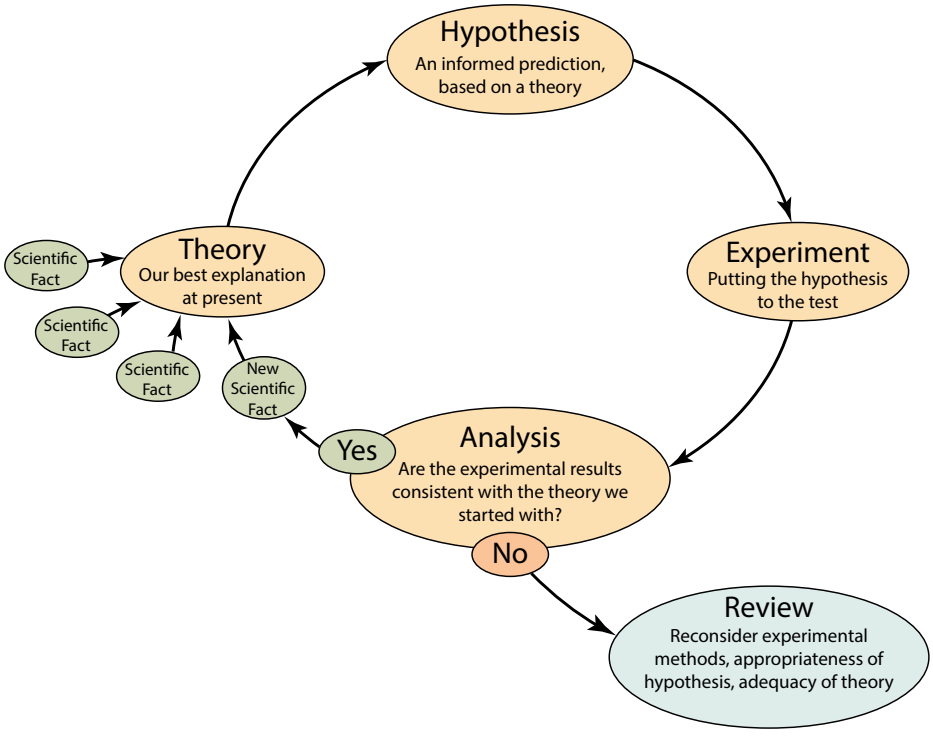


FIGURE 1.6. *The Cycle of Scientific Enterprise.*

Thus, theories are not truth claims about nature. In fact, since we are not God, we do not know the actual, whole truth about nature. So we continue to explore and learn more about the nature of reality indefinitely, continuing the Cycle of Scientific Enterprise. All scientists agree (and hope) that as we learn more and more, our theories grow closer and closer to the actual truth. If we actually do hit on the truth, we have no way of knowing it. All we know is that we have a theory that repeatedly produces hypotheses that are supported by experiment and observation.

As a quick example, consider the atomic theory that all matter is composed of atoms. Is this the truth? We do not know. We do know that this theory has stood up for over 200 years under the most rigorous and sophisticated tests scientists have thought to devise, so we are pretty confident in the claim that matter is made of atoms. Most of us probably believe the claim to be true. But there could come a time when we discover that matter only appears to be composed of atoms and that something else is going on—that matter is composed of strings or loops or springs or has some crazy structure we have never even imagined.

You use the Cycle of Scientific Enterprise in your everyday experience. As an example, imagine you are home alone and you hear a loud chirping noise. Your perception of that noise is an observation you seek to explain. The theory you must work

with includes the facts that there are several smoke detectors in your house capable of making such a sound, that a detector only chirps when the battery needs to be replaced, and that it has been a while since you changed any batteries on these detectors. So you form the hypothesis that one of them has a low battery and is alerting you. One by one, you inspect the battery-life indicator on each detector; this is the experiment that tests your hypothesis. All the batteries seem to be fine, so your hypothesis is not supported by the evidence. Assuming that your battery tests are correct, this experimental result indicates that your theory may need revising. The detector chirps again. Since no detectors indicate a dead battery, you must now form a new hypothesis. Perhaps there is an explanation for a chirping detector other than a weak battery? You go downstairs, following the sound of the chirp. This leads you to the carbon monoxide detector. You read the side label on the detector and realize that the chirping sound indicates a moderate level of carbon monoxide in your house. A new fact has been revealed, and your theory revised accordingly. With the revised theory, you act quickly to open windows, get outside, and call the fire department.

FIGURE 1.7. *A scientist presents the results of his experiments to a group of other scientists. Great progress is made as scientists engage with and build upon one another's work.*

In the biological sciences, it is often the case for knowledge to increase from experimental results without the prior formation of a hypothesis. For example, a molecular biologist may run a *screening experiment* in which a known protein X is incubated with whole-cell *lysate*[2] to find out what other proteins will bond to protein X. If binding partners are found, the biologist can then investigate further the nature and purpose of the binding interaction. This example shows that not all experiments are designed to address hypotheses; some are done in the spirit of "going fishing" or "let's see what happens."

In the scientific world, the Cycle of Scientific Enterprise repeats itself as many thousands of scientists conduct research, publish papers, and engage with one another's work, as illustrated in Figure 1.7. Truly, science is an exciting, ever-changing field, with new discoveries made every day, all over the world. As we continue, let's now examine some of the instruments biologists use to make these important discoveries.

1.1.4 *Instruments and Measurement*

The study of biology relies heavily on a number of specialized instruments and techniques. One of the most important of these instruments is the microscope, which enables one to see organisms too small to be perceived by the naked eye (*microorganisms*).

Magnifying devices have been part of recorded history since the time of the ancient Greeks (circa 400 BC). These simple devices consist of a single convex lens that bends light, creating a magnified image of the object being viewed. No doubt

2 Lysis is the process of breaking down a cellular membrane. A fluid containing the contents of lysed cells is called a lysate.

you have used a magnifying glass to observe objects more closely, as illustrated in Figure 1.8.

FIGURE 1.8. *Magnifying glasses bend light so that objects appear to be larger than they really are. The magnification of a simple lens such as a magnifying glass is limited to about 2×.*

However, the *magnification* of these simple devices is limited to producing an image appearing about 2× larger than the object's actual size. Magnification is defined as the ratio of an image size to the object's actual size.

The first *compound light microscopes* were invented in the late 1500s in Holland. These microscopes were the first to use two lenses in order to achieve higher magnification than that of a single lens. The basic compound microscope works the same way a simple telescope does: a telescope makes a distant object that appears small appear larger, while a microscope makes an object that really is small appear larger. A typical compound light microscope, similar to what you might see in a biology classroom, is shown in Figure 1.9. This instrument allows you to see organisms at varying magnifications, usually in the range of 40–1000×.

FIGURE 1.9. *Compound light microscope. Instruments such as this provide magnifications of 40–1000×.*

Despite the higher magnification of the early compound light microscopes, their resolution was quite limited. *Resolution* is a measure of how clearly the image of an object appears. Mathematically, it is the measure of the minimum distance that two objects can be separated and still be viewed as distinct objects. If you wear corrective lenses, you may already be familiar with this concept. Without your contacts or glasses on, two distant objects near one another may appear to be blurry or fuzzy, melding into a single unclear image. However, with your contacts on you see two separate sharply focused objects.

The first light microscope to achieve both high resolution and magnification was that of Dutch scientist Antonie van Leeuwenhoek in the 1670s. This revolutionary breakthrough opened up the world of microorganisms—previously unknown to mankind.

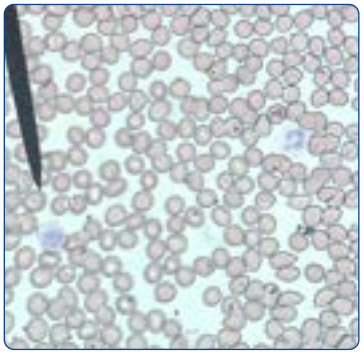


FIGURE 1.10. *A light microscope image of human blood, stained with a special dye to increase contrast. Most of the gray-colored cells are red blood cells, which do not have a nucleus. On the left and right center of the image, you can see two purple-stained white blood cells. The nucleus of these two cells is visible, though not in much detail.*

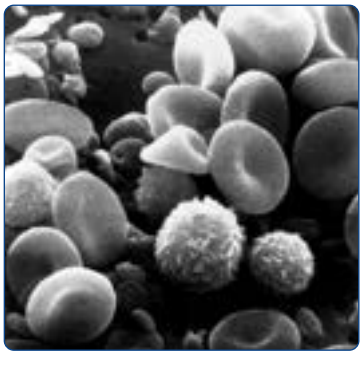


FIGURE 1.11. *An image of human blood using an SEM. The donut-shaped cells are red blood cells, the cells that carry oxygen to each cell in the body. The rounder, large cells with projections on the surface are white blood cells. The tiny objects are platelets—cell fragments involved in blood clotting whenever there is an injury.*

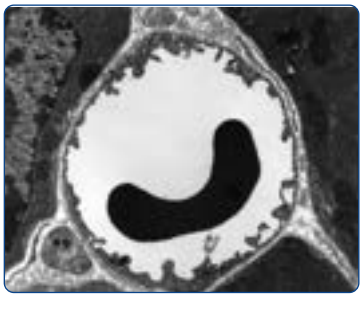


FIGURE 1.12. *A transmission electron micrograph (TEM) of a red blood cell in a capillary (cross-section). The small diameter of a capillary forces red blood cells to pass through in single-file.*

While the light microscope is incredibly useful for viewing cells and microorganisms, its resolution is limited to about 200 nm (nanometers), or 2.0×10^{-7} m. This limit is due to the wavelength range of visible light (400–700 nm), which limits the minimum distance between objects a microscope can clearly resolve.

Because of this resolution limit, the inner workings of the cell are too small to see with a light microscope. To remedy this problem, in the 1930s physicists Ernst Ruska and Max Knoll constructed an *electron microscope* with a resolution that far exceeds that of the light microscope. This type of microscope uses a beam of electrons, rather than light, to generate an image of a sample. In a previous science class, you may have learned that electrons exhibit both particle and wave behaviors (just as light does). The electron microscope exploits the wave nature of the electron, which exhibits wavelengths in the picometer (10^{-12} m) range. The electrons are blasted at a sample at high speed. The interactions between the electrons and the atoms in the sample produce signals that are used to construct an image of the sample. The smaller wavelength of electrons (about 1000× shorter than the wavelengths of visible light) means that much higher resolution can be achieved—down to about 2 nm—and thus much smaller structures can be imaged.

Figure 1.10 shows an image generated by a light microscope. This sample of human blood was stained with a special dye to increase contrast, in other words, to make the cells stand out from the background. Though the boundaries of the cells are clearly visible, other features are not. Figure 1.11 shows an image of human blood produced by a *scanning electron microscope* (SEM). Note that one can watch living things under a light microscope, but only non-living things can be imaged in an SEM.

Figure 1.12 shows an image produced by a *transmission electron microscope* (TEM). In this picture, the dark crescent shape is the cross-section of a single red blood cell, traveling through a capillary. Capillaries are tiny blood vessels where oxygen delivery and waste pickup occur throughout the body.[3] We are looking at a cross-section of this capillary, as if we cut a thin slice of the tube and look at it end-on. That is why the capillary looks like a light-colored circle, rather than a long tube. Notice the rich detail, which enables the study of the surrounding cells (capillary walls are only one cell thick). Multiple organelles (small parts of cells that carry out specific functions) are visible within these surrounding cells. In contrast to three-dimensional SEM images, TEM images are two-dimensional. However, their high magnification and resolution allows for detailed images of organelles within cells, making the TEM an extremely powerful tool in biology.

In addition to light and electron microscopes, there is a host of other instruments and techniques that are useful to bi-

3 Waste products include carbon dioxide and various nitrogen compounds produced by cellular metabolism.

ologists. We address some of these experimental techniques in later chapters.

Finally, it is important that we are able to measure lengths and other parameters in order to study living things accurately. In the United States, we commonly use units such as miles, inches, and feet. But scientists all over the world use the *SI* or *metric system* of units. In this text, we assume you are familiar with this system. However, if you need information about the metric system, you can find it in Appendix A. Table 1.1 shows the metric prefixes commonly used in biology. You should commit these to memory, if you haven't already done so.

The symbol μ is the Greek letter mu, and merits some comment. We pronounce μm "micrometer" (MY-kro-mee-ter), which is not to be confused with a tool for making small measurements called a micrometer (my-KROM-it-er). However, in biology, this measurement is so common that the micrometer has a special nickname—the micron. In fact, the symbol μ is sometimes used by itself to mean a micron (equivalent to micrometer). Therefore 1 μ (one micron) is the equivalent to 1 μm (one micrometer, which can also be read as one micron).

Prefix	Abbreviation	Mathematical Equivalent	Level of Biological Organization	Example Application
kilo	k	10^3	biosphere	circumference of earth, ~10^4 km
centi	c	10^{-2}	organism	average human height, ~180 cm
			organ	width of human heart, ~10 cm
milli	m	10^{-3}	tissue	width of largest human vein, ~24 mm
micro	μ	10^{-6}	cell	cell, ~5–120 μm
nano	n	10^{-9}	biomolecule	biomolecule, ~10–1000 nm
angstrom	Å	10^{-10}	molecule	average length of a chemical bond, ~1 Å
pico	p	10^{-12}	atom	typical atomic width, ~100 pm

TABLE 1.1. *Metric prefixes commonly used in biology. Note: The angstrom is a non-SI unit of measure, not a metric prefix.*

Another unit of measure commonly used in biochemistry is the angstrom. An angstrom is equivalent to one ten billionth of a meter (10^{-10} m) or one tenth of a nanometer (0.1 nm). The angstrom is not an official unit in the SI system, but it is a handy unit of measurement because many individual atoms and chemical bond lengths are about an angstrom wide. This unit of measure is named after Swedish spectroscopist Anders Jonas Ångström (1814–1874).

1.2 What Is Life?

1.2.1 *Life vs. Non-life*

Now that we have discussed what science is, how science works, and some of the important tools and measurements that biologists use, it is time to explore the nature of life.

"Is it alive?" is a question that even the youngest of children ponder. My 5-year-old daughter recently asked me, "Are earthquakes alive?" It was a good question, and not only because we live in Southern California where earthquakes are common. Earthquakes move, can cause massive damage, and act without apparent cause.[4] Nonetheless, earthquakes are not alive.

So then, what is it that makes something alive? Just like an earthquake, you move, cause damage (hopefully not very often!), and act according to your own will. Unlike an earthquake, however, you are composed of cells, metabolize energy-containing molecules, grow, have the potential to reproduce, respond to stimuli, and adapt to the changing environment. All in all, the characteristics that distinguish life from non-life are summed up in one idea—living things have an inner source of activity by which they act to accomplish their purpose; non-living things also exist for a purpose, but they are acted on from the outside and do not have within themselves the ability to act.

Let's take a minute to think about purpose and design in the world.

In the Physical Science text that is sister to this text, the author states that the universe comprises three basic things—matter, energy, and intelligence.[5] Matter is anything that has mass and takes up space; energy is what holds everything together and enables any process to happen; and intelligence is the wisdom of God in his creation that causes everything to work together in an orderly and beautiful way. Since God created all that exists, his wisdom is evident everywhere (Psalm 104:24),[6] including in the laws of nature that govern how everything in the universe works.

In the study of life, we distinguish between non-living and living matter. Like everything else in the universe, living things are made of matter, use energy, and obey the laws of nature. However, non-living things do not act from within according to a guiding purpose as living things do. The word purpose implies an end or goal (or, in Aristotle's language, a *telos*), such that

4 Of course, you learned in Earth Science that earthquakes are caused by the shifting of tectonic plates due to the buildup of stress. However, this cause was not apparent to my young daughter.

5 The most rigorously scientific way of describing this trio would be matter, energy, and order, where the order in nature is due to the laws of nature. The truth behind the order observed in nature is that this order is a manifestation of the intelligence of the Creator.

6 We know this is true, as Psalm 104 tells us, even though the world is presently in bondage to corruption, and groans (Romans 8:21, 22). We hold these two ideas in tension; this is a mystery.

everything a living organism does is aimed toward accomplishing one or more meaningful purposes. Non-living matter does not act from within in a purposeful fashion; it simply responds to physical processes.

Consider a common inanimate (non-living) object, such as a rock. The rock is made of atoms and molecules (matter). Its existence is the result of the God-given laws of nature (being formed by chemical reactions and weathering). Like all of creation, the rock exists for a purpose, but it does not act from within to fulfill that purpose. If the rock moves, it is because an outside force acts on it. A living thing, on the other hand, acts from within to fulfill a guiding purpose that directs all its more specific characteristics. The simplest purpose a living thing can pursue is to survive and reproduce. Beyond this, organisms display more complex purposes such as supporting other life in an ecosystem. Finally, human beings have many purposes, including the most noble "chief end" of all: "to glorify God and to enjoy Him forever."[7]

Beyond the general characteristic of acting from within to fulfill a purpose, all living things possess six specific characteristics, listed in Table 1.2. You can also think of these six characteristics as six requirements that must be met in order for a thing to be regarded as alive. Note that all six move an organism toward the fulfillment of its purpose. In the following subsections, we examine these six characteristics in more detail.

1. Living things are composed of cells and operate on many levels of organization.	Living things are made of matter, and are arranged according to highly organized, complex, purposeful designs. The most fundamental level of organization that displays all the characteristics of life is the cell.	Section 1.2.2
2. Living things metabolize.	Living things use materials from the environment and excrete waste, a process called metabolism. Waste products are broken down and used again. Energy is continually supplied from the Sun, converted, and used by organisms, which produce waste heat in the process.	Section 1.2.3
3. Living things grow, develop, and reproduce.	Organisms proceed through various life stages, typically of increasing complexity, until maturity is reached and the organism is able to reproduce.	Section 1.2.4
4. Living things use and transmit genetic information.	Living things share a common genetic code. The information in the code dictates how an organism functions and is passed on to offspring.	Section 1.2.5
5. Living things respond.	Living things have some sort of sensory system by which they respond to light, sound, motion, or other stimuli. They process the information received and respond accordingly.	Section 1.2.6
6. Living things adapt to their environments.	Populations of organisms adapt to a changing environment, as each generation favors survival of organisms with the most suitable traits.	Section 1.2.7

TABLE 1.2. *Six characteristics of life.*

7 The "chief end of man," as described by the Westminster Catechism.

To illustrate, my children were recently subjected to a dramatic experience that highlights the dividing line between life and non-life. At a school picnic, they excitedly participated in a number of games, winning three live goldfish in little plastic baggies. Not wanting to dampen their enthusiasm for their new pets, I invested in a small aquarium, colorful gravel, plastic plant-like decor, and a small pink castle, not to mention the required chemical additives to make tap water safe. We carefully transferred the fish to their new home, making sure that their baggies had time to adjust temperature so as not to shock the fish. At first, everything went along swimmingly (pun intended). Our three goldfish beautifully displayed to us that they were alive, displaying the characteristics listed in Table 1.2. Were they composed of cells? Check. Did they metabolize? Yes. They utilized matter and energy. We fed them diligently twice a day, and it was evident that they were producing excrement. They energetically swam back and forth, using the energy that the food gave them. Did they grow, develop, and reproduce? Though we didn't get to see it ourselves, as living things, these fish most certainly grew from small eggs. And my children most earnestly hoped that there might be both a male and female goldfish among them so that they might have babies. Did they use and transmit genetic information? Yes. They had genes that specified where exactly their fins should be placed, the bright orange color of their scales, and how they would breathe the oxygen dissolved in their tank water. Any of their offspring would have displayed similar traits. Did they respond? Yes, indeed! If they swam too close to a spiky plastic plant, they turned around and swam in another direction. If they swam underneath the "waterfall" produced by the filter water being returned to the tank, they swam more vigorously in order to stay on their intended path. Did they adapt to their environment? Here is where we ran into trouble.

After a few days, our goldfish friends began to behave strangely. One, whose name was Buddy, decided he preferred to hang out in one spot near the gravel. We could see his gills and mouth moving back and forth, though, so we knew he was alive. Another one, whose name was Buddy Jr., floated to and fro, but didn't exert the same energy that he once did. It was almost as though he became paralyzed, subject to the forces of the filter water alone. However, his gills and mouth continued to move, indicating that he was still alive. I looked for information as to why they might be behaving this way. Based on the best available "goldfish care theory," I came up with the hypothesis that their water had too much ammonia and needed to be partially changed. I set out to complete this change, making sure to place the correct number of drops of water conditioner, and to bring the new water to room temperature before adding it to the aquarium. Just minutes after the fish had their new water, I heard a distressed shriek coming from upstairs. One of my children saw that the goldfish were no longer breathing. Their

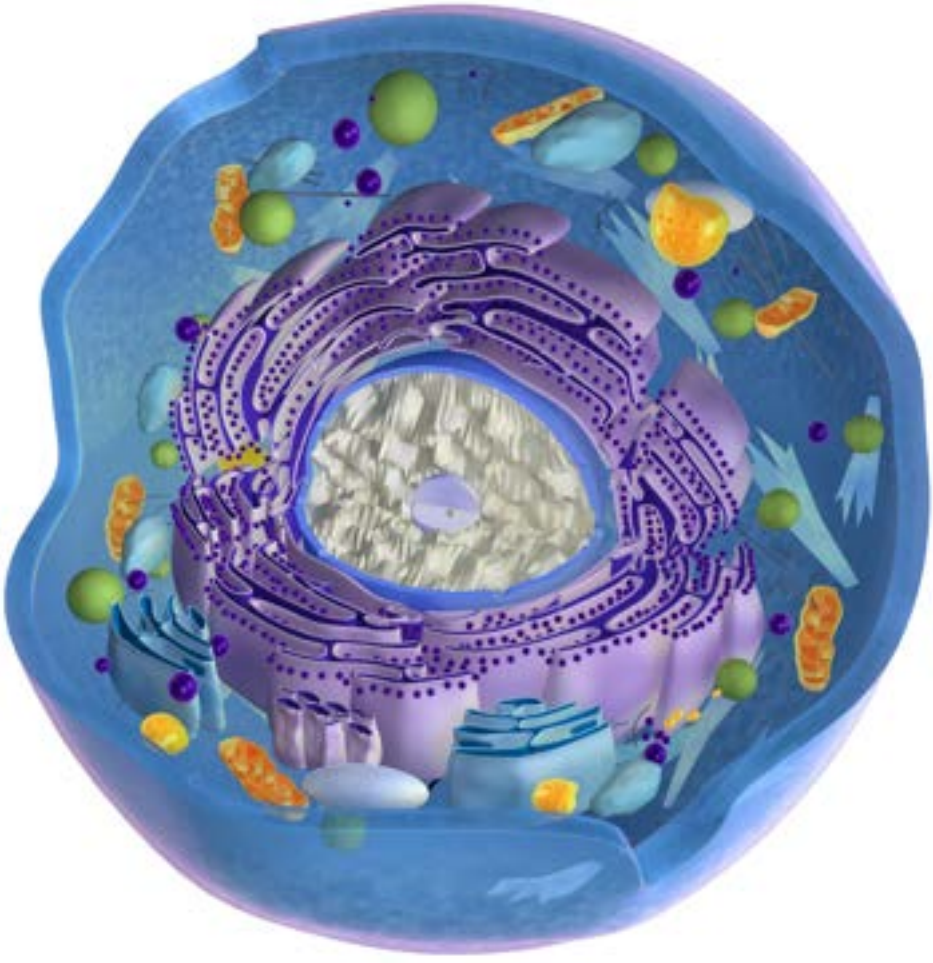


FIGURE 1.13. *A 3-D model of an animal cell. Cells range in size from 10–100 µm across, and thus are only visible with the aid of a microscope. The genetic material (DNA) is shown in grayish-white around the center.*

gills and mouths were now completely still. Our pets had ceased living.

What happened? Most likely, the stress of being a carnival prize, being driven home in a baggie, and "lovingly" handled by a six-year-old child, followed by the shock of entering a new aquarium environment was too much for these sweet goldfish to handle. They could not adapt to the stressful environment, and then, one by one, lost all the other attributes of life.[8]

The living goldfish had purpose. They lived to survive and reproduce (and to bring delight to my children). The bodies of the deceased goldfish became non-living matter. They moved because of outside forces (like the filter waterfall), not of their own accord. Now laid to rest in backyard graves, their cells are decomposing into smaller building blocks. They no longer possess any of the attributes of life.

8 If I had maintained a large population of goldfish in a much larger tank, perhaps a few fortunate ones might have survived the unfavorable water conditions. After several generations, all the resulting fish would have been much more resistant to the ammonia in the water.

Next, we take a closer look at these six characteristics of life. In later chapters, we address aspects of these characteristics in much greater detail.

1.2.2 *Cellular Structure and Levels of Organization*

So far as we know, all living things are made of cells. Just as atoms are the fundamental building blocks of matter, cells are the fundamental building blocks of life. A cell is a self-contained living factory, surrounded by a barrier called a *membrane*, containing genetic material (instructions for operation), capable of self-replication, and keeping itself alive by cycling matter and energy. Rocks, air, water, cars, and computers are not made out of cells, and thus are not alive. Living things—such as bacteria, mold, trees, badgers, and human beings—are composed of cells. Figure 1.13 shows a generalized, cut-away diagram of a cell and its parts. Chapters 3–5 of this book are devoted to delving into the fascinating details of cells and how they work. For now, just note that living beings have cells as their fundamental unit—either as a single-celled organism or as many cells working together.

The fact that living beings are composed of cells illustrates a larger principle in biology—that life operates on many levels of organization, as illustrated in the three-page Table 1.3, containing Figures 1.14a–1.14l. These levels entail differing size scales (from the size of an atom to the size of the Earth itself), and each higher level encompasses all the levels below it. At each step, higher levels of complexity and more intricate interactions are present, illustrating how the creative power of God sustains life itself. These levels of organization are foundational to the study of biology, and you must commit them to memory.

The first thing to notice about Table 1.3 is that the smallest level of organization we depict in biology is the *atom*. Figure 1.14a shows a simplified diagram of a hydrogen atom. Atoms are the fundamental unit of all matter and are much too small to be imaged except by very advanced instrumentation. Matter in general (i.e., anything made out of atoms or parts of atoms) is not necessarily alive, but it is important to know that all living things are ultimately made out of atoms.

As we discuss in Chapter 2, atoms join together (by sharing their electrons) to make molecules, as illustrated in Figure 1.14b. Simple molecules comprise compounds that you may encounter in everyday life, such as water, oxygen gas, carbon dioxide, or sodium bicarbonate (baking soda). These simple molecules are also very much active in biological processes—all of them are necessary to keep you alive. About 60% of your body's mass comes from water, you inhale oxygen gas and exhale carbon dioxide, and sodium bicarbonate regulates the acidity of your blood. However, simple molecules can also be combined to form biomolecules (also called macromolecules), the next level of organization.

Many biomolecules are formed from repeating units of simple molecules, as illustrated in Figure 1.14c. This biomolecule is a fragment of genetic material called DNA that holds the operating instructions for the cell. Other biomolecules include proteins, carbohydrates, and lipids, discussed in Chapter 2.

The next level of organization is the *organelle* (Figure 1.14d). Organelles are small parts of cells that carry out one or more specific functions. For example, one organelle, called the nucleus, is located at the center of the cell, and contains most of the cell's genetic material (DNA). I must pause here to clarify. You already know the term "nucleus" as the central part of an atom, the region holding the atom's protons and neutrons. The nucleus of a cell is a completely different thing, and in fact is many orders of magnitude larger than the nucleus of an atom. Their only common feature is being the centrally located portion of a fundamental building block—of either atoms or cells.

Now, organelles are amazing because they are composed of different parts that work together. Going back to the example of the nucleus of the cell, several parts of the nucleus collaborate to control many other cellular activities, making the nucleus the command center of the cell. (The nucleus itself is regulated by signals it receives from outside itself via hormones.) Several types of biomolecules of different shapes and sizes are found in the nucleus. These parts intricately work together like a finely tuned factory, sending out messages that control what many other organelles in the cell do. Every other organelle in the cell has specific functions as well, and we elaborate more on these roles in later chapters.

Although each organelle has different parts that work together to do a job, a lone organelle cannot be considered a living organism. Consider why this is the case. Let's return again to our example organelle, the cellular nucleus. By itself, does a cellular nucleus possess all six of the characteristics of life?

1. Is it made of cells? (no)
2. Does it cycle matter and energy? (partly)
3. Can it reproduce itself without the help of other organelles? (no)
4. Does it use and transmit genetic information? (yes)
5. Does it respond? (yes, molecular signals control how many messages are sent out based on environmental conditions)
6. Does it adapt? (no, adaptation occurs in populations, not individuals)

Based on the first characteristic alone—that living things are made of cells—an organelle does not qualify as a living thing. But additionally, the nucleus of a cell doesn't completely cycle matter and energy (other organelles called the mitochondria are necessary to produce the chemical energy the organelle uses) and it cannot reproduce itself. An organelle by itself does

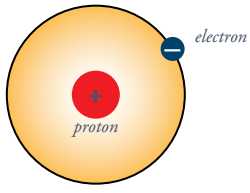
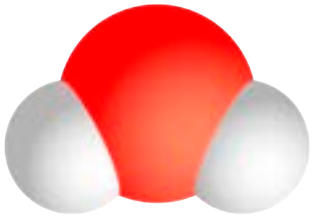
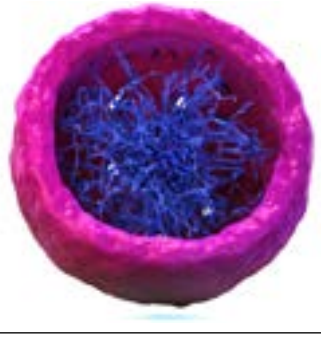
Level	Size scale (length or diameter in meters)	Illustration	Caption
*Atom	10^{-11}–10^{-10}		Figure 1.14a. An atom is the fundamental unit of matter, made of protons, neutrons, and electrons. Only 118 types of atoms have been found in creation (or synthesized), and are cataloged in the periodic table. This diagram depicts a hydrogen atom, which does not have any neutrons.
*Simple molecule	10^{-10}–10^{-9}		Figure 1.14b. Molecules are made by joining atoms together through the sharing of electrons. This illustration depicts a water molecule, which joins two hydrogen atoms (white) to one oxygen atom (red).
*Biomolecule	10^{-9}–10^{-7}		Figure 1.14c. A biomolecule is a large molecule that often consists of many repeating small molecules attached together. DNA, RNA, proteins, lipids, and carbohydrates are types of biomolecules. This image shows a model of a small (13 base pair) segment of DNA.
*Organelle	10^{-8}–10^{-6}		Figure 1.14d. Organelles are groups of different biomolecules that work together to perform one or more functions within a cell. This image depicts the nucleus of a cell. Present are the nuclear membrane (pink) and DNA (blue).

TABLE 1.3. *Levels of organization in biology. For items (f)–(k), fish are used as examples.*

*Note that by themselves, entities smaller than a cell are not alive. However, when put together in the organized, purposeful array of a cell, they comprise life.

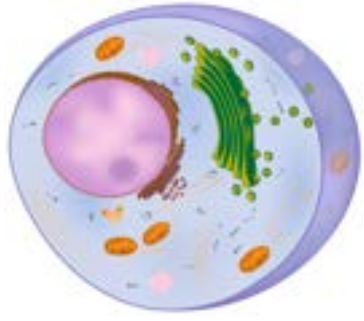
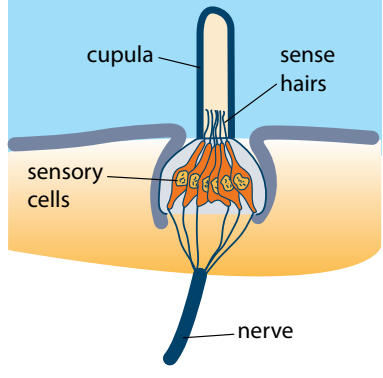
Level	Size scale (length or diameter in meters)	Illustration	Caption
Cell	10^{-7}–10^{-4}		Figure 1.14e. Cells are the fundamental unit of life. Cells contain multiple organelles as well as DNA, and are enclosed by a cell membrane. These parts work together to perform all the functions of life (metabolism, reproduction, growth, response, and adaptation). The nucleus of this cell is shown in light pink.
Tissue	(varies)		Figure 1.14f. A tissue consists of many cells of the same type that work together. In the center of this figure are several sensory cells that work together to sense pressure changes underwater in the lateral line of a fish. (The entire structure shown is called a neuromast.)
Organ	(varies)		Figure 1.14g. In an organ, tissues work together to perform particular tasks that benefit the whole body. This picture shows the lateral line of a fish, where each of the dark spots lined up horizontally is one neuromast (illustrated in Fig 1.14f). The lateral line can sense pressure changes in the water.
Organism	10^{-7}–30		Figure 1.14h. An organism is an individual living being, composed of organs that work together to perform the functions of life. (Some organisms consist of a single cell.) This photo shows a parrotfish.

TABLE 1.3 *(continued). Levels of organization in biology. For items (f)–(k), fish are used as examples.*

Level	Size scale (length or diameter in meters)	Illustration	Caption
Population	(varies)		Figure 1.14i. A population is a group of organisms of the same species living in a particular region. This photo shows a group of parrotfish.
Community	up to 10^6		Figure 1.14j. A community consists of multiple, interacting populations in a particular region. This photo shows a number of fish species swimming in close proximity.
Ecosystem	up to 10^6		Figure 1.14k. An ecosystem is a community of living things that interact with nonliving components of their environment. Here are a number of fish and coral polyps that interact with water, sunlight, and calcium carbonate to form a coral reef ecosystem.
Biosphere	10^7		Figure 1.14l. The biosphere consists of all ecosystems on earth.

TABLE 1.3 *(continued). Levels of organization in biology. For items (f)–(k), fish are used as examples.*

not include all the characteristics of life, but organelles have specialized roles so that an overall cell does include all the characteristics of life. It is for this reason that the cell is the lowest level of organization considered in and of itself to be alive. Another diagram of a cell is shown in Figure 1.14e.

Just as organelles have specialized roles in order to serve all the necessary functions of life for the cell, cells actually specialize their functions as well, to serve the needs of the entire organism—your body, for example. A group of cells of the same type that work together to perform a function is called a *tissue*.

In Figure 1.14f, we illustrate tissue with an example, found in most fish, called the *lateral line system*. As underwater creatures, most fish have a sort of "sixth sense" in that they are able to sense pressure and movement in the water. For example, if a crustacean swims nearby, a fish senses the resulting waves, and knows where to swim to find its next meal. In the center of the structure shown in 1.14f is a tissue composed of the hair cells that sense these very subtle changes in the water. This group of identical cells is part of a larger organ-like structure called a *neuromast*, which contains other cell types in addition to the hair cells.

Collectively, the tiny neuromasts form the lateral line, which is visible on the body of a fish and shown in Figure 1.14g. This lateral line is the organ responsible for the ability of a fish to sense water movement. This amazing ability helps fish to find prey, avoid predators, and stick together in groups.

An *organism*, or individual living being, typically comprises many coordinated organ systems. Figure 1.14h shows an individual parrotfish. Generally, an organism may consist of one or many cells. In the case of a many-celled organism, the cells stick together, forming specialized tissues and organs that coordinate their efforts so that the entire organism may survive and reproduce.

A *population*, or group of organisms of the same species living and interbreeding in the same area, is shown in Figure 1.14i. Populations do not exist alone, however. They are typically part of *communities*—groups of populations living and interacting in the same area. These interactions could include both cooperation and competition, or even species eating (preying upon) others. Even if one species does not prey upon others, it can still compete with others for food resources or living space. Figure 1.14j shows a community of various fish species living in close proximity.

Moving on, living things interact with the non–living components of their environment. The colorful tropical fish in Figure 1.14k are part of a coral reef ecosystem. An *ecosystem* is a system of living communities interacting with non-living parts of their environment. In order to survive, this ecosystem must remain within a very narrow temperature range (just around 80°F) and must be located at just the right depth so that it is

always covered with water, yet close enough to the surface to receive sufficient sunlight. Water level and temperature are just two non-living factors that play roles in the survival of the species of this ecosystem. The fish pictured in Figure 1.14k, small organisms called *phytoplankton* (not visible to the naked eye and not evident in the picture), and *coral polyps* that secrete calcium carbonate are just a few of the numerous amazing creatures found in coral reefs. We are naturally awed and inspired by the beauty of ecosystems such as these, and we should embrace our human vocation of caring for the earth.

Finally, the *biosphere*, shown in Figure 1.14l, encompasses all living things on earth and is the largest level of biological organization we know of.

1.2.3 *Living Things Metabolize*

Living things carry out a group of chemical reactions collectively known as *metabolism*, by which they use matter and energy to power all their processes. In order to remain alive, an organism must repeatedly take in matter from the environment. These molecules are then transformed into different molecules that the organism uses, harnessing energy in the process. Unusable matter is returned to the environment in the form of waste. Other organisms in the environment then transform the waste into usable building blocks, and the matter-recycling process continues.

Some types of organisms, such as plants, also have the amazing ability to make direct use of energy in the form of sunlight, storing that energy in the bonds of molecules. These molecules are then used by other organisms or broken down to retrieve the energy. All organisms need energy in some form to power all their processes. Without usable energy, they would cease living.

FIGURE 1.15. *An example of a producer (moss), consumer (hedgehog), and decomposer (mushroom) in the same ecosystem.*

In essence, there are three major categories of organisms that participate in these metabolic processes, illustrated in Figure 1.15. *Producers*, such as moss, harness the Sun's energy, storing it in molecules that can be eaten and used by other organisms. *Consumers*, such as the hedgehog, eat plants and other animals, reusing the matter and stored energy for their own purposes. Finally, *decomposers*, such as mushrooms, secrete special biomolecules that break down excreted waste products or the remains of dead organisms, allowing those molecules to be used again. In short, energy flows from the Sun to producers, then to consumers, then to decomposers, and ultimately is dissipated as heat. The matter continues to be broken down and built back up again, recycled over and over. The cycling of matter and energy takes place in thousands upon thousands of metabolic reactions, which we discuss in much more detail in Chapter 4.

Reflect for a moment on the exquisite design of this system. The interplay of producers, consumers, and decomposers shows the wisdom of our great God. Consider how God

provides everything we need for life, and how wonderful it is that all God's creatures support one another in their needs for matter and energy. Not only that, but God designed these processes to be sustainable, such that the cycle continues on and on without ever being exhausted (at least so long as the Sun continues to shine!). Let us thank God for His goodness to us and resolve to care for the Earth He designed for us so that we don't compromise this exquisitely designed environment through carelessness, waste, or pollution.

1.2.4 *Living Things Grow, Develop, and Reproduce*

Living things exhibit a pattern of growth over the course of their lifespan that leads toward reproduction. This does not simply mean that organisms get bigger—although attaining larger size is part of the process. Living things also develop into organisms of increasing complexity as time progresses. The purpose of this process is to reach sexual maturity and reproduce so that life may continue.

Let's look at the stages of human life as an example. The stages of fetal development are illustrated in Figure 1.16. A person begins as a single cell—the joining of a sperm and an egg, with a unique genetic makeup. At that point, the cell begins to divide (a process called mitosis), becoming two cells, then four, then eight. As the process of cell division continues, certain cells begin to specialize their roles. Though each cell has a complete copy of the person's DNA, specific cells begin using only certain parts of the DNA. As a result, some cells are set on a path to become blood cells, others are destined to become brain cells, and so on. As time progresses, the expressed genes in each cell begin to dictate how cells are arranged within the body. The growing baby begins to look more and more human-like, with a head, a

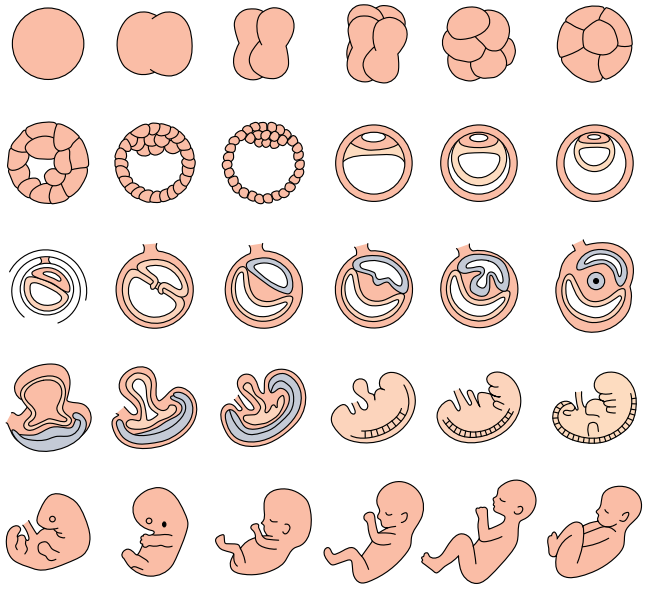


FIGURE 1.16. *The stages of human fetal development.*

face, a torso, two arms, and two legs. The heart begins to beat just a few weeks after fertilization of the original egg, before the mother even knows she is pregnant!

Dramatic growth and differentiation of cells continues for about nine months, until the baby's lungs are ready to breathe oxygen, and then the baby is born. Yet even after birth, the process of growth and development continues.

The baby gradually gains the ability to roll over, crawl, walk, and talk. As the child reaches school age, she learns to read and write. She reaches physical milestones, such as losing baby teeth and gaining permanent ones. Eventually, the child reaches puberty, initiating a series of changes by which she becomes physically capable of sexual reproduction.

All the while, the person gains increasingly complex reasoning abilities. She can now visualize abstract concepts as she studies subjects in increasing depth throughout her high school career and beyond.

Development continues throughout adulthood as well. Around middle age, signs of aging appear—such as graying hair, diminishing eyesight, and menopause in women.

As you can see, human beings do not remain stagnant, but follow a predictable pattern of growth and development throughout life—from conception through death. This process includes becoming sexually mature so that one may reproduce, creating more individuals and continuing the human race.

All organisms go through some developmental process. Butterflies progress from caterpillars to adult butterflies; frogs change from eggs to tadpoles to adult frogs. Even bacteria pass through phases as they repeatedly go through the process of cell division. In summary, growth, development, and reproduction are processes pertaining to all life.

1.2.5 *Living Things Use and Transmit Genetic Information*

Before a building is built, an architect draws up the plans for the workers to follow. Before a play is performed, the actors require a script. Computers require software in order to perform any task.

In a similar way, an organism requires a set of instructions to control its functions. However, rather than being drawn on a sheet of drafting paper or typed in word processing software, these instructions are encoded in a special language stored in the structure of a biomolecule called *deoxyribonucleic acid*, or *DNA*. Amazingly, all organisms use DNA as their genetic material—from the simplest bacteria to mammals and even to human beings. We delve into the fascinating details about DNA in Chapters 5 and 7. For now, we simply note the following four highly significant properties of DNA:

1. DNA can be copied in its entirety, so that the copies may be transmitted to offspring.
2. Small portions of DNA can be copied.
3. The information in DNA is stored in chemical "language" composed of an alphabet of four compounds. We represent these compounds by four letters of the English alphabet. The chemical language is read and understood by other parts of the cell.
4. The language of DNA is the same for all living species on Earth.

Each of the trillions of cells in your body contains instructions that are billions of DNA "letters" long. That's the amount of DNA required to encode for you! Though all living things share the same alphabet and the same language, no two species, nor even two individual humans, share the exact same genetic makeup. As a rough analogy, there are many novels written in English, but the sequence of letters in each novel is vastly different from the next. There are genres of similar novels, such as mystery, romance, historical fiction, and so on. These genres are analogous to the similar genetic codes shared by members of the same species. But because of the versatility inherent in a written language, no novel is identical to another. Consider the great care God has for us that He specifically designs each person with a unique genetic makeup. As Psalm 139:13–14 says, "For you formed my inward parts; you knitted me together in my mother's womb. I praise you, for I am fearfully and wonderfully made. Wonderful are your works; my soul knows it very well."

When living things reproduce, they pass their genetic material on to their offspring by asexual or sexual reproduction. In asexual reproduction, a single-celled organism makes a copy of its DNA and then divides into two identical daughter cells. Thus, asexual reproduction results in offspring that are, in most cases, genetically identical. In sexual reproduction, two parents each contribute half of the DNA in the resulting offspring. The result is a genetically unique individual.

To summarize, the DNA in all living things provides the foundational information used to determine how each life function is carried out. The DNA in every cell contains the instructions governing how the cell can behave. Living things transmit their DNA (in part or in whole) to the next generation, through either sexual or asexual reproduction.

1.2.6 *Living Things Respond*

As illustrated in the sad tale of the pet goldfish, living things respond. We use the word *stimulus* to indicate any event that causes an organism to react. The stimulus causes a signal to be transmitted in the form of light, sound, pressure, motion, temperature, or *chemical gradient* (a region of varying concentration in solution). An organism must have a sensory system in place in order to detect the stimulus, process the information,

and then respond accordingly. The goldfish could sense the presence of the plastic plants in the aquarium and turn around and swim in the other direction.

To cite another example, say you are outside playing volleyball with your friends. Suddenly, the ball is rushing directly towards your head. You detect this stimulus by seeing the shadow of the ball passing in front of you, and you hear your friends yelling "heads up!" Your brain detects the information transmitted from your eyes and ears, interprets that information as an imminent threat, and immediately directs your muscles to respond. If you are like some of us, you might duck, cover your head, and move out of the way. Those who are more athletically inclined might instead jump towards the ball and spike it over the net. Either way, you sense a signal, process it, and respond.

You might think that this property is only present in higher (more intelligent) organisms, but even simple bacteria respond to stimuli. Many bacteria exhibit *chemotaxis*, the ability to sense concentrations of molecules and swim towards them (or away from them, if they are poisonous). This ability helps the bacteria both to find food and to avoid threats.

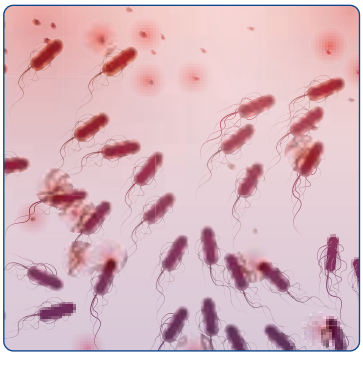


FIGURE 1.17. *An artist's concept of bacterial chemotaxis in* E. coli.

Figure 1.17 shows an artist's rendition of bacterial chemotaxis. The rod-shaped *E. coli* cells each have multiple *flagella* (singular: *flagellum*), string-like projections extending from their cell bodies. These flagella spin like propellers, enabling the bacteria to move. When no chemical attractants are present, the flagella all spin facing outward from the bacterium, and the bacterial cell tumbles in place. However, when an increasing concentration of a nutrient molecule is present (such as sugar), the bacterium aligns the flagella in the same direction. This alignment causes the bacterium to propel forward toward the sugar. Thus, bacteria respond to the stimulus of the chemical gradient caused by the sugar's presence and are rewarded with a tasty treat.

Though bacteria don't have a nervous system as you and I do, they can sense the presence of these molecules in the water and respond accordingly. Perhaps, then, it isn't really fair to consider *E. coli* a "simple" organism (though it is tiny and single-celled). The flagellum itself is a rather complex marvel of mechanical engineering. (I'll leave that one for you to look up yourself.) Life and its processes are truly amazing, no matter how large or small the organism.

Now, let's pause briefly to consider how stimulus and response relate to the notion of *homeostasis*. While organisms respond to stimuli in isolation, they also maintain various continuous stimulus/response processes so that the organism remains within life-sustaining limits.

To understand homeostasis, let's consider the thermostat on the heating/cooling unit of a house. The thermostat constantly monitors the temperature of the surrounding air, keeping it at a specified temperature. When I lived in the snowy

state of Minnesota, I kept my thermostat set at 68 degrees Fahrenheit all winter long. If the temperature dropped below 68°F, the thermostat signaled the furnace to blast warm air until the temperature reading returned to the desired temperature. Now living in sunny California, I set my thermostat to 78°F during the summer. If the temperature should rise above this value, the thermostat signals the air conditioner to infuse cold air until the reading returns to 78°F. Rather than a one-time stimulus/response event, this is a continuously occurring stimulus/response feedback cycle.

In the same way, to maintain homeostasis organisms monitor pH, salt concentration, temperature, nutrient levels, and a whole host of other parameters—adjusting their responses to these conditions so that the body remains in an acceptable, life-sustaining range. Maintaining homeostasis is a particularly complex manifestation of the stimulus/response characteristic of life.

1.2.7 *Living Things Adapt to the Environment*

In a constantly changing environment, organisms must have strategies in place so they can survive, thus fulfilling their common purpose. One strategy is *adaptation*. In the present context, adaptation is not exhibited by an individual organism, but by a population of organisms over several generations. This process of adaptation depends upon genetic variability among the members of the population.

As an example, consider the finches that Darwin observed during his voyage to the Galapagos Islands in 1845, illustrated in Figure 1.18. Since the Galapagos Archipelago originated from volcanic activity, its species had to migrate there from other locations. These finches probably came from a population of birds on the mainland. As the finches settled on the different islands of the archipelago, differing food sources led to higher reproductive success for birds with certain traits. For example, in Figure 1.18, the large ground finch labeled #1 (*Geospiza magnirostris*) has a large, short beak that is adapted to cracking nuts. Isolated on an island with nuts as the major food source, birds in the original population that had this trait already (by the nature of genetic variability) were more successful in obtaining food. Better fed, birds with the large, short beak were also more successful at reproduction. After several generations, this trait

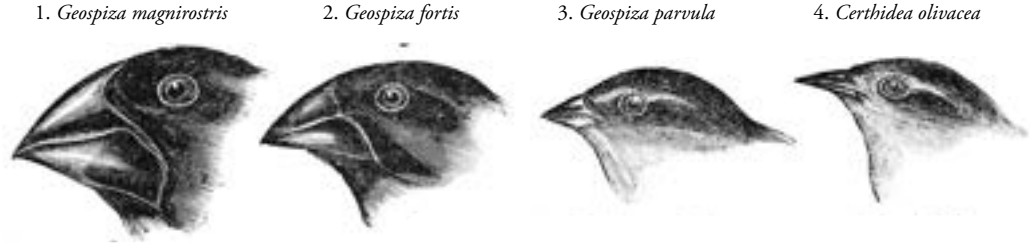


FIGURE 1.18. *Finches of the Galapagos Islands, with beaks adapted to varying food sources.*

became predominant in the population, due to the survival advantage it conferred.

On the other hand, the medium ground finch #2 (*Geospiza fortis*) lived on an island with small soft seeds as its major food source. As a result, finches with the beak most adept at eating these seeds had a reproductive advantage. After several generations, most of the medium ground finches in the population possessed a beak like the one pictured here (drawn in 1845). Interestingly, since that time, scientists observed that a drought in the 1970s caused further adaptation in this population. The drought shifted the major food source from small soft seeds to much harder seeds, and as a result, scientists observed a 10% change in the sizes of the beaks of the medium ground finch. This adaptation to changes in climate enabled the birds to utilize the changing food source more effectively.

Adaptation can also be readily observed in bacterial populations. In 1943, penicillin was introduced as an effective antibiotic against a disease-causing bacterial species, *Staphylococcus aureus* (*S. aureus*). After just two years, up to 20% of *S. aureus* infections became resistant to penicillin treatment. These resistant strains of *S. aureus* had adapted to the threat (by producing more of an enzyme called *penicillinase*, which breaks down the deadly antibiotic). Later, another antibiotic called methicillin was introduced as an effective treatment. Yet today, MRSA (methicillin-resistant *Staphylococcus aureus*) infections are a formidable problem in medicine. As new antibiotic treatments are introduced, bacterial populations develop resistance to antibiotics and are not so easily killed off. This is why it is important to use antibiotics cautiously. We want to avoid generating antibiotic-resistant strains we cannot treat.

Adaptation is an important feature of life that enables populations to survive amid changing conditions. We explore the process of adaptation (also called microevolution) further in Chapter 9.

1.3 The History of Biogenic Theory (Can life emerge from non-life?)

So far, we have explored the answers to two questions: 1) By what methods do we make new discoveries in the field of biology? 2) By what criteria do we judge something to be alive? We now use this information to explore how ideas about biology have changed over time.

1.3.1 *Aristotle and Redi*

The ancient Greek philosopher Aristotle (Figure 1.19) wrote extensively about nature (natural philosophy). Aristotle was the first philosopher to propose the theory of *spontaneous generation*. Spontaneous generation is the idea that living things can arise randomly out of non-living matter. To Aristotle, spontaneous generation made sense according to everyday observa-

tion. Frogs were seen to emerge from the mud alongside riverbeds—therefore, frogs must be made out of the mud. Worms are seen on rotting meat—therefore, worms must come directly from that meat.

Spontaneous generation was not seriously questioned for about 2,000 years. Then, in 1668, Tuscan naturalist Francesco Redi (Figure 1.20) performed an experiment showing that maggots found on rotting meat did not actually originate from the decaying flesh. Redi assembled a large array of different types of meat, leaving some samples to rot in uncovered jars, and others in jars covered with cheesecloth. The uncovered samples became infested with maggots after a few days. However, the covered samples had no maggots directly on the meat. Instead, adult flies were seen hovering over the cheesecloth, where they laid their eggs. Redi concluded that maggots are simply part of the life cycle of flies, and that an adult fly must lay its eggs on the rotting meat in order for the maggots to develop there. Redi thus showed that life does not proceed directly from decaying matter, as the proponents of the theory of spontaneous generation argued. Rather, life proceeds from parent to offspring. Redi's famous conclusion in Latin was *omne vivum ex vivo*, or *every living thing from a living thing*.[9]

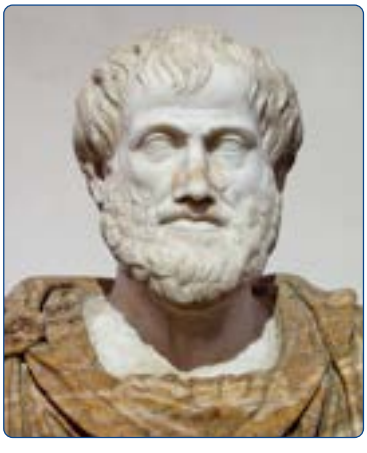


FIGURE 1.19. *A statue of ancient Greek philosopher Aristotle (384–322 BC).*

FIGURE 1.20. *Tuscan naturalist Francesco Redi (1626–1697).*

1.3.2 *Needham and Spallanzani*

Eighty years passed before another test of the theory of spontaneous generation occurred. While Aristotle and Redi could only observe the generation of macroscopic animals, by the 18th century the invention of the microscope allowed investigation of microorganisms as well. In 1748, Irish priest John Turberville Needham (Figure 1.21), in collaboration with French aristocrat Comte de Buffon, performed an experiment that supported the theory of spontaneous generation. Needham heated mutton gravy in stoppered glass tubes until boiling,

9 In most texts, the Latin phrase is translated less accurately as "all life comes from life." However, *omne* means every, and *vivum* and *vivo* are both forms of the Latin word for "living thing."

FIGURE 1.21. *Irish priest John Turberville Needham (1713–1781).*

FIGURE 1.22. *Italian scientist Lazzaro Spallanzani (1729–1799).*

under the assumption that boiling killed all life within. Afterwards, Needham was able to observe many quickly-moving microorganisms and smaller so-called "organic molecules" in the fluid. According to Needham's theory supporting spontaneous generation, these "organic molecules" had a special life-giving force and clumped together to form larger microorganisms called "animalcules." Thus, he concluded that the microorganisms arose spontaneously from previously non-living matter.

In 1765, Italian priest Lazzaro Spallanzani (Figure 1.22) questioned the results of Needham and Buffon, repeating their experiment with some modifications. First, Spallanzani hermetically sealed his tubes, meaning that he melted the glass to make an airtight seal. Second, he boiled the gravy for at least an hour. As a result, Spallanzani did not observe any microorganisms forming after treatment. Since they were contemporaries, Spallanzani's results stirred up a debate between Needham and himself. Spallanzani argued that Needham did not boil his gravy long enough to kill all the organisms, and that by not tightly sealing his vessels airborne microorganisms could cause contamination. Needham responded that Spallanzani's tight seal and excessive boiling destroyed the "life force" in the air that is necessary to generate new life.

Applying what we know about the Cycle of Scientific Enterprise, we see how both Needham and Spallanzani were attempting to use competing theories to account for the same body of scientific facts. According to the theory of spontaneous generation, Needham could predict (make a hypothesis) that new life would emerge after all previous life had been killed. He performed an experiment that seemed to support this hypothesis, and so evidence for spontaneous generation was strengthened. Spallanzani, suspecting that all scientific facts did not, in reality, support the theory of spontaneous generation, proposed instead that Needham's results were the result of experimental flaws. Spallanzani repeated Needham's experiment with improved design, achieving the opposite result—no microbial growth. Throughout the history of science, conflicting results and even fierce debates have been common. These debates cause the Cycle of Scientific Enterprise to move forward toward ever stronger theories, as experimental flaws and other hidden mysteries are revealed by further testing. Scientists might not be as motivated toward repeated testing and improved experimental design if everyone agreed all the time.

1.3.3 *Pasteur and the Victorian Debates*

A century passed, and the question of spontaneous generation was still far from settled. In the 1860s, French scientist Louis Pasteur (Figure 1.23) designed an experiment to settle this question as part of a contest hosted by the French Academy of Sciences. Needham's criticism of Spallanzani's experiment was that it excluded outside air, thus impairing some "vital force" from forming new life. As a result, Pasteur designed a

new kind of flask that would allow air inside, but not heavy dust particles. This apparatus, known as the "swan-necked flask," is shown in Figure 1.24. Pasteur predicted that contamination from the outside air was the cause of bacterial growth after boiling a broth or gravy, and his swan-necked flask was designed to prevent this contamination.

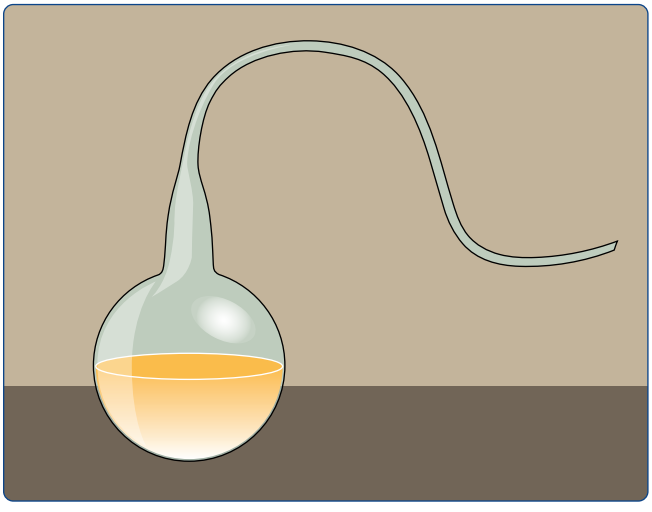


FIGURE 1.24. *A sketch of Pasteur's swan-necked flask.*

First, Pasteur boiled broth in several of his special flasks. In the first sample, he broke off the neck of the flask, exposing the broth to the air above. This sample grew microorganisms after a few days. In the next sample, he simply boiled the broth, and allowed it to sit for months. Even though it was exposed to the air through the open swan-necked flask, no bacterial growth occurred. After many months, Pasteur finally tipped the flask over, exposing the broth to the dust that had settled at the edge of the opening, and then setting it upright again. After a few days, bacterial growth occurred. Pasteur's experiment gave strong support to *biogenic theory* (*biogenesis*)—all life comes from previously existing life.

Though many synopses on the history of spontaneous generation end with Pasteur, it should be noted that the debate raged on for at least another decade. English scientist Thomas Henry Huxley (Figure 1.25) popularized Pasteur's results, while another English scientist named Henry Charlton Bastian (Figure 1.26) fervently debated Huxley in favor of spontaneous generation. Bastian, a supporter of Darwin's newly published theory of evolution, theorized that the first life must have emerged from non-life, and that microorganisms continue to do so. Rather than use the term spontaneous generation, Bastian instead coined the term *archebiosis*. This word implies that life, while emerging from nonliving matter, does so according to natural law. Previous supporters of spontaneous generation

FIGURE 1.23. *French scientist Louis Pasteur (1822–1895).*

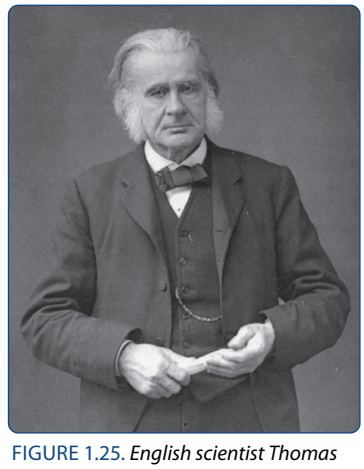


FIGURE 1.25. *English scientist Thomas Henry Huxley (1825–1895).*

FIGURE 1.26. *English physiologist Henry Charlton Bastian (1837–1915).*

FIGURE 1.27. *Irish physicist John Tyndall (1820–1893).*

viewed the process as completely random and coincidental, not necessarily following any natural laws.

In the midst of these debates between Huxley and Bastian, Irish physicist John Tyndall (Figure 1.27) entered the fray. Tyndall's early experiments heating "infusions" in an enclosed dust-free environment showed no microbial growth. However, after some time repeating these experiments, Tyndall did begin to notice bacteria forming after heating in the dust-free air. He eventually traced these unexpected results to a bale of hay that he recently had moved into the room. Repeating his experiments in a different building yielded the original results—no bacterial growth. A subsequent discovery by another scientist, Ferdinand Cohn, showed that the presence of hay can lead to an interesting phenomenon. Bacteria that live in hay are notorious for forming heat-resistant spores that can survive excessive boiling. These spores from the hay contaminated Tyndall's experiments that showed bacterial growth, which explains why moving his experiment to another building solved the problem. Tyndall then invented a procedure for killing the spores—by intermittent and repeated boiling and cooling. It seemed that the debates over spontaneous generation were definitively put to rest, as nearly everyone then accepted that life arises only from previously existing life.

1.3.4 *Modern Vocabulary*

To summarize this section, throughout history scientists (and the public at large) have taken different stances on the question of spontaneous generation. Aristotle and Redi focused on the generation of macroscopic life, arguing for and against its emergence from non-living matter. Needham and Spallanzani focused on the question of microorganisms, and whether they can arise from a broth that has been heat-sterilized. Pasteur and his contemporaries in the 1860s focused on the experimental complications of previous work, correcting for contamination by airborne microorganisms and heat-resistant bacterial spores. Additionally, they engaged in a fierce public debate over the theory of biogenesis. By the end of this era, the theory that life must proceed from life (biogenesis) became nearly universally accepted.

Today, scientists sometimes use the term *abiogenesis* rather than spontaneous generation or archebiosis. Huxley coined this term, where *a–* is a prefix meaning "not," *bio–* means "life," and *–genesis* means "beginning." The term abiogenesis implies an event that may have occurred just once in the past, resulting in the first cellular life. This idea is controversial; however, it is important that you understand the distinctions between the three terms: spontaneous generation (life emerges randomly from non-living matter on a regular basis); archebiosis (life emerges from non-living matter on a regular basis according to natural law); and abiogenesis (life may have emerged from non-

In the Wonderworld of Biology

Comparing Orders of Magnitude

The study of biology can be an overwhelming prospect because there are so many facts to learn. To illustrate, let's look at some mathematical comparisons relevant to biology.

Say you live to be 100 years old. Doing some simple math, the span of your life would be:

$$100\ \text{yr} \cdot \frac{365\ \text{dy}}{\text{yr}} = 36{,}500\ \text{dy}\ (\simeq 10^4\ \text{dy})$$

$$36{,}500\ \text{dy} \cdot \frac{24\ \text{hr}}{\text{dy}} = 876{,}000\ \text{hr}\ (\simeq 10^6\ \text{hr})$$

$$876{,}000\ \text{hr} \cdot \frac{60\ \text{min}}{\text{hr}} = 52{,}560{,}000\ \text{min}\ (\simeq 10^8\ \text{min})$$

$$52{,}560{,}000\ \text{min} \cdot \frac{60\ \text{s}}{\text{min}} = 3{,}153{,}600{,}000\ \text{s}\ (\simeq 10^9\ \text{s})$$

So, if you live to be 100, you will have lived about three billion seconds. By comparison, your body has about 3.7×10^{13} cells and a similar number of bacterial cells that live inside your gut. Of your cells, your brain contains about 100 billion (10^{11}) neurons, each of which can make 1,000 connections for a capacity of 10^{14} overall connections.

The number of cells in your body far exceeds the number of seconds in your lifespan, so there is no way you could ever catalog them all. Yet, our heavenly Father, who created all life on earth, knows the number of hairs on your head (Matthew10:30) and the number of cells in your body. God also intimately knows every base pair of the DNA (numbering about 3.2 billion) in each of your cells and how that information makes you uniquely you.

Taking our investigation a step further, it has been estimated that in all human history about 175 million books have been written. With only about 53 million minutes in your lifespan, you have no hope of reading them all (as if you wanted to). Some say that all these books could be preserved digitally in about 175 terabytes of data. (Tera is the SI prefix that represents 10^{12}, so 175 terabytes is about 10^{14} bytes). Furthermore, current estimates of the amount of information contained on the Internet fall in the zettabyte range (or 10^{21} pieces of data). These numbers regarding books written and information on the Internet represent a significant fraction of the sum of information that humanity has collectively produced over time.

By contrast, it has been estimated that the total of all DNA base pairs contained in the biosphere, that is, in every living thing on earth, numbers around 5×10^{37} base pairs. Thus, the number of base pairs contained in all life is about 10^{16} (10 quadrillion) times larger than the total number of bytes of data necessary to preserve all the information contained in books and on the Internet.

Let that sink in for a minute. How much greater is God's knowledge and power than our own—that the blueprint for life exceeds the sum total of human accomplishment by 16 orders of magnitude! No wonder the study of biology can be overwhelming. There is no way we can understand the intricacies of life the way God does.

On the other hand, God didn't design our world in a random or chaotic way, but infused the created world with order and beauty. Not only that, but God endowed humanity with the ability to understand, question, and explore the world. Biology is unified by general logical principles that we can begin to understand. And as we do, we can experience ever greater awe toward the One who designed it all.

life just once a long time ago; since that time, life has proceeded from other life).

The origin of life is a subject that is still actively being researched today. We now know just how complex a single cell is, and it is not easy to discern exactly how its individual components could have come together in just the right way to form a living—and self-replicating—entity from a non-living one. However, just as everything in nature follows orderly, beautiful, mathematical principles designed by the Creator Himself, we can trust that God's creative work is ultimately responsible for all living things, past, present, and future. As the apostle Paul writes in Colossians 1:17, "He is before all things, and in Him all things hold together." Maybe you will one day participate in the Cycle of Scientific Enterprise yourself, perhaps even making your own contribution to the theory of biogenesis.

Chapter 1 Exercises

SECTION 1.1

1. Distinguish between truth and scientific facts.
2. Write three true statements and three factual statements (that are correct so far as we know).
3. Distinguish between theories, hypotheses, and experiments.
4. Explain how the Cycle of Scientific Enterprise works.
5. Make a table listing the three types of microscopes discussed in this chapter. For each, include columns with the following information: 1) how the image is obtained, 2) range of magnification, 3) degree of resolution, 4) types of structures that can be observed, and 5) description of the resulting image.
6. For each of the seven common measurements/metric prefixes listed in Table 1.1, identify two additional applications to actual objects or phenomena in nature.
7. Compare the micron and the angstrom (i.e., calculate their ratio).

SECTION 1.2

8. List and briefly describe the six characteristics of life.
9. Make a table listing the levels of biological organization. For each level, list a defining characteristic.
10. Explain why a cell is the simplest level of organization considered to be alive.
11. Other than those discussed in the book, give an example of a cell type, tissue, organ, and organism. Make sure that each example is a component of the level of organization above it.
12. Write a paragraph distinguishing between producers, consumers, and decomposers.
13. Compare and contrast sexual and asexual reproduction.
14. Briefly trace the developmental process of a human being, from conception through death.
15. Besides those mentioned in the book, think of three examples of an organism responding to a stimulus. For each example, explain what the stimulus is, how the organism senses it, and how the organism appropriately responds.
16. Define homeostasis, giving an example of an organism exhibiting homeostasis.
17. Define adaptation, giving an example of a population adapting to changing environmental conditions.

SECTION 1.3

18. Compare and contrast the theories of Aristotle and Redi.
19. Compare and contrast the experiments of Needham and Spallanzani. How were their theories different from those of Aristotle and Redi?
20. Describe the experiments and conclusions of Louis Pasteur.
21. What experimental modifications did John Tyndall make, and how did his results strengthen biogenic theory?
22. Distinguish between Bastian's term *archebiosis* and Huxley's term *abiogenesis.* How are these two terms different from the term *spontaneous generation*?

CHAPTER 2

The Chemistry of Life

The physical and chemical properties of substances underlie all of biology, determining how and why life's processes work the way they do. Here, you can see how water behaves when in contact with the waxy coating on the surface of a leaf. Water molecules are polar—a characteristic stemming from the way hydrogen and oxygen atoms share electrons. As a result, water molecules stick to each other, a property known as cohesion. Because water molecules are attracted to each other and tend to avoid contact with the nonpolar molecules on the leaf, water beads into spherical droplets despite the constant downward pull of gravity. A droplet at the bottom edge of the leaf clings to it (displaying adhesion) one last moment before gravity pulls it to the ground. We can use the physical and chemical properties of biologically important molecules to explain the related properties at higher levels of organization, giving us amazing insight into how all things work together according to the laws of nature designed by the Creator.

OBJECTIVES

After studying this chapter and completing the exercises, you should be able to do each of the following tasks, using supporting terms and principles as necessary.

SECTION 2.1

1. Describe the major features of two types of intermolecular interactions. Give an example of a substance and its physical properties that result from each type.
2. List and describe seven unique properties of water that result from hydrogen bonding.

SECTION 2.2

3. List the six most common elements found in living organisms.

SECTION 2.3

4. List and draw six of the most common functional groups.
5. Define monomer and polymer.
6. Describe the roles of monomers and polymers in the structures of biomolecules.
7. List and describe the types of monomers that combine to form the four major classes of biomolecules.
8. Describe the purpose of each of the four major types of carbohydrates.
9. Describe the major roles and properties of lipids.
10. Describe the characteristics of the four major groups of amino acids. Give an example of each.
11. Describe the major roles and properties of proteins.
12. Describe the major roles and properties of nucleic acids.
13. Describe the structure of DNA.
14. List three major differences between DNA and RNA.

In Chapter 1, we spend time discussing the hierarchical organization of living things, beginning at the most fundamental level of the atom. Atoms are the building blocks of all matter—including living organisms. These atoms are chemically bonded to one another to form molecules. As you know from your chemistry class, the two major types of chemical bonds are covalent and ionic. Covalent bonds hold atoms together by the sharing of electrons between the different atoms. Covalent bonds are found in molecules, such as oxygen gas and methane. Ionic bonds form as a result of a transfer of electrons from one atom to another. These bonds hold salt crystals together, such as in sodium chloride. In this chapter, we explore several basic but important topics in the chemistry of life.

2.1 Water and Hydrogen Bonding

2.1.1 *Intermolecular Interactions*

In addition to covalent and ionic bonds, there are other, weaker forces or interactions between neighboring molecules—*hydrogen bonding* and *Van der Waals* forces. The stronger of the two interactions is hydrogen bonding. When a hydrogen

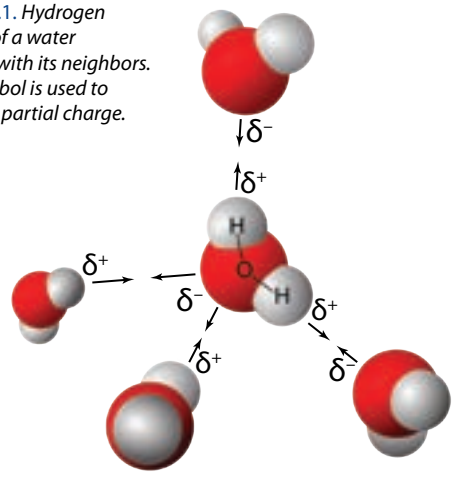


FIGURE 2.1. *Hydrogen bonding of a water molecule with its neighbors. The δ symbol is used to indicate a partial charge.*

FIGURE 2.2. *The gecko can walk up vertical walls due to Van der Waals forces. This gecko is native to Hawaii.*

FIGURE 2.3. *The underside of a gecko's foot on glass, showing the high surface area available for Van der Waals interactions that allow this lizard to scale vertical, smooth surfaces.*

atom is covalently bonded to a highly electronegative element, such as oxygen, nitrogen, or fluorine, the more electronegative atom pulls the electrons in the bond toward itself. The result is a partial positive charge on the hydrogen and a partial negative charge on the electronegative atom. The charge imbalance produces a highly polar molecule that readily interacts with other polar molecules.

Figure 2.1 depicts how this partial charge causes water molecules to be attracted to each other. In fact, each water molecule has room for up to four hydrogen-bonding interactions with its neighbors. One hydrogen bond forms at each of the two hydrogen atoms, while oxygen forms two additional hydrogen bonds on its own. The presence of hydrogen bonding among water molecules leads to a number of fascinating properties, without which life as we know it could not exist. We discuss these properties and their implications soon.

The weakest intermolecular interactions are the Van der Waals forces. These occur between molecules that are mildly polar (Keesom force), between polar and nonpolar molecules (Debye force), and between nonpolar molecules (London dispersion force). In molecules that are nonpolar—i.e., their electrons are equally shared—a temporary polar character can be induced by a nearby polar molecule, or it can form from little random fluctuations in charge that lead to a fleeting, temporary, partial charge. These random fluctuations are caused by the electrons being momentarily denser on one side of the molecule. When this fleeting partial charge occurs, it induces a momentary partial charge in a neighboring molecule, and then the neighbor's neighbor, and so on as electrons rush towards a neighboring partial positive charge. These interactions are quickly formed and quickly dispersed, and they are not nearly so strong as hydrogen bonding. However, they do account for interactions between nonpolar molecules, and can exhibit strength when present in large numbers. In fact, Van der Waals forces give the gecko the amazing ability to stick to a smooth

surface so it can walk up walls, as shown in the photo of Figure 2.2. The highly folded nature of the gecko's foot, shown in Figure 2.3, gives it a large surface area with which to have Van der Waals interactions. Therefore, though a single Van der Waals interaction is relatively weak, the large number of these interactions between the gecko's foot and the vertical surface enable the gecko to fully support its weight.

2.1.2 *The Structure and Properties of Water*

As described above, hydrogen bonding between water molecules causes the molecules to tend to stick together. Interestingly, each instance of hydrogen bonding in liquid water only lasts about 10 ps (picoseconds). The brevity of the bonding contributes to water's fluidity. However, because of the large number of water molecules present at any given time in any sample of water (about 10^{20} molecules per drop!), there is a great deal of hydrogen bonding present at any given time. As a result, water molecules really stick together. Because of the relative strength of hydrogen bonding, water displays seven unique properties that are necessary for life. In this section, we discuss these properties, arranged into four major categories.

Cohesion, Surface Tension, and Adhesion

Cohesion is the tendency for molecules of the same kind to stick together. Water's hydrogen bonding tendencies cause it to be highly cohesive. This property is easily seen in the way water forms beads, as shown in the opening image of this chapter. You can also observe cohesion by slightly overfilling a glass of water and noticing how the water bulges upward without spilling over, as in Figure 2.4. In this case, the cohesive forces between water molecules overcome the force of gravity.

FIGURE 2.4. *Because of its hydrogen bonding tendencies, water displays cohesion—the tendency to stick to itself. Here, a glass of water is slightly overfilled. Notice how the water bulges upward above the rim of the glass.*

Related to cohesion is *surface tension*. Because water molecules tend to stick together, the surface of water has a special resistance to being broken. This resistance, or surface tension, is commonly seen when insects such as water striders literally walk on the water without breaking the surface, as shown in Figure 2.5.

FIGURE 2.5. *Water striders are named for their ability to walk on water.*

While cohesion describes how water sticks to itself, *adhesion* refers to water's tendency to stick to other substances or objects. If you've done any work with laboratory glassware, you are already familiar with adhesion. In Figure 2.6, a sample of water sits in a *buret*, ready to be measured. Because water displays adhesion, some water creeps up the sides of the glass, forming the curved surface of the meniscus. The presence of a meniscus means that a special measurement technique is required when reading volumes in glassware such as burets and graduated cylinders. In order to obtain an accurate reading, you must position yourself so the meniscus is at eye level and read the water level at the bottom of the meniscus.

FIGURE 2.6. *Water in a buret.*

Trees and other plants use cohesion and adhesion working together in a rather magnificent way. In order to survive, a tree

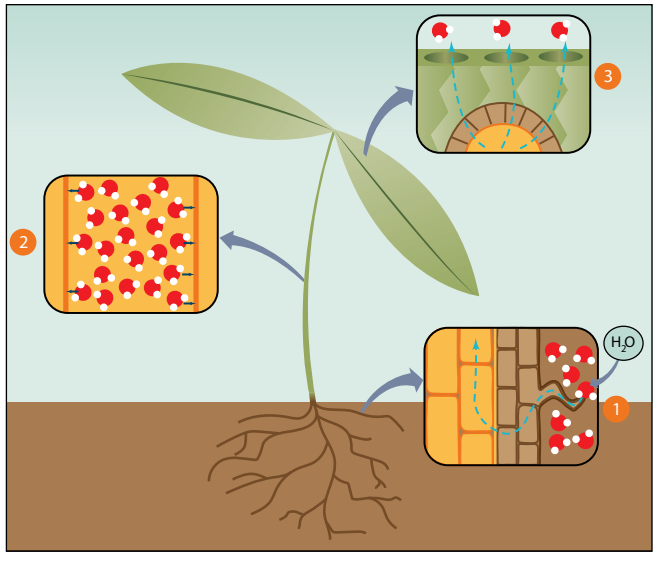


FIGURE 2.7. *Transpiration in plants. 1) Water moves passively into the plant roots. 2) Water molecules stick to the sides of the plant vessels by adhesion. Cohesion holds the water molecules together, forming a column or chain of water. 3) When stomata open, water molecules evaporate, drawing water up through the xylem.*

must pull large amounts of water from its roots up to its leaves on a daily basis. While humans and other animals have hearts to pump blood all over the body, trees have no such organ. In a process called *transpirational pull* (part of cohesion-tension theory), water actually pulls on itself and on the inner tissue of the tree (the xylem) to cause the bulk flow of water in large amounts. This process is illustrated in Figure 2.7. A large oak tree moves about 110 gallons of water (the amount of water held in an extra-large bathtub) from roots to leaves every day.

Thermal Properties: Specific Heat Capacity and Boiling/Freezing Point

Hydrogen bonding also explains water's temperature stability and its unusually large temperature range from melting point to boiling point—both necessary for maintaining life.

When we measure the temperature of a substance, we are in effect measuring the average speed of the molecules. The faster the molecules are moving, the hotter the substance is. Because of the hydrogen bonding in water, water molecules stick to their neighbors. As a result, it takes a lot of energy for molecules to break free of those hydrogen bonds in order to begin moving more quickly. The property of substances relating quantities of heat to changes in temperature is the *specific heat capacity*. The specific heat capacity of a substance is the amount of heat required to raise the temperature of one gram of the substance by one degree Celsius. Because of hydrogen bonding in water, water has a greater specific heat capacity than almost any other substance.

This property is extremely important for life—at both the organism and ecosystem levels of organization. Individual organisms cycle matter and energy through many thousands of chemical reactions. In the process of these reactions, large amounts of heat are released. Because cells have such a high wa-

FIGURE 2.8. *On Earth, water regularly exists in all three phases—solid, liquid, and vapor—often simultaneously. This photo shows a glacier (solid) surrounded by liquid water and vaporized water in the air, some of which has condensed into clouds. (Note that clouds are not water vapor—water vapor is invisible!)*

ter content, most of this heat is safely absorbed without appreciably raising the body temperature of the organism. This is just one way that organisms can maintain homeostasis—keeping body temperature within the narrow range necessary for life.

At the ecosystem level, large bodies of water such as oceans resist changes in atmospheric temperature. This provides a stable environment for marine life, but the large body of water in turn moderates the atmospheric temperature, and thus coastal areas tend to have a more moderate climate than inland areas. In Southern California, this temperature pattern is especially evident during the summers. Cities along the coast tend to remain in the 70s (Fahrenheit), while the farther inland one goes the higher the temperature is. Traveling even 100 miles inland can result in temperatures well above 100°F! (Take-home tip: If you want to cool off, go to the beach.)

Finally, water has a large range of temperatures at which it exists as a liquid. As you know, water freezes at 0°C and boils at 100°C. Most other molecules of similar size boil at very low temperatures and exist mainly as gases. Because of hydrogen bonding, it takes a much higher temperature for water molecules to overcome those attractive forces and jump out of a container to fly away as water vapor. As a result, here on Earth, we regularly find water in all three phases—solid, liquid, and vapor—with all three playing a role in maintaining life. All three phases are shown in Figure 2.8.

Density

Water has a very unusual density pattern. For most substances, solids are denser than liquids, which in turn are denser than gases. This pattern makes sense because molecules occupy more room moving around as they warm up and speed up. In general, faster moving molecules take up more space.

However, in the case of water, ice is actually less dense than liquid water below 4°C (39°F). Figure 2.9 shows the hydrogen bonding pattern of H_2O molecules forming solid ice. In liquid water, the molecules are constantly tumbling over one another, engaging in and moving away from hydrogen bonding with other molecules. But ice forms a stable hydrogen bonding pattern. The ideal geometry of these bonds forces the water mole-

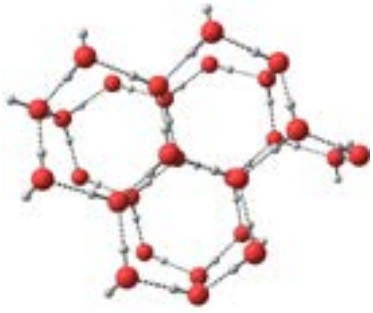


FIGURE 2.9. *The regular, hexagon-shaped, hydrogen-bonded structure of solid ice means that the molecules are farther apart than those of liquid water.*

cules to move farther apart as they approach the freezing point. This means water molecules take up more space in solid ice. As a result, ice is less dense than liquid water and floats.

This property is phenomenally important for maintaining life. In colder climates, lakes freeze over in winter. But because ice floats, only the top of the lake freezes. This top layer of ice then shields the liquid water underneath from the cold temperature, preventing freezing of the entire lake. Fish and other aquatic life survive in the liquid underneath.

Additionally, when the ice melts in the spring, a phenomenon called *spring overturn* occurs. As ice melts, it becomes dense, cold, liquid water and falls to the bottom of the lake. As this denser water falls, it pushes the slightly warmer water originally at the bottom up to the top. This process circulates nutrients and oxygen and is essential for the health of all life within the lake.

Solubility

Finally, because water molecules are polar, water dissolves most other substances that are polar or ionic, earning it the nickname of *universal solvent*. As an example, if you place a spoonful of table salt in a cup of water the polar water molecules begin to pull apart the salt crystal, ion by ion, as illustrated in Figure 2.10. Since water has both partial negative and partial positive charges, these are attracted to their opposite charges in ions in the crystal. The attraction is so strong that it overcomes the ionic attractions in the crystal structure of the salt. Once the salt completely dissociates, each ion is surrounded by water molecules.

Because it is polar, water dissolves a great many substances due to electrostatic attractions between the solvent and solute particles. These solutions are of great importance in biology. Many nutrients cannot be transported or used unless they are first dissolved in water, blood, or other bodily fluids. Also, the interior of cells (the *cytoplasm*) consists of a solution of many useful molecules.

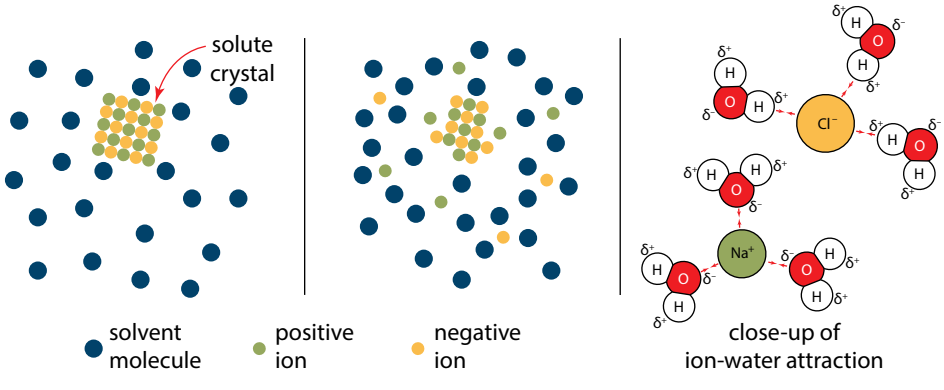


FIGURE 2.10. *Here, water dissolves a salt crystal. The partial positive and partial negative charges of the water molecules attract the negative chloride ions and the positive sodium ions, respectively. In the end, each ion becomes surrounded by water molecules. At this point the salt is in solution, no longer visible to the eye, not even with a light microscope.*

2.2 The Elements of Life

So far as we know, life cannot exist without water. You may already know that water constitutes 50–60% of human body weight. (Interestingly, the averages for adult men and women differ significantly: 58% and 48%, respectively.) Accordingly, the oxygen and hydrogen atoms in water are the most abundant elements in living organisms. The acronym CHNOPS is often used to summarize the six elements that are the basic building blocks for all biological molecules—carbon, hydrogen, nitrogen, oxygen, phosphorus, and sulfur. Figure 2.11 depicts the relative diameters of these key atoms. In all, about two dozen elements are required by living organisms.

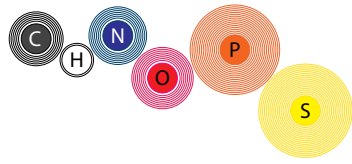


FIGURE 2.11. *Relative diameters of the CHNOPS atoms, the basic building blocks of all biological molecules on Earth.*

Of the atoms in the human body, 62% are hydrogen and 24% are oxygen. Carbon is the next most common element, contributing 12% of the atoms. The next is nitrogen, making up 1.1% of the atoms in the human body. Percentages for the next few elements are: calcium (0.22%), phosphorous (0.22%), sulfur (0.038%), sodium (0.037%), potassium (0.03%), chlorine (0.024%), and magnesium (0.015%). Notice that even though calcium is not in the CHNOPS list, its abundance in the human body exceeds that of phosphorus and sulfur. This is because calcium comprises 39% of the atoms in our bones.

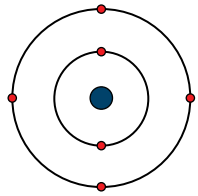


FIGURE 2.12. *Bohr model of carbon, showing four valence electrons available for bonding.*

2.3 Biomolecules

2.3.1 *Functional Groups*

Carbon is a particularly important element in biology because of its central role in building biomolecules—large molecules composed of repeating arrays of smaller molecules. Generally, any molecule containing carbon is called an *organic molecule*.[1] The study of organic chemistry focuses on the science of organic molecules—their composition, synthesis, and how they react with other molecules. This central role of carbon in organic molecules is due to the unique structure of the carbon atom.

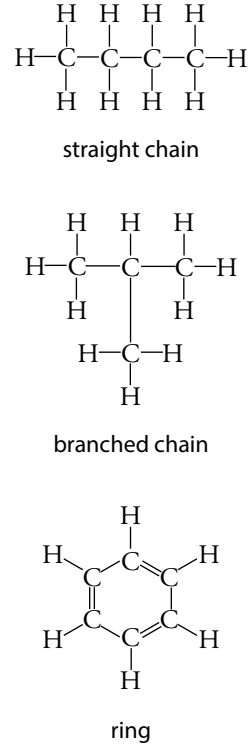


FIGURE 2.13. *Some examples of carbon's versatility in bonding.*

As the Bohr model in Figure 2.12 depicts, carbon atoms have four valence electrons, the electrons in the valence shell available to participate in chemical bonding. This means that carbon atoms can form up to four covalent bonds with atoms of other elements or with other carbon atoms. One of the most basic organic molecules is the *hydrocarbon*. This is a molecule composed of a chain of carbon atoms with hydrogen atoms attached to the carbons. Carbon can form single, double, and triple covalent bonds, in which one, two, or three pairs of electrons are shared. The covalent bond between carbon and hydrogen is always a single bond. Additionally, carbon atoms

1 There are a few exceptions, such as CO, CO_2, carbides, some carbonates, and molecules of pure carbon. These are regarded as inorganic molecules.

can bond together in straight chains, branched chains, or rings, as illustrated in Figure 2.13. Carbon is thus an extremely versatile element and forms the basis for almost all biologically relevant molecules.

The chemistry of organic compounds makes routine use of special groups of atoms bonded together to form groups with unique chemical properties. These groups of atoms are called *functional groups*. There are several common functional groups that can be found on many different molecules. Below are descriptions of some of the most common functional groups in biological systems. These are summarized in Table 2.1. In the formulas below, R represents the molecule the functional group is attached to, typically a hydrocarbon or other organic compound. R′ represents another generic molecule, perhaps different from R.

1. *Hydroxyl group* (R—OH) This functional group is composed of an oxygen atom and a hydrogen atom. It is a polar functional group, which means it exhibits hydrogen bonding with water. Some examples of molecules that contain hydroxyl groups are alcohols, carbohydrates, steroids, and proteins.
2. *Methyl group* (R—CH_3) The methyl group is a central carbon surrounded by three hydrogen atoms. It is a nonpolar functional group and is found in DNA, some proteins, and some carbohydrates.
3. *Carbonyl group* (R—CO—R′) The carbonyl group is a carbon atom, found within a hydrocarbon or other large molecule, that attaches to an oxygen atom by means of a double bond. It is a polar functional group, with the carbon being poor in electron density and the oxygen rich in electron density. This means that an electron-rich atom may react with the carbon to lengthen the molecule while the oxygen gets a hydrogen to make a hydroxyl. The carbonyl is found in other functional groups such as carboxylic acids. Carbonyls are common in steroids, fats, and proteins.
4. *Carboxyl group* (R—COOH) This functional group is composed of a carbon atom bound to two oxygen atoms—to one by means of a double bond, and to the other with a single bond. The oxygen with the single bond is also attached to a hydrogen atom. You can think of this group as a combination of carbonyl and hydroxyl groups. Carboxyls tend to give up the hydrogen atom in aqueous environments, which means they are acids. They also play an important role in the formation of covalent bonds between the amino acids of proteins.
5. *Amino group* (R—NH_2) Amino groups consist of a nitrogen atom bound to two hydrogen atoms. They are capable of accepting a third hydrogen atom under certain conditions, which means they are weakly basic molecules. Ami-

nos, like carboxyls, play an important role in the formation of peptide bonds between amino acids.

6. *Phosphate group* (R—PO_4^{2-}) The phosphate group contains a single phosphorous atom surrounded by four oxygen atoms. It is a negatively charged, polar functional group found in DNA, the energy molecule ATP, and a special group of lipids called phospholipids.

7. *Sulfate group* (R—SO_4^-) The sulfate group is composed of a sulfur atom bound to four oxygen atoms. It is a negatively charged, polar functional group found in carbohydrates, proteins, and lipids.

Complete familiarity with these functional groups is essential in the study of biochemistry. Scientists identify different classes of molecules based on the presence or absence of certain functional groups on the molecule. We make note of some of these in the molecular descriptions in the next section.

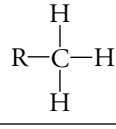
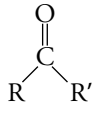
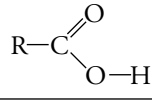
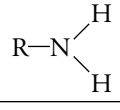
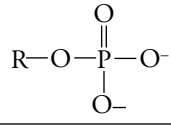
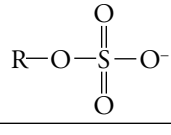
Functional Group	Formula	Lewis Structure	Where Found	Properties
hydroxyl	R—OH		in alcohols, steroids, proteins, and carbohydrates	polar, exhibits hydrogen bonding
methyl	R—CH_3		attached to DNA, amino acids, carbohydrates	nonpolar
carbonyl	R—CO—R′		in steroids, amino acids, and waxes	polar, exhibits hydrogen bonding
carboxyl	R—COOH		in fatty acids and all amino acids	forms peptide bonds, acidic
amino	R—NH_2		in all amino acids	forms peptide bonds, basic
phosphate	R—PO_4^{2-}		in all phospholipids, nucleic acids, and ATP	polar, negatively charged
sulfate	R—SO_4^-		attached to carbohydrates, proteins, lipids	polar, negatively charged

TABLE 2.1. *Functional groups common in biological molecules.*

2.3.2 *Monomers and Polymers*

There are four classes of biomolecules. Many biomolecules are built up by repeating smaller units. The small molecules are called *monomers*, from the Greek *mono* meaning single, and *meros* meaning part. The larger *polymers* (meaning many parts) are built up by chaining many monomers together. Monomer subunits are joined together through a process called *dehydration synthesis*. In this reaction, a hydroxyl group is removed from one monomer and a hydrogen is removed from the other monomer to form a water molecule. This results in the formation of a covalent bond joining the two monomers together. This process is easily reversed through a *hydrolysis* reaction in which water enters and breaks the covalent bond. These two reactions are used to build polymers and break polymers apart respectively. (See also Figure 2.17 below.)

In Sections 2.3.3 and 2.3.5, we describe the four classes of biomolecules. In between these sections, Section 2.3.4 introduces amino acids.

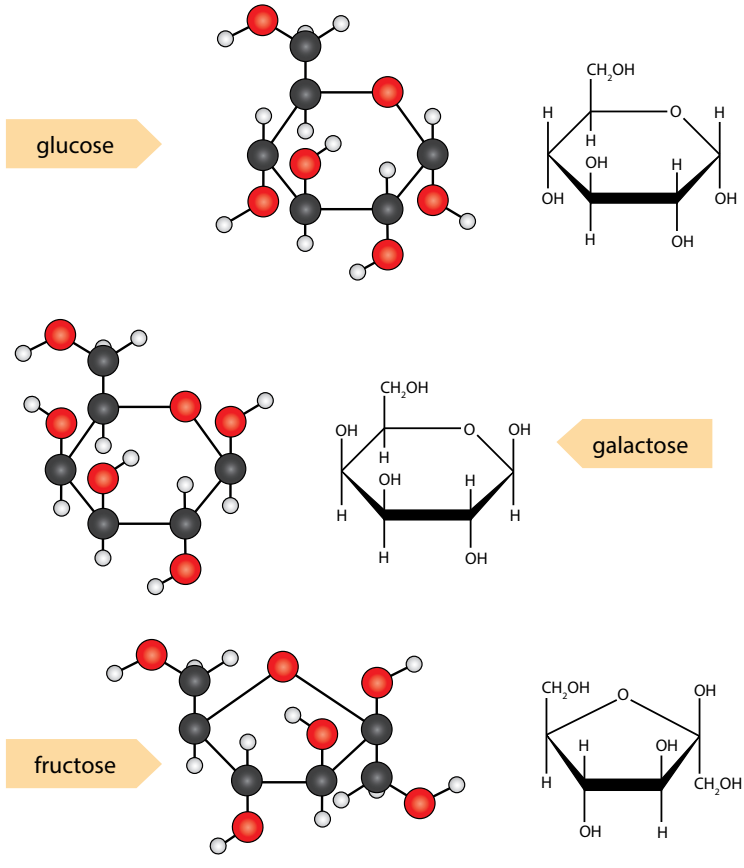


FIGURE 2.14. *Chemical structures of glucose, galactose, and fructose, each displayed with two types of models: ball-and-stick models and line diagrams. All are monosaccharides. Carbon atoms are shown in black or charcoal gray in ball-and-stick models. Carbon atoms typically are not shown in the corresponding line diagrams.*

2.3.3 *Carbohydrates and Lipids*

Carbohydrates

The first class of biomolecule to consider is the *carbohydrates*. These are composed of monomers incorporating carbon, oxygen, and hydrogen atoms in the ratio 1:2:1, i.e., CH_2O, hence the name *carbo–* (meaning carbon) and *hydrate* (referring to water).

The monomers of carbohydrates are called simple sugars or *monosaccharides* (meaning single sugar). These sugars are generally composed of a ring with five or six carbon atoms and an oxygen atom, with a hydroxyl group attached to each carbon atom. Three common sugars are glucose, galactose, and fructose. These differ in their placement of the hydroxyl groups relative to one another and the shape of their rings, as depicted in Figure 2.14.

Glucose is a sugar the body breaks down to produce energy and is a major component of some foods. Fructose is found in many fruits, as well as in honey. Sucrose—table sugar—is a *disaccharide* (two simple sugars connected together) of glucose and fructose. Figure 2.15 shows the chemical structure of sucrose.

When many sugars are joined together, a *polysaccharide* is formed. When vast numbers of sugars chemically combine, the result is a carbohydrate biomolecule. There are four major types of carbohydrates, described below.

1. *Starch* Starch is a polymer of glucose, formed as a mixture of two types of chains: amylose and amylopectin, represented in Figure 2.16. Starch is the major dietary source of glucose, and is found in rice, bread, potatoes, corn, and

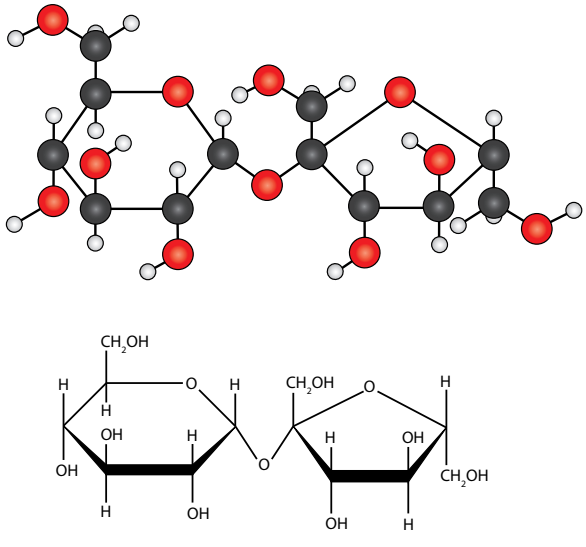


FIGURE 2.15. *Sucrose (table sugar) forms from the chemical reaction that joins glucose and fructose. (The orientation of the fructose part of this molecule is upside down compared to the fructose molecule illustrated in Figure 2.14.)*

other foods. Our bodies break down starch by means of the hydrolysis reaction depicted in Figure 2.17 *(hydro* = water, *lysis* = break down).

2. *Glycogen* Glycogen, depicted in Figure 2.18, is a branched-chain polymer of glucose, found mainly in the liver and muscles. Its purpose is to store glucose for future energy usage. Many marathon or long-distance runners "carb load" the day before a big race. By eating large quantities of starch, their bodies store the excess glucose in the form of glycogen. During the race, as the glucose in their systems runs out their bodies begin breaking down glycogen.
3. *Cellulose* Cellulose, depicted in Figure 2.19, is a straight-chain (unbranched) polymer of glucose, in which the glucose monomers are bonded in a different orientation than they are in dietary starch. This orientation allows hydrogen bonding to occur between neighboring strands, resulting

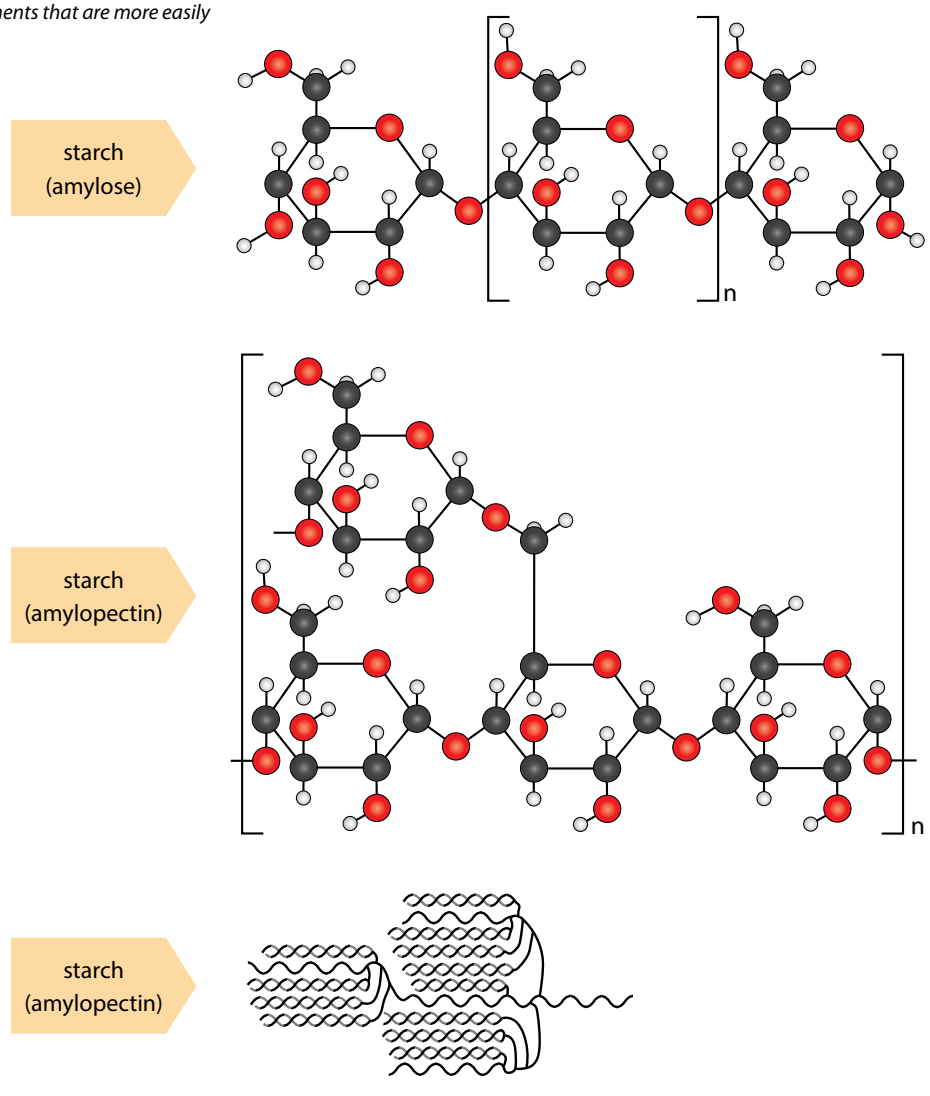


FIGURE 2.16. *The chemical structure of starch. Starch is a polymer of glucose, comprising a mixture of two types of chains: amylose (top) and amylopectin (middle and bottom). The* $[\,]_n$ *notation means the glucose monomer is repeated in a chain n times, where n varies widely—many thousands of glucose units are possible in a single chain. Amylose consists of straight (unbranched) chains in a helical structure; it is more soluble in water (because the hydrophobic regions are inside the helix), but more difficult to digest. Amylopectin consists of branched chains, also in a helical structure, and is more digestible (because the branches expose segments that are more easily broken).*

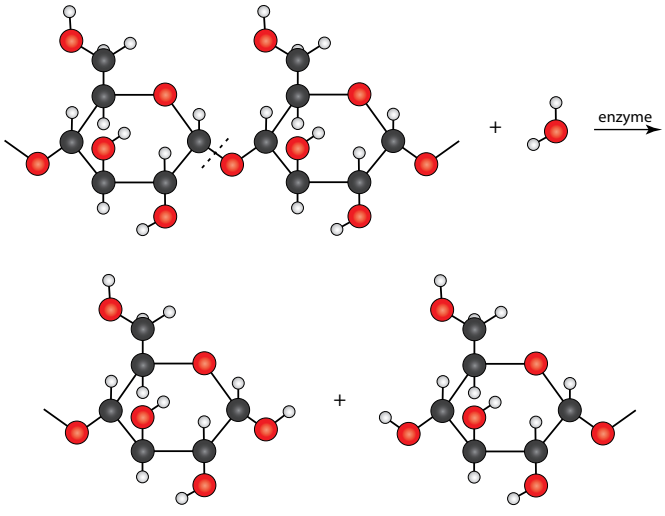


FIGURE 2.17. *Hydrolysis: With the addition of a water molecule and an appropriate enzyme, the starch polymer is broken down into glucose. (For simplicity, only two monomers in the starch molecule are shown.)*

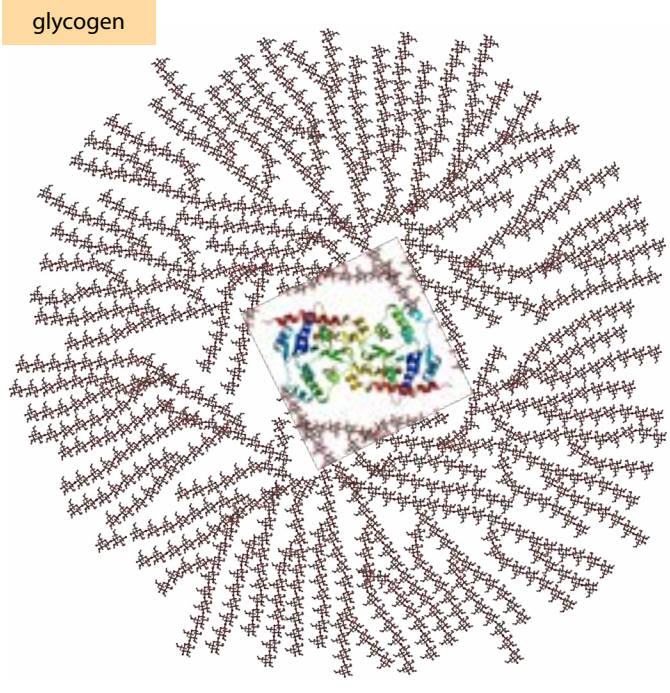


FIGURE 2.18. *Glycogen, another branched chain polymer of glucose, with the branches held together in the center by a protein molecule to form one large molecule.*

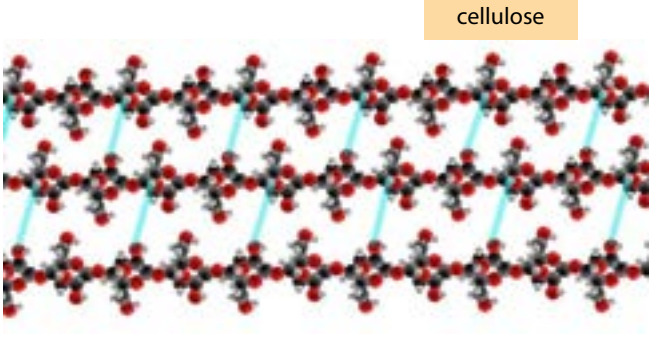


FIGURE 2.19. *Cellulose. Its chemical structure is similar to that of starch, but the different orientation of the monomers makes cellulose indigestible by humans. Blue lines indicate hydrogen-bonding sites between strands.*

in an extremely strong fiber that gives strength to plants and trees. Cellulose is a major component of celery, making it difficult to chew. The human body is not able to digest cellulose, so celery has little caloric value. (It does, however, contribute many helpful compounds such as vitamins and antioxidants.)

4. *Chitin* The glucose monomers in this carbohydrate are linked together in the same indigestible manner as those of cellulose. However, one of the hydroxyl groups on each glucose monomer is replaced by an acetyl amine group ($HNCOCH_3$, shown at the bottom of Figure 2.20). These groups allow for hydrogen bonding between chains, giving chitin (pronounced KAI-tin) the ability to form a strong, flexible, two-dimensional sheet. This makes chitin the perfect material for the exoskeleton of insects and crustaceans, as well as in the cell wall of some fungi. If you have ever pulled the tail off a shrimp before eating it, you know what chitin feels like. The outer shell (exoskeleton) of crayfish, such as the one shown in Figure 2.21, is composed of chitin.

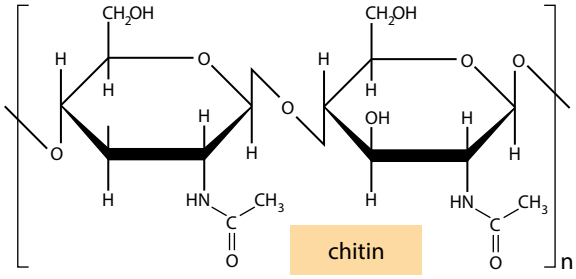


FIGURE 2.20. *Chitin. Hydrogen bonding between polymer chains enables chitin to form a strong, flexible, two-dimensional sheet.*

One universal feature of carbohydrates is that the many attached hydroxyl groups make them polar molecules, just like water molecules. As a result, we call carbohydrates *hydrophilic* (water-loving). The next class of molecules is just the opposite.

FIGURE 2.21. *A crayfish, whose strong yet flexible exoskeleton is composed of the carbohydrate chitin.*

Lipids

The *lipids* is a class of molecules that are *hydrophobic* (water-fearing). These molecules consist mainly of hydrocarbon chains and are nonpolar. Their electrons are shared and distributed evenly around the molecule, so that no partially charged regions are created. Molecules in this class do not dissolve in water.

There are three major types of lipids, described below.

1. *Triglycerides* These molecules consist of three *fatty acids*—modified hydrocarbon chains, depicted in Figure 2.22—attached to a small molecule called *glycerol*, depicted in Figure 2.23. In animals, lipids in the form of triglycerides store energy for long-term use. Fatty acids are described as *saturated* or *unsaturated*. In saturated fatty acids, all the carbon-carbon bonds are single bonds and a pair of hydrogen atoms is attached to each carbon (except the last carbon, which has three hydrogen atoms attached). This arrangement represents the greatest possible number of hydrogen atoms in a carbon chain, hence the term "saturated." The fatty acid shown in Figure 2.22 is a saturated molecule. Saturation results in straight carbon chains that pack tightly together. Because of the tight packing, these fats are solids at room temperature. Animal fats, such as butter or bits of fat that one trims from a piece of meat, are composed

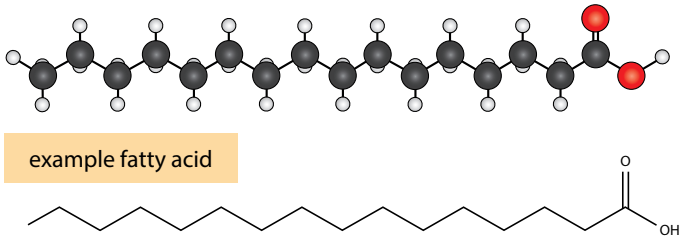


FIGURE 2.22. *An example of a fatty acid. The left side consists of a long nonpolar chain of carbon and hydrogen. The right end has a polar acid group that attaches to glycerol in order to make a triglyceride.*

of saturated fats. In unsaturated fats, one or more double bonds are present between the carbon atoms in the chain, with a single hydrogen atom bound to each carbon joined by the double bond. An example of an unsaturated structure is depicted in Figure 2.24. Double bonds kink the carbon chain so that it bends, preventing the fat molecules from being so close to one another. As a result, unsaturated fats tend to be liquids at room temperature; those that are liquids are called *oils*. Unsaturated fats are often derived from plants. Olive oil, avocado oil, and vegetable oil used for cooking are examples of unsaturated fats.

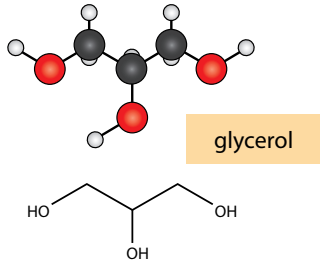


FIGURE 2.23. *Glycerol, the molecule to which three fatty acids attach to make a triglyceride.*

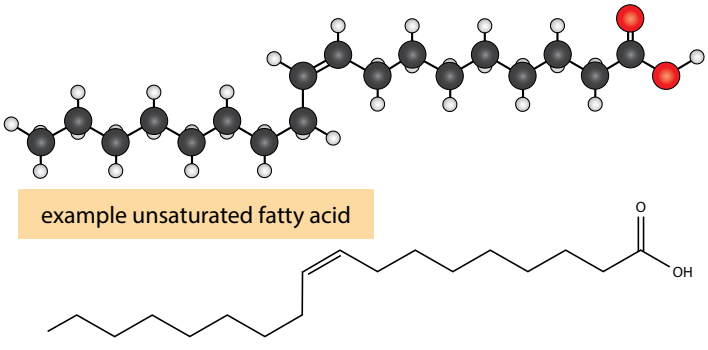


FIGURE 2.24. *An example of an unsaturated fatty acid. Unsaturated fatty acids have one or more double bonds between carbon atoms, making the resulting molecules less tightly packed than in saturated fats and more likely to be a liquid at room temperature.*

2. *Phospholipids* Phospholipids consist of only two fatty acids bound to a glycerol molecule, with the third spot being taken by a phosphate group, as depicted in Figure 2.25. These fats have the property of being hydrophobic at one end (where the fatty acids are) and hydrophilic at the other end (where the phosphate group is). The result is that this molecule forms a *lipid bilayer*, the major component of the cell membrane (see Figures 3.41 and 3.42). Since all cells are enclosed by a cell membrane, the ability of phospholipids to form a lipid bilayer is an extremely important property for biochemistry.

3. *Steroids* Steroids are relatively small molecules made of carbon rings. One steroid you have probably heard of is *cholesterol*, shown in Fig 2.26A. Found on the nutrition labels of many foods, this molecule is also a component of the cell membrane. Two other steroid molecules are testosterone and estrogen, shown in Figures 2.26B and 2.26C. These are the sex hormones that control some of the functions specific to male and female physiology.

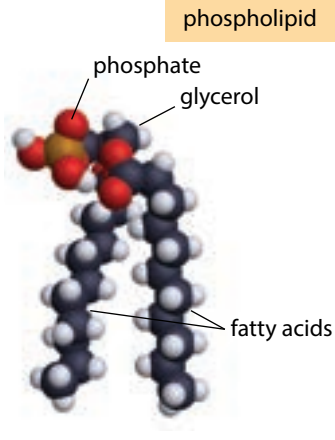


FIGURE 2.25. *A phospholipid molecule. The phosphate group is shown in orange and red.*

FIGURE 2.26A. *Cholesterol is a steroid that is an important component of the cell membrane. Note the connected carbon rings, an essential part of steroid structure.*

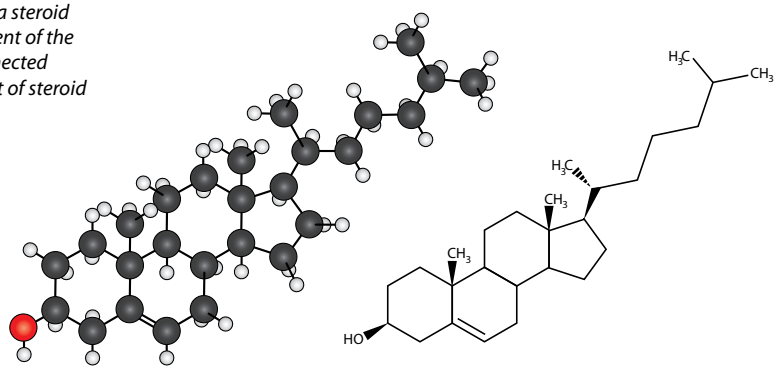


FIGURE 2.26B. *Testosterone, a steroid that functions as the major male sex hormone.*

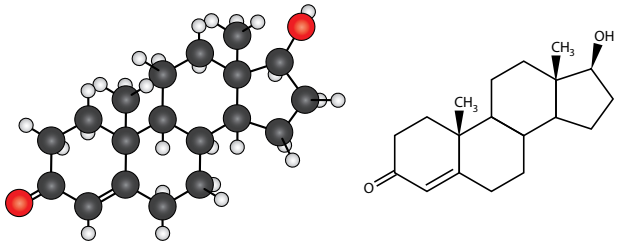
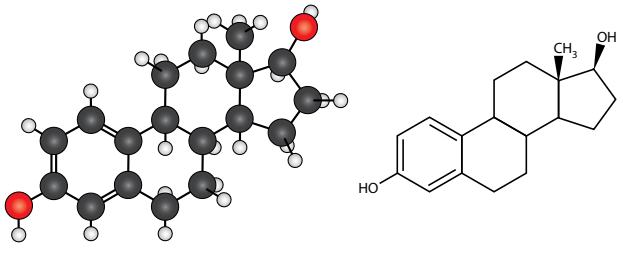


FIGURE 2.26C. *Estrogen. Estradiol, pictured here, is the predominant form of estrogen that is active during a woman's reproductive years.*



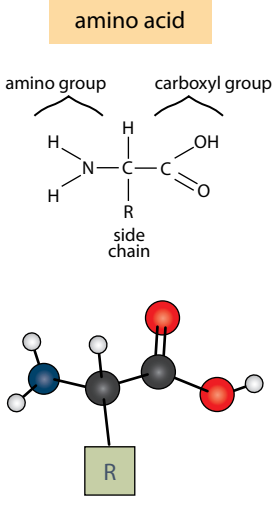


FIGURE 2.27. *General structure of an amino acid.*

The structures of the carbohydrate and lipid molecules we have discussed are fairly uniform. Starch is composed of many repeating units of identical glucose monomers. Similarly, triglycerides contain three nonpolar fatty acid molecules, and fatty tissue is a collection of similar lipid molecules. In contrast, in the next two sections we explore classes of biomolecules in which the monomers are less similar, giving rise to greater diversity of both form and function.

2.3.4 *Amino Acids*

Proteins are a class of biomolecule like no other. The human body is capable of producing some two million different proteins, yet this vast array of molecules is built from an "alphabet" of just 20 different monomers, called *amino acids*. The basic structure of the amino acids is shown in Figure 2.27. Each amino acid has an identical backbone, consisting of a central carbon atom, an amino group on one end, and a carboxyl group on the other, so the generic amino acid formula can be written R-CH(NH_2)COOH. The structures of the 20 amino acids are shown in Figure 2.28.

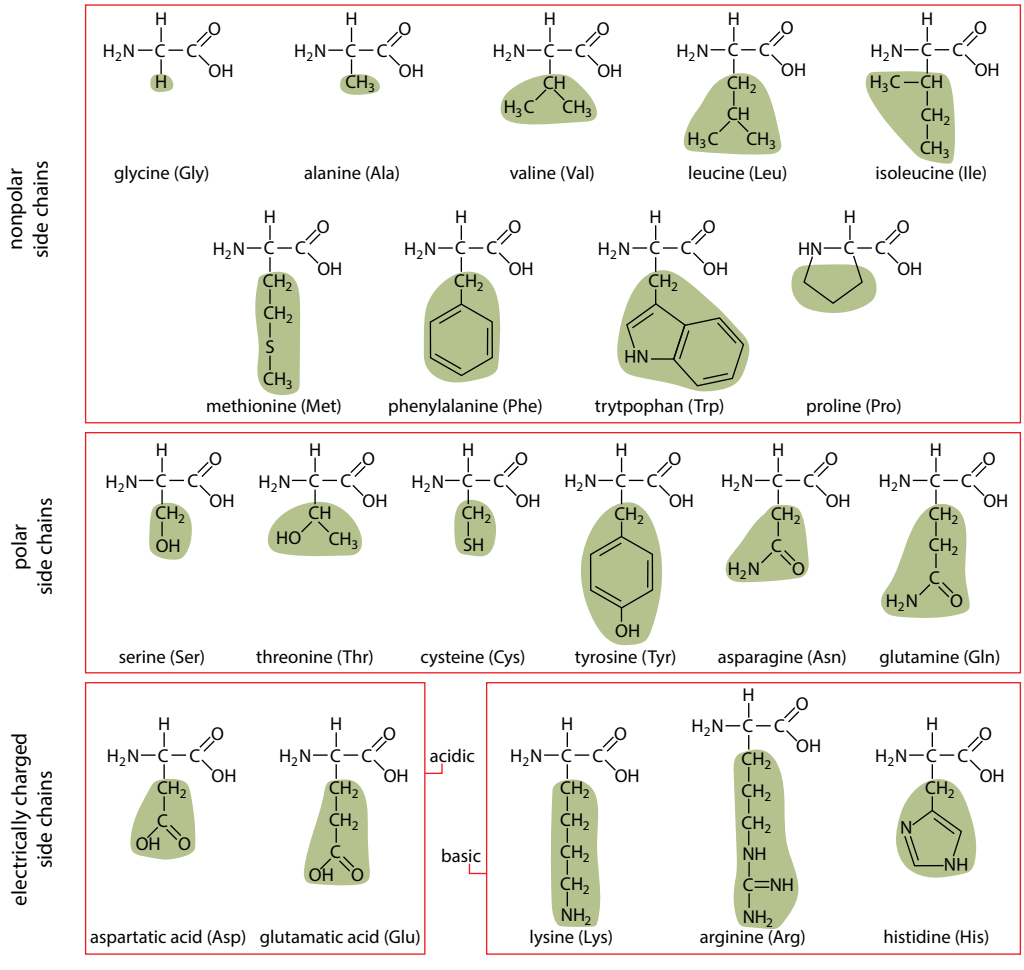


FIGURE 2.28. *The 20 amino acids that combine to construct proteins. Side chains are indicated by a green background.*

The difference between the amino acids is the "R" group, also called its *side chain*. The twenty amino acids can be organized into four different groups. The largest group of amino acids comprises those that have nonpolar side chains. Some nonpolar amino acids, such as alanine and glycine, have very small side chains, while others have large, bulky side chains composed of rings of hydrocarbons. The largest one of these is the amino acid tryptophan.

A second group of amino acids are those with polar side chains. These six amino acids contain side chains with hydroxyl, carbonyl, or amino groups that contribute to their polar character. (Both the polar and nonpolar groups of amino acids have no net charge; they are neutral.) One unique amino acid in this group is cysteine, which has a sulfur atom on its side chain. Two cysteine amino acids can form a special covalent bond between them called a *disulfide bond*. Insulin, a protein hormone that helps regulate the levels of glucose in our blood, is one molecule in which disulfide bonds are put to use. It is a small protein composed of two polypeptide chains—the A-chain, composed

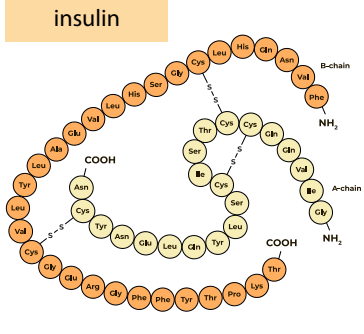


FIGURE 2.29. *The protein structure of insulin. In the active form of insulin, disulfide bonds hold the two polypeptide chains together.*

of 21 amino acids, and the B-chain, composed of 30 amino acids. These two chains are held together by two disulfide bonds to produce the active form of insulin, pictured in Figure 2.29.

The amino acids in the remaining two groups each have a positively or negatively charged side chain. Amino acids that have a positively charged side chain are called basic amino acids. The side chains of these amino acids contain more amino groups than carboxyl groups. The amino group (—NH_2) can accept a hydrogen ion (—NH_3^+), giving the side chain a positive charge. In contrast, amino acids that have a negatively charged side chain are considered acidic amino acids. These amino acids contain more carboxyl groups than amino groups on their side chains. The carboxyl groups (—COOH) can give up a hydrogen ion (—COO^-), resulting in a negatively charged side chain.

The amino acids exhibit a vast array of physical and chemical properties, making them the perfect components for producing the proteins needed to perform the huge number of functions that occur in the bodies of complex organisms, such as ourselves. Just as the English language has an alphabet of 26 letters that combine to make millions of words, which can be combined to create a practically infinite number of writings of diverse meanings, the amino acids combine in different ways to produce a similarly fantastic array of diverse proteins. Short chains of amino acids are called *peptides*; longer chains are called *polypeptides*.

2.3.5 *Proteins and Nucleic Acids*

Proteins

We have seen that sugars join together to create complex carbohydrates. In a similar way, proteins are formed by chains of amino acids, connecting the amino group of one amino acid to the carboxyl group of the next. However, the sequence of the amino acids to be joined is of crucial importance, and is thus exquisitely designed and controlled. We discuss this process in a later chapter. For now, we focus on the major structures and functions of proteins. The main idea is that a protein's amino acid sequence determines its three-dimensional structure, and its structure determines its function.

The making of a protein molecule begins with a single, straight chain of amino acids—a polypeptide. The sequence of amino acids present in this single chain defines the protein's *primary structure*. Once the polypeptide is assembled, or even while it is being assembled, it begins folding into a 3-D structure that gives it machine-like abilities. The *secondary structure* refers to some common 3-D structures that can occur in parts of a polypeptide chain. Two types of secondary structure are the *alpha helix* and *beta sheet*, both of which rely on regular patterns of hydrogen bonding within the chain of amino acids. In the alpha helix, every backbone N – H group hydrogen bonds to

In the Wonderworld of Biology

Does Eating Protein Promote Healing?

You may have heard people say that you should eat lots of protein after an injury because the body needs protein to repair itself. Is this correct? If so, how does this process work? It might be tempting to think that our bodies directly take the proteins of beans, cows, or chickens and use them to cover our wounds like some sort of molecular band-aid. However, the real story is a bit more complicated. It turns out that eating protein does promote healing, but in a more complex, elegant fashion.

Whenever we ingest animal or plant protein, we chew the food in our mouths to increase the surface area, and then the specialized environment of the stomach takes over. In this highly acidic organ, large protein molecules (with tertiary or quaternary structure) are first denatured[1] by the acid, and then cut into smaller polypeptide chains by an enzyme called pepsin. As the partially digested food proceeds to the small intestine, the pancreas secretes the enzymes trypsin and chymotrypsin. These enzymes digest the polypeptides into even smaller pieces by preferentially cutting at specific amino acids. At this point, much of the protein is degraded into individual amino acids that are absorbed into the bloodstream by special cells called enterocytes. The absorbed amino acids are transported all over the body for use by the cells—and they are especially needed at the site of a wound.

When our bodies heal an injury, molecular building blocks are needed by healthy cells in order to divide and generate new cells to replace the damaged tissue. One major need is for amino acids to form new proteins according to the instructions in our DNA. Some of the 20 amino acids used to build proteins can be synthesized from other molecules in our bodies; these are called nonessential amino acids. In contrast, essential amino acids cannot be synthesized by our bodies and must be obtained from our diets. Both types of amino acids are needed in the correct proportions to keep protein synthesis moving along—and so it is important to be mindful of eating a variety of foods, especially when relying on only plant-based protein sources. Finally, the body needs a source of nitrogen for nucleotide synthesis, so that cells may continue copying DNA as each cell division occurs. In other words, healing from an injury is costly in terms of new cell growth and the need to synthesize new DNA and new proteins.

The building blocks supporting tissue growth—amino acids and the nitrogen for biosynthesis—mostly come from dietary protein. If these molecules aren't supplied from the diet, a condition called negative nitrogen balance ensues, when the body breaks down muscle tissue in order to supply the necessary amino acids.

Other food sources, such as carbohydrates, fats, and many vegetables, are important sources of calories, minerals, and vitamins, but protein is the key supplier of the precursors to rapid cell division that support the healing of a wound.

In summary, after an injury you can enjoy your legumes, omelets, cheese, and steaks knowing that these sources of protein are supplying your body with the amino acids and nitrogen it needs to synthesize the proteins required for all the new cells used in healing a wound. And for you vegetarians out there, eating a varied diet of multiple protein sources such as lentils, tofu, nuts, tempeh, and all kinds of beans works just as well.

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1 With respect to proteins in biology, denaturing refers to breaking weak, noncovalent interactions or hydrogen bonds that give a protein its structure, causing it to unfold. Denaturing causes a protein to lose its quaternary, tertiary, and secondary structure, only retaining its primary structure.

FIGURE 2.30. *An alpha helix—a type of secondary protein structure. Two alpha helices of the protein keratin are depicted here.*

the backbone C = O group four monomers earlier in the same chain, creating a helical structure. In the beta sheet, hydrogen bonding occurs between the same two groups located on separate, parallel strands, creating a flat structure. Figure 2.30 shows a model of an alpha helix. The two helical ribbons in the diagram are simply symbolic for the long, complex, helically shaped molecule. The ribbon diagram is superimposed over the atoms in the illustration—a common way that biochemists indicate the presence of an alpha helix in models of 3-D protein structures.

Tertiary structure describes the fully formed 3-D structure of a single polypeptide chain. The final shape, such as in the example shown in Figure 2.31, is the product of many different chemical interactions between the R side chains. These interactions include those described in Section 2.1.1—covalent bonds, ionic bonds, hydrogen bonding, and Van der Waals interactions.

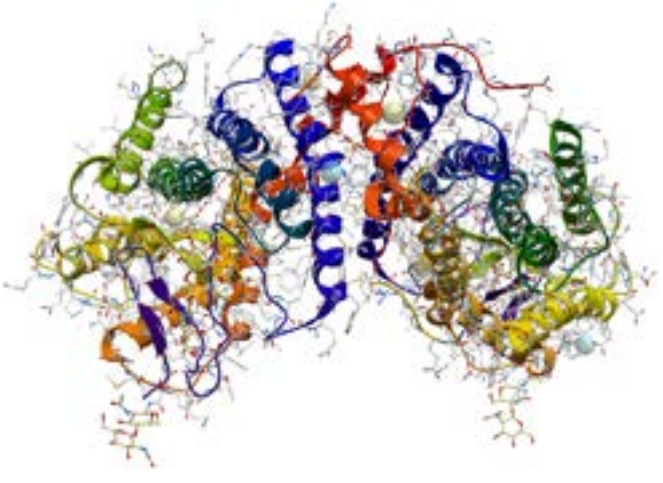


FIGURE 2.31. *Tertiary structure of a protein, showing the 3-D interactions of all the amino acids. Note the ribbon indication of alpha helices as well as the flat, lime-green ribbons representing beta sheets. Underneath the ribbon notation, you can see the actual atomic/chemical structure. This protein is called rhodopsin—a protein involved in vision under low-light conditions.*

Finally, *quaternary structure* is reserved for those proteins whose functions depend on several individual polypeptides, called *subunits*, interacting with one another to perform the final function, as illustrated in Figure 2.32. These subunits often associate with one another through hydrogen bonding or Van der Waals interactions.

Each protein has its own unique structure that arises from the sequence of amino acids in the polypeptides If there is a change in a single amino acid, the protein structure may lose its integrity and even its function. Let's look at a well-known example involving the protein hemoglobin, illustrated in Figure 2.33. Hemoglobin is a protein consisting of four subunits, two

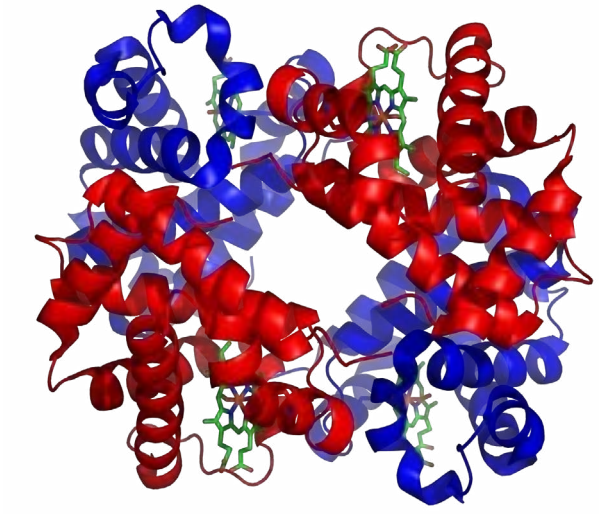
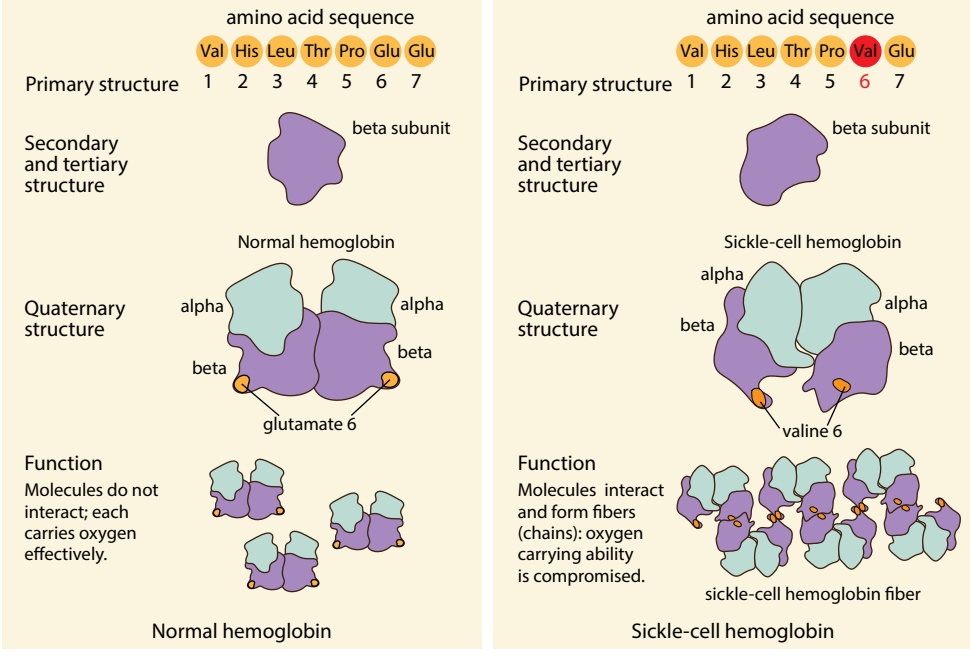


FIGURE 2.32. *Quaternary structure, in which multiple protein subunits come together to create a single functional protein. Pictured here is hemoglobin, the protein that carries oxygen in red blood cells. Hemoglobin has four subunits (separate polypeptide chains), bonded to four heme molecules that each carry oxygen. The subunits are colored red and blue. The heme molecules are shown in green. For clarity, only the ribbon structure is shown for most of the protein, and not the underlying atomic structure.*

alpha subunits and two beta subunits. Hemoglobin within red blood cells carries oxygen. The sixth amino acid in the beta subunit polypeptide is glutamic acid, a polar acidic amino acid. The genetic disease known as sickle cell anemia results from the sixth amino acid being valine instead of glutamic acid. Valine is a nonpolar amino acid. The presence of the valine results in the misfolding of the beta chain of hemoglobin, resulting in a small protrusion in the subunit. As a result, the abnormal hemoglobin attaches to other misshapen hemoglobin molecules, forming a chain, which then crystalizes into firm fibers inside the red blood cells. These fibers cause the red blood cells to distort and assume the sickle shape that gives the disease its name.

This example illustrates the high degree of specificity in a protein's 3-D structure, and therefore its function in an organ-

FIGURE 2.33. *A change in the amino acid sequence of hemoglobin results in an improperly folded protein, causing sickle cell anemia. Note that glutamate is the ionized form of glutamic acid.*



ism. Protein structure can be sensitive to changes in temperature, pH, and salt concentration. Without finely tuned conditions, proteins fall apart, or *denature*—they lose their 3-D shape, resulting in a loss of function. This is just one reason why it is so important for an organism to maintain homeostasis. Otherwise, all its molecular machines come to a screeching halt! Proteins have different temperature or pH requirements, depending on which part of the body they reside in or which organism they are part of. For example, the chemical environment in the stomach is highly acidic, with a pH range of 1.5–3.5. Enzymes in the stomach that participate in digestion of proteins and other molecules are correctly folded for operation in these acidic conditions. In fact, these stomach enzymes are within their optimum pH range in the stomach and do not function as well in an environment with a neutral pH. By contrast, proteins that exist in the blood, such as hemoglobin, are perfectly suited to form their optimum 3-D shape at a pH in the range of 7.35–7.45 and can be much less effective in other pH ranges. Think about how many of these finely tuned parameters must govern every single enzyme in your body in order for you to be alive. Now extend that to every enzyme present in every living creature on earth. Multitudes of writers have noted the fine-tuning found everywhere in creation, and this divine fingerprint is certainly everywhere to be seen in living organisms.

Now that we have seen how proteins are structured, let's look at some of the various functions that proteins perform within a cell.

1. *Enzymes* An enzyme is a *catalyst*—a molecule that facilitates a chemical reaction without itself being consumed. Your body is a fantastic chemical factory, employing many thousands of chemical reactions—those that turn food into energy, those that build up tissue, those that control growth and development, and many others. To ensure that these reactions occur in a controlled manner, they are controlled by enzymes. As illustrated in Figure 2.34, these enzymes bind to particular chemicals, called *substrates*, that place them in just the right direction to react with other substrates and then release a newly formed product, ready to begin the process again. Without an enzyme to help, most chemical reactions in the body would simply take too long, being too energetically unfavorable to occur. Enzymatic control of chemical reactions means that every process can be finely tuned, helping the body to maintain homeostasis.
2. *Transporters* Many substances must cross cell membranes in order to keep the body running smoothly. Proteins act as transporters to facilitate this process.
3. *Signaling Proteins* Cell membranes have a number of proteins that relay signals from other cells to the cell's interior. Other proteins may form a complex cascade of signaling pathways.

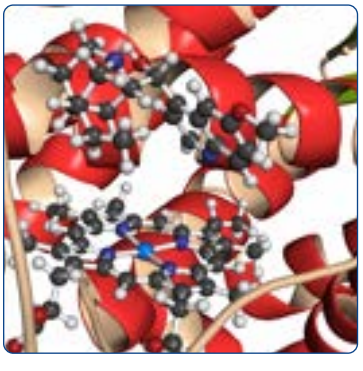


FIGURE 2.34. *Substrates (shown as ball-and-stick molecules) bind to enzymes (shown as ribbons). Enzyme-substrate interactions are very specific, and are often mediated by exactly placed hydrogen bonds and Van der Waals interactions.*

4. *Structural Proteins* Proteins are the structural components in the body. These include collagen in the extracellular matrix; keratin in hair, nails, and intermediate filaments; and other proteins in the cytoskeleton filaments extending throughout the interior of all cells and giving them their shape.
5. *Motor Proteins* Proteins enable our bodies to move. These proteins include myosin for muscle contraction, kinesin for vesicle transport, and dynein for cell division.
6. *Regulatory Proteins* Proteins such as antibodies, hormones, and receptors regulate and control processes throughout the body.

These are just a few of the many tasks performed by proteins—the workhorses of the cell.

Nucleic Acids

The fourth major class of biomolecules is the *nucleic acids*. Of these, there are two major types—DNA and RNA. DNA stands for *deoxyribonucleic acid*, illustrated in Figure 2.35. RNA stands for *ribonucleic acid*.

The monomers present in DNA and RNA are called *nucleotides*, an example of which is illustrated in Figure 2.36. A nucleotide consists of three parts: a phosphate group, a five-carbon sugar, and a nitrogenous base (or *nucleobase*). In the case of DNA, the sugar is *deoxyribose*, accounting for part of DNA's long name. The *nucleic* part of the name refers to the fact that DNA is located in the nucleus of a cell. The nitrogenous base is one of four molecules: *guanine*, *cytosine*, *adenine*, or *thymine*, often referred to by the letters G, C, A, and T. Their structures are shown in Figure 2.37, along with other details pertaining to the bonding arrangements in DNA.

The sequencing of these four "letters" is a code storing the information needed to run an organism, including the blueprints for the amino-acid sequences of the huge number of proteins in the body. DNA molecules can be hundreds of millions of nucleotides in length. This stunning structure is one of the most amazing discoveries of contemporary science.

The power of DNA lies in the complementarity of its two strands, paired together by a very specific hydrogen bonding pattern. Wherever there is a guanine base on one strand, it pairs (via three hydrogen bonds) to a cytosine base on the opposite strand, as illustrated in Figure 2.37. Likewise, adenine pairs with thymine (two hydrogen bonds). Figure 2.37 shows how the structures of the four bases are perfectly suited to hydrogen bonding. This complementarity gives DNA all kinds of advantages, including redundancy, so that even when repeatedly copied the genetic information maintains stability over long periods of time. The two strands in the DNA can also briefly separate so that the code can be copied and its information sent to other parts of the cell.

FIGURE 2.35. *Three-dimensional structure of DNA. Structured rather like a spiral staircase, the "backbones" or "handrails" on the outside of the double helix contain phosphate groups and sugar units (deoxyribose). The "stairs" are nucleobases, stacked in pairs, one pair above the other.*

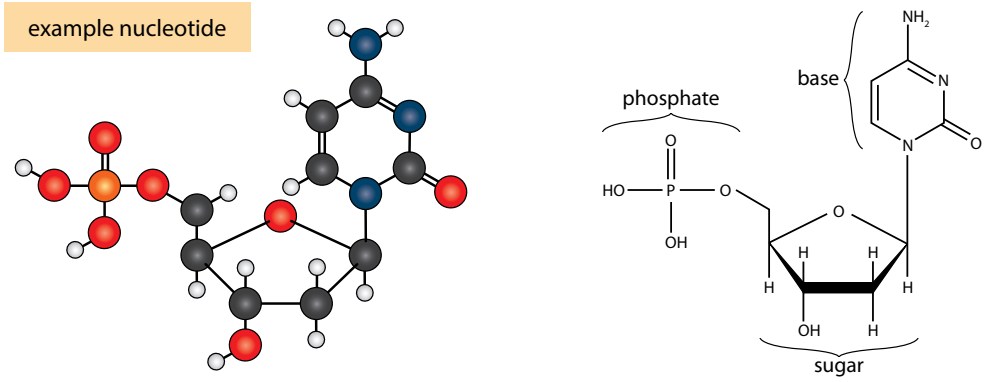
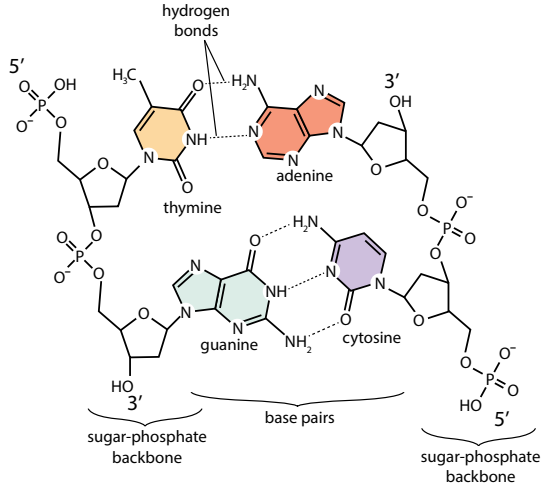


FIGURE 2.36. *An example of a nucleotide. The base shown here is cytosine, but can be any of four bases (cytosine, guanine, adenine, and thymine). The sugar shown is deoxyribose, present only in DNA. In the case of RNA, the sugar is ribose, with an additional OH group present. The phosphate structure remains the same regardless.*

FIGURE 2.37. *DNA bonding details. The figure shows the four DNA nucleobases, the sugar-phosphate backbones, A–T and G–C pairing, and the hydrogen bonding between strands. Note that the smallest human chromosome is about 48 million base pairs long!*



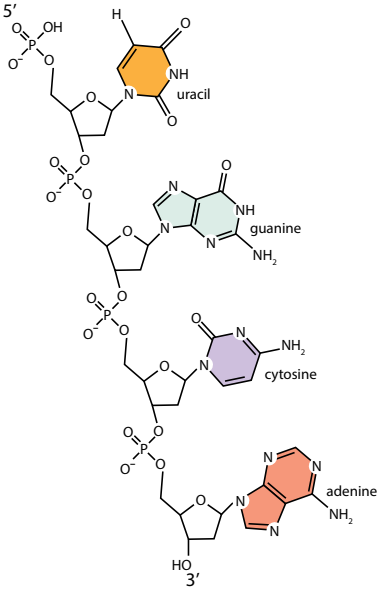


FIGURE 2.38. *RNA bonding details.*

RNA is similar to DNA but differs in three key ways. First, the sugar in RNA is ribose, so the full name of RNA is *ribonucleic acid*. Ribose is also a 5-carbon sugar, having an extra hydroxyl group that deoxyribose lacks, hence *de–* (lacking) *oxy–* (oxygen). The other two differences are illustrated by comparing the short RNA strand depicted in Figure 2.38 with the short DNA strand illustrated in Figure 2.37. Like DNA, RNA is assembled from four bases. Three of these are identical to those in DNA (adenine, guanine, and cytosine) but one of them is different—thymine in DNA is replaced by *uracil* in RNA. Uracil and thymine connect with adenine in the same way through hydrogen bonding. Finally, while DNA exists in a stable double strand, RNA is typically single-stranded and can be folded into various 3-D shapes. We explore the roles of DNA and RNA further in Chapters 5 and 7.

Biochemical research in recent decades indicates that in addition to hydrogen bonding between base pairs in DNA, there also exist in DNA "base-stacking interactions," that is, pushing and pulling forces between bases due to their being stacked on

In the Wonderworld of Biology

DNA as a Wire

An artist's rendition of the DNA molecule, with the backbones drawn as hollow tubes. Prominent are the flat DNA bases.

DNA is a wondrous molecule. In addition to a host of other properties, it has the curious ability to behave as an electric wire, conducting electricity. As mentioned in the discussion of nucleic acids, the DNA bases (A, T, C, and G) are flat molecules that stack on top of one another or side-by-side, like crackers in a pack or books on a shelf. The close stacking of the DNA bases (3.4 nm apart) means that the orbitals of their atoms overlap, forming one continuous, giant wire-like orbital where electrons reside. This arrangement means that electrons can freely flow through these orbitals, just as the conduction electrons do in metal wires. This property of DNA was discovered by Caltech chemist Jacqueline Barton and her colleagues in the 1990s. For many years it was an interesting phenomenon observed in the laboratory only. However, the question remained, is there any biological role for electron transfer through DNA?

More recent studies have shown that there indeed is a biological role for DNA's current-carrying properties. In fact, it may play a central role in the regulation of many DNA-related processes in the cell. One such process is DNA repair.

In the normal base-pairing arrangement, A pairs with T, and C pairs with G. However, sometimes the copying enzymes make mistakes, just as you might make a spelling mistake while typing. Additionally, sometimes free radicals can damage a DNA base, causing it to pair with the wrong partner. In both cases, DNA repair enzymes go back and fix the mistakes just as you fix your typos by proofreading. Now, if you are proofreading an essay, there are essentially two ways you can check for mistakes. The first way is to re-read your work very carefully, scanning for mistakes as you go. The second way is to run a spell-check or find-and-replace so the process proceeds at the speed of electrons running through the circuit board of your computer.

For years it was thought that DNA repair enzymes work like careful proofreading, slowly and methodically crawling along the DNA strand searching for damage or mismatches to repair. Recent work by Professor Barton and her colleagues has shown that electron transfer through the DNA stack plays a role, enabling the repair process to proceed much faster.

Multiple DNA repair enzymes bond to the DNA at distant sites, sending an electron from one to the other, essentially playing a game of catch. If the catch is successful, it means that the intervening DNA is healthy and correctly placed. However, if the catch is unsuccessful, then DNA damage has interrupted the electron transfer through the DNA base stack, just like a broken circuit would cause the lights to go off in your house. At that point, the repair enzyme is located relatively close to the damage—and can quickly crawl toward the offending base and fix it.

This exciting model illustrates how the Cycle of Scientific Enterprise works. Sometimes interesting discoveries are made in the lab whose far-reaching implications aren't apparent for many years. Decades of repeated testing and well-designed experiments can lead to modification of scientific models, as was the case with the model describing how DNA repair enzymes find damage to repair. Of course, this model also illustrates just how spectacular the engineering is in living things! The heavens—and our bodies—declare the glory of God. We are indeed fearfully and wonderfully made!

top of each other in the spiraling molecule. Looking once again at Figure 2.35, the nitrogen atoms in the bases are shown in blue, in the center of the molecule. The shape of the bases is flat, and in the view captured in the model, we are viewing the bases on edge, rather than the "front view" shown in Figure 2.37. The bases lie quite flat on top of each other—they stack, like the crackers in a pack of crackers. Research now shows that the base-stacking interactions may even be more significant in holding the DNA strands together than the hydrogen bonding between base pairs.

Chapter 2 Exercises

SECTION 2.1

1. What is hydrogen bonding? What kinds of atoms participate in hydrogen bonding and under what conditions?
2. How do van der Waals interactions occur? What kinds of molecules participate in these kinds of interactions?
3. What is cohesion? Adhesion? Using these two terms, briefly describe how water flows upward in trees and other plants against the force of gravity.
4. Explain why water striders can walk on water.
5. How does adhesion affect the way one takes a volume reading with a graduated cylinder or a buret?
6. Define specific heat capacity. What are two ways that water's specific heat capacity benefits life (at the organism and ecosystem levels)?
7. Consider a planet having high levels of H_2S (hydrogen sulfide) instead of H_2O (water). Do you think life could exist there? Why or why not? Explain your answer in terms of hydrogen bonding and resulting properties. Hint: the electronegativities of hydrogen, oxygen, and sulfur are 2.20, 3.44, and 2.58, respectively.
8. Describe how the unique density properties of water support life.
9. Consider a nonpolar substance such as oil. Is water able to dissolve it? Explain, in terms of electrostatic interactions, why this is the case.
10. Describe the process by which water dissolves a salt crystal at the molecular level.

SECTION 2.2

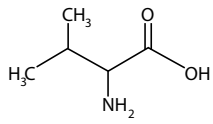
11. What are the six most abundant elements found in living organisms? What additional element is a significant part of the composition of the human body and why?
12. Do some research on the abundance of elements in the composition of the earth. How does this compare to the percentage of elements in living things?

SECTION 2.3

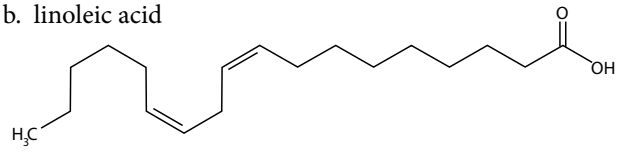
13. Why is the element carbon a major component of biological molecules?
14. Draw the following functional groups: hydroxyl, amino, carboxyl, phosphate. Describe their unique properties.

15. Sketch the following molecules on your own paper. Then circle and label as many different functional groups as you can.

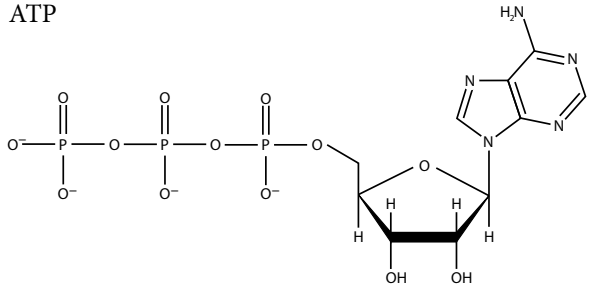
a. valine



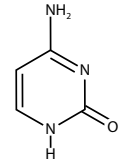
b. linoleic acid



c. ATP



d. cytosine



16. Describe the chemical structure of a simple sugar.
17. Describe the four major types of carbohydrates. What is the nature of each one's chemical structure? What are the roles of these carbohydrates in biology?
18. Compare and contrast carbohydrates and lipids.
19. Describe the three major classes of lipids in terms of their chemical structures and roles in the cell.
20. Describe the chemical structure of an amino acid. Identify and label the functional groups.
21. Structurally speaking, how are amino acids joined together to create proteins?
22. List and describe the four main groups of amino acids. Give an example of an amino acid from each group.
23. Explain how the different amino acids give a protein its specific structure.
24. Describe six major roles for proteins in the cell.
25. Make a table defining the four levels of protein structure: primary, secondary, tertiary, and quaternary.
26. Explain what happens to a protein if it is placed in an environment with an excessively high temperature.
27. Describe the basic features of DNA's structure.
28. Name the four bases found in DNA and indicate how they base-pair with one another.
29. What type of bonding or interaction do DNA bases use to achieve complementary base pairing? Why do you think the structures of these bases are suited for this type of bonding?
30. Compare and contrast the structures of DNA and RNA.
31. What are base-stacking interactions?

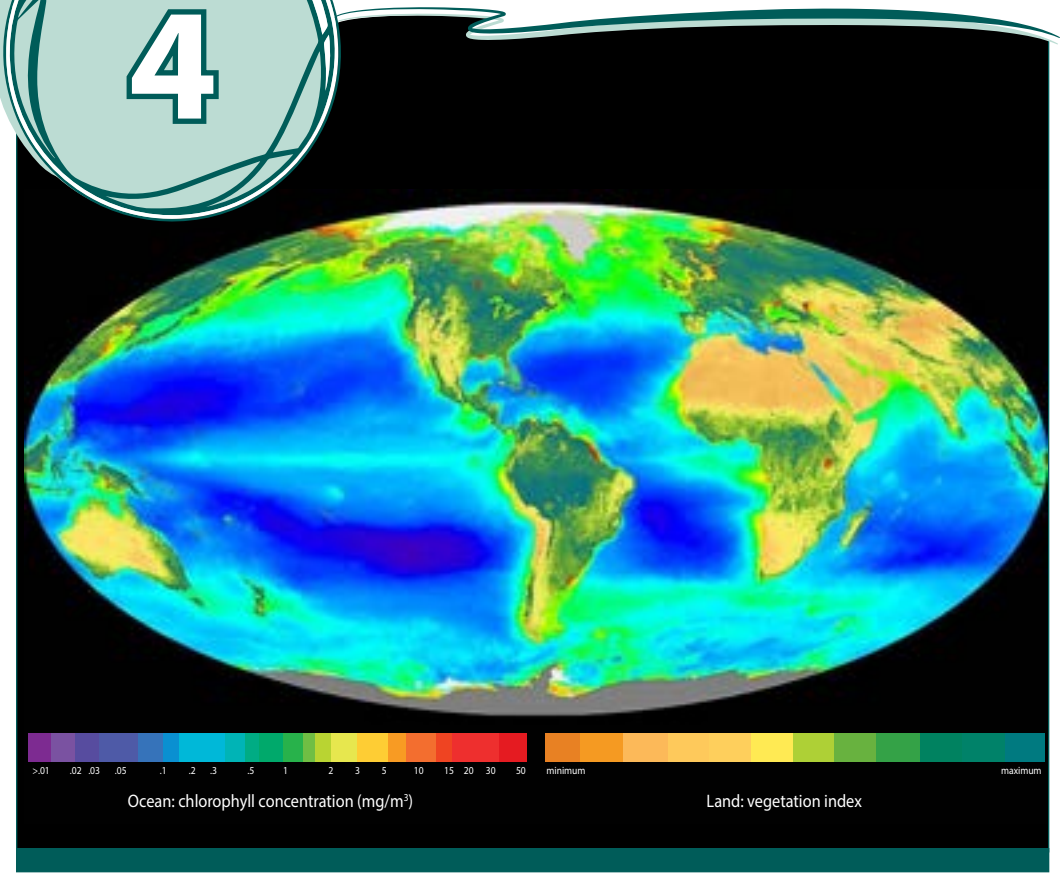
REVIEW QUESTIONS

32. List the six characteristics of all life.
33. Describe three types of microscopes. What types of structures can be visualized with a light microscope?
34. Compare and contrast light and electrons as used for visualizing biological specimens.
35. Define the terms resolution, magnification, and contrast.
36. Are light microscopy or electron microscopy appropriate tools for studying biomolecules? Why or why not?

CHAPTER

4

Energy in the Cell



All life on earth depends on the photosynthesis of plants, algae, and cyanobacteria. This image is color coded to indicate where photosynthesis is occurring on earth. In the ocean, oranges and reds indicate high levels of chlorophyll, the key molecule for photosynthesis; blues and purples indicate lower concentrations. On land, teal green shows the highest density of plant life; yellows and browns show lower densities. The Amazon Rainforest stands out as the photosynthetic champion on land, spilling out into an oceanic hotspot of photosynthesis where the Amazon River meets the Atlantic Ocean. On the flip side, the Australian Outback, Sahara Desert, and much of the Middle East appear barren. What other patterns do you observe in this picture? Can you formulate a generalization as to the most favorable places on earth for photosynthesis?

OBJECTIVES

After studying this chapter and completing the exercises, you should be able to do each of the following tasks, using supporting terms and principles as necessary.

SECTION 4.1

1. Describe seven forms of energy.
2. Given a chemical equation, classify it as combustion, redox, or hydrolysis.
3. Explain how the chemical structure of ATP makes it ideally suited for the role of energy currency in the cell.
4. Using a reaction pathway diagram, explain the difference between exergonic and endergonic reactions.
5. Define activation energy and relate it to the function of enzymes.
6. Explain how feedback inhibition helps control metabolism in the cell.

SECTION 4.2

7. Describe the overall goal of cellular respiration.
8. Briefly summarize the general matter and energy transformations in each stage of cellular respiration: glycolysis, the oxidation of pyruvate, the Krebs cycle, and oxidative phosphorylation, including where each process occurs.
9. List and describe the roles of four major coenzymes involved in cellular respiration (coenzyme A, NADH, $FADH_2$, and coenzyme Q).
10. Draw a skeleton diagram of glycolysis (not including chemical structures), naming the inputs, outputs, and their destinations.
11. Describe the breakdown of glucose into pyruvate that occurs during glycolysis.
12. Draw a skeleton diagram of the Krebs cycle (not including chemical structures), naming the inputs, outputs, and their destinations.
13. Explain how active transport functions in the electron transport chain.
14. Describe the mechanism of action of the ATP synthase.
15. Differentiate between aerobic respiration, anaerobic respiration, and fermentation.
16. Explain how other biomolecules are broken down to produce energy for the cell.

SECTION 4.3

17. Explain why chlorophyll is green.
18. Explain how light makes electrons available for the electron transport chain in photosynthesis.
19. Describe the process by which ATP and NADPH are synthesized in the light-dependent pathway.
20. Trace the path of an electron from water through the electron transport chain.
21. Explain how ATP and NADPH are used in the Calvin cycle.
22. Describe the major inputs and outputs of the Calvin cycle.
23. Compare and contrast the Krebs cycle and the Calvin cycle.
24. Compare and contrast the electron transport chains of cellular respiration and photosynthesis.
25. Describe two photosynthetic strategies some plants use to conserve water in dry, hot climates.

4.1 Energy in Chemical Reactions

4.1.1 *Energy*

Energy is difficult to define, but it is fundamental to the workings of the universe. Dictionaries usually relate energy to "the ability to do mechanical work," which does not really get at what energy *is*. Energy is one of the three fundamental components of the universe, the other two being matter and intelligence (seen in the mathematical structure of the laws of nature).

We can describe energy as the thing that holds everything together and that enables most processes to occur. Without energy, you could not move or think. Even non-living objects—such as a rock sitting on a mountaintop—contain energy in the motions of the molecules of which they are composed, the energy states of the electrons in the atoms, the chemical potential energy in the bonds holding the molecules together, and the gravitational potential energy due to being in the gravitational field of the earth.

So far as we know, all the energy that exists was created in the first moment of the existence of the universe. Since that time, all this created energy has obeyed the *law of conservation of energy*, which states that *energy can be neither created nor destroyed, only changed in form*.

Although energy is difficult to define, it is not difficult to understand the different forms in which energy exists, some of which are described in Table 4.1. For example, a boulder sitting at the edge of a tall cliff contains gravitational potential energy due to the earth's gravitational field. If the boulder falls off the cliff, this energy is converted to kinetic energy as the rock rushes toward the ground. Energy conversions—particularly those involving electromagnetic radiation and chemical potential energy—are among the most basic processes supporting the life and growth of living things.

TABLE 4.1. *Major forms of energy.*

Energy Type	Definition
kinetic energy	The energy of bodies in motion.
gravitational potential energy	Energy contained in the gravitational attraction between two objects.
chemical potential energy	Potential energy that is stored in the chemical bonds of molecules.
electrical potential energy	Potential energy that is stored in the separation of electric charges.
internal energy	The sum of the kinetic energies from the motions of individual molecules in a substance. The average kinetic energy of the molecules in a substance is proportional to its temperature.
nuclear energy	Energy from nuclear reactions, in which nuclei of atoms are rearranged and mass is converted to energy. Ultimately, almost all life on earth is powered by nuclear energy released from the sun, where hydrogen nuclei fuse together to form helium.
electromagnetic radiation	Massless energy that exists in a spectrum of wavelengths. Energy from the sun is transferred to earth as electromagnetic radiation.

4.1.2 *Chemical Reactions*

One of the characteristics of a living thing is that it cycles matter and achieves energy flow. This is achieved through thousands of chemical reactions, collectively known as *metabolism*. In this section, we highlight a few important terms that apply to chemical reactions in biological processes.

In a combustion reaction, a fuel such as methane combines with oxygen gas to yield carbon dioxide and water:

$$CH_4 + 2O_2 \rightarrow CO_2 + 2H_2O$$

Many biological reactions are of this type, such as the reaction that provides the energy for ATP synthesis, discussed in the next section.

A second important reaction type is the *oxidation-reduction* or *redox reaction*. In redox reactions, electrons are transferred from one reactant to another, forming new products in the process. Atoms that gain electrons are said to be reduced, while atoms that lose electrons are said to be oxidized. As you know, combustion reactions are a type of redox reaction, but for our purposes we are considering them separately.

A third biologically important reaction type is *dehydration synthesis*, in which a hydroxyl group is removed from one monomer and a hydrogen is removed from another monomer to form a water molecule; in the process, a covalent bond is formed joining the two monomers together. The opposite reaction is *hydrolysis*, a term that comes from the Greek *hydro–* (water) and *–lysis* (breaking apart). In hydrolysis reactions, water serves as a reactant that slices another molecule into two smaller product molecules (see Figure 2.17).

Besides products and reactants, other molecules, called *catalysts* or *enzymes* facilitate reactions without being transformed themselves. An example of a catalyst is the material inside the catalytic converter in automobile exhaust systems. Car exhaust contains pollutants such as carbon monoxide (CO) and nitrogen oxides that result from imperfect combustion. The catalyst causes the nitrogen oxides to react to become nitrogen and water, while the CO is converted to CO_2. In the chemical equation describing a catalyzed reaction, the catalyst is typically indicated over the yields arrow.

In a chemical reaction, there is always a net gain or loss of chemical potential energy between the reactants and the products. Of the forms of energy listed in Table 4.1, the most important form of energy for biology is chemical potential energy, energy stored in the chemical bonds of molecules. Each chemical bond stores a different amount of energy. Chemical reactions play a key role in releasing or storing this chemical potential energy.

4.1.3 *ATP as Energy Currency*

Figure 4.1 illustrates a hydrolysis chemical reaction involving the nucleotide molecule *adenosine triphosphate* (*ATP*).

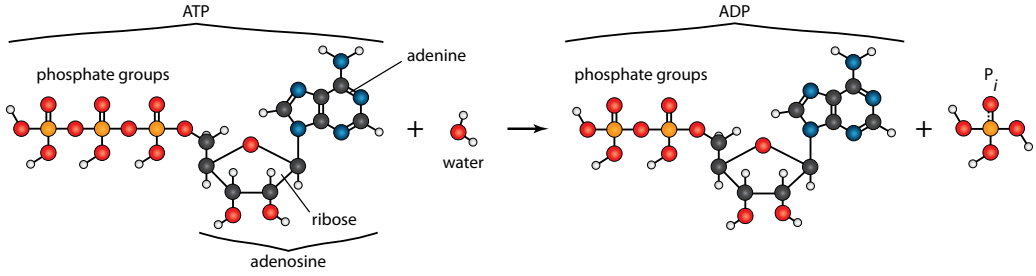
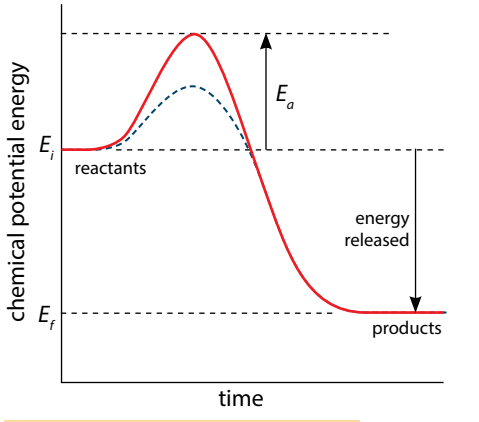
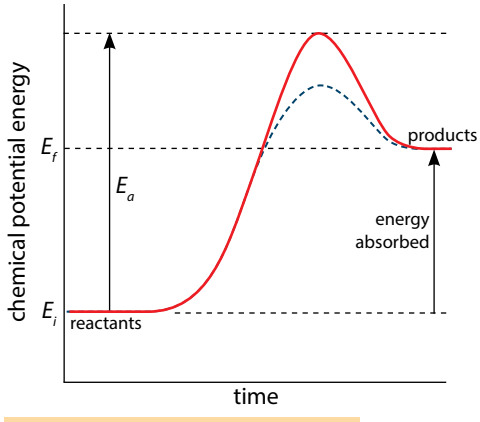


FIGURE 4.1. *The hydrolysis reaction of ATP to ADP (adenosine diphosphate) and a phosphate group. In the models, black = carbon, white = hydrogen, blue = nitrogen, red = oxygen, and orange = phosphorus.*



exergonic reaction pathway



endergonic reaction pathway

FIGURE 4.2. *Reaction pathway graphs, showing the energy of a chemical reaction as it proceeds. The horizontal axis represents time, as the reaction progresses from start to finish. The vertical axis shows chemical potential energy stored in the bonds of the participating molecules.*

ATP is composed of the nitrogenous base *adenine* (the part of the molecule including the nitrogen atoms, colored blue in the figure), the sugar *ribose* (the ring in the center), and three phosphate groups (each containing a phosphorous atom, orange in the figure). Phosphate groups have a phosphorus atom attached to four oxygen atoms and one or more hydrogen atoms. When detached from the rest of the molecule, biologists call the phosphate group *inorganic phosphate*, denoted as P_i. Since ATP has three phosphate groups, it is called a *triphosphate*. The third phosphate group is attached to the rest of the molecule by a high-energy bond that holds a large amount of chemical potential energy. Chemical potential energy is analogous to the energy in a boulder sitting on a tall cliff. The boulder contains high gravitational potential energy that can be converted into a large amount of kinetic energy by falling off the cliff. In an analogous fashion, when the third phosphate in the ATP molecule is removed, a lot of energy is released.

For this reason, the ATP molecule serves as the universal energy currency of the cell. Whenever the outermost phosphate group is lost or transferred, energy is released to drive other reactions. In this fashion, ATP hydrolysis supplies the energy for most other processes in the cell. The reactants in Figure 4.1 are ATP and water. The products are *adenosine diphosphate* (*ADP*) and P_i.

Exergonic (i.e., exothermic) reactions release energy into the surroundings or make that energy available for other processes. Endergonic (i.e., endothermic) reactions absorb energy, storing chemical potential energy in the bonds of the reaction products. The energy driving an endergonic reaction must come from an outside source in order for the reaction to proceed. Indeed, the chemical reaction in Figure 4.1 can be reversed, reattaching the phosphate to ADP with a net input of energy. This important and complex process is achieved by cellular respiration, our topic in the next section.

The nature of endergonic and exergonic reactions is illustrated by the graphs shown in Figure 4.2. The initial climb in

energy is called *activation energy*, E_a. It represents the amount of energy input required to get the chemical reaction started. The activation energy usually destabilizes the chemical bonds of the reactants enough that they can break and re-form in a new configuration. When a person lights a fire with a match, the heat from the match provides the activation energy to begin the combustion of the fuel. (The activation energy for lighting the match is the heat produced by friction during the striking of the match.)

ATP is required for endergonic reactions because ΔG, the change in the Gibbs free energy, is positive and typically too large to come from the thermal energy of the surroundings (e.g., body heat). ATP is not required for exergonic reactions because ΔG is negative and the E_a to break existing covalent bonds is supplied by thermal energy of the surroundings.

Now, let us look at the energy levels of the chemical bonds both before and after the reaction takes place. In the exergonic reaction pathway, we see that the chemical potential energy in the reaction products is lower than the chemical potential energy in the reactants. Thus, energy is released into the environment as the reactants are transformed into products. In the endergonic reaction pathway, the final energy of the products is higher than that of the reactants, indicating a net gain in the energy stored in the chemical bonds of those molecules.

4.1.4 *Enzymes*

Referring again to Figure 4.2, note that two curves are shown for each pathway—a red curve and a dotted blue curve. This is where catalysts come into play. Without being used up itself, a catalyst serves to lower the activation energy of a reaction, enabling it to proceed much more readily. The dotted blue curves indicate the lower activation energy required when a catalyst is present.

Biological catalysts are called *enzymes* and are typically proteins with a very specific structure. Enzymes interact with one or several reactant molecules, perhaps stretching them out or orienting them in the right position for a reaction to occur.

Figure 4.3 shows the *active site* of one such enzyme. The active site, colored blue in the figure, is the specific spot in the protein that holds onto the reactants, usually through hydrogen bonding or other intermolecular interactions. Reactants in enzymatic reactions are called *substrates*. The amino acids in an enzyme are in *just the right 3-D structure* to enable the protein to adjust its shape to grasp perfectly its substrate, a phenomenon known as *induced fit*. This figure depicts the enzyme hexokinase, as it binds to two substrates—ATP and the sugar xylose. Notice how the enzyme encloses around the substrates for a perfect fit.

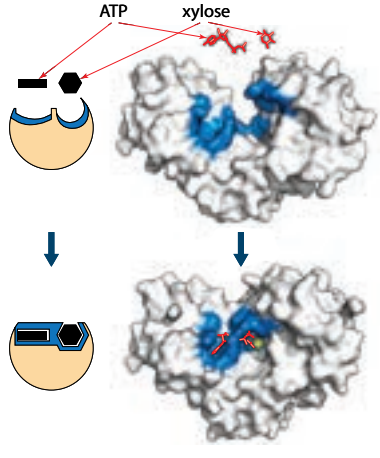


FIGURE 4.3. *This figure depicts the enzyme hexokinase, as it binds to two substrates—ATP and the sugar xylose. Hexokinase facilitates the first step in glycolysis, with the sugar glucose. But xylose, with a similar shape to glucose, competes with glucose and acts as an inhibitor.*

Let's revisit the chemical reaction of Figure 4.1 and consider why, if it is so easy for ATP to lose a phosphate group by a simple reaction with water (which is almost everywhere in the

cell by virtue of the aqueous cytosol), the cell does not spontaneously explode with all the ATPs losing their phosphates all at once. The reason relates to the activation energy.

Here is an amazing thing about cells. Of all the thousands of chemical reactions at play, every single one of them requires a specific enzyme to proceed. Enzymes are designed to facilitate specific chemical reactions, with an active site whose amino acids are exactly aligned to grasp the reactant molecules and contort them, destabilizing the bonds and making them easier to break, which lowers the activation energy so that the chemical reaction can proceed. Without the enzyme, the reaction would have such a high activation energy barrier that it would take thousands of years for the reaction to proceed. That's how carefully God has designed the metabolism of the cells that keeps us alive!

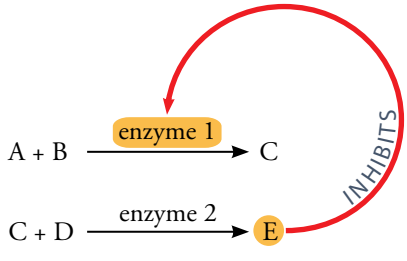
Enzymes are precisely controlled in a number of ways. First, their production can be increased or decreased in response to an external stimulus. Second, the activity of enzymes can be inhibited or enhanced by an immediate or downstream product of its chemical reaction. The process of inhibition is called *feedback inhibition* and is an important method of preventing chemical reactions from making too many product molecules.

Consider this hypothetical series of biochemical reactions: A and B are converted to C in a reaction catalyzed by enzyme 1. Next, C joins with D to produce E, in a reaction catalyzed by enzyme 2. The chemical equations for these reactions are:

$$A + B \xrightarrow{\text{enzyme 1}} C$$

$$C + D \xrightarrow{\text{enzyme 2}} E$$

A common pattern is that the E molecule then serves to inhibit enzyme 1:



With less of the active enzyme 1 around, fewer molecules of C and E are produced. Molecule E can do this by physically sitting in the active site of enzyme 1 (blocking its action) or binding to another site in the protein, making its conformation (folded shape) unfavorable for catalyzing the transformation of A and B into C.

To understand the phenomenon of feedback inhibition, consider this analogy. Suppose people are laying down sandbags in preparation for a possible flood, as shown in Figure 4.4. To

facilitate this, workers line up and pass sandbags from one to the next. Feedback inhibition is like the worker at the end of the line shouting to the first person in the line, "Slow down a minute! We have too many sandbags!" Interestingly, in other cases where extra products are needed, a mechanism similar to feedback inhibition serves to produce the opposite effect—a downstream molecule actually speeds up its own production.

FIGURE 4.4. *In metabolism, a series of chemical reactions can be thought of as similar to a group of people lined up to pass sandbags from one location to the other. Feedback inhibition occurs when a downstream product builds up too much, serving to inhibit an earlier step.*

4.2 Cellular Respiration

We are now ready to consider the complex and fascinating process by which ATP energy is generated in the cell—*cellular respiration*. This section contains a lot of information and terminology. Learning this topic cannot be rushed, so take the time to study this section carefully.

4.2.1 *Production of ATP*

Many of the reactions that take place during metabolism serve to break down macromolecules into usable building blocks. The general word for these processes is *catabolism*. Other reactions take the building blocks and build up macromolecules. The general term for these is *anabolism*. Catabolic processes release the energy required to drive the anabolic processes. Additionally, active transport, DNA replication, cell division, and protein synthesis all require large amounts of energy to proceed. In fact, without a continuous supply of energy in the form of ATP, cells cannot continue living. Generating this ATP is the overall goal of cellular respiration. We now consider how ATP is generated. The overall process is represented by the following simplified chemical equation:

$$\text{glucose} + O_2 + \text{ADP} + \text{phosphate} \rightarrow \text{ATP} + CO_2 + H_2O \quad (4.1)$$

The overall process represented by Equation (4.1) occurs in two major chemically separate phases. The second of these phases is the generation of ATP from ADP:

$$\text{ADP} + \text{phosphate} \rightarrow \text{ATP} + H_2O \quad (4.2)$$

Notice that this reaction is the reverse of the one presented in Figure 4.1. As such, the reaction is extremely endergonic—it requires a large energy input in order to proceed. The energy for the production of ATP is supplied by an exergonic chemical reaction:

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6H_2O + 6CO_2 + \text{energy} \quad (4.3)$$

We can think of Equation (4.3) as digested food + oxygen yields exhaled carbon dioxide, water, and energy (in the form of ATP). The two reactions in Equations (4.2) and (4.3) are part of a continuous cycle of energy production and energy consumption in the cells, illustrated in Figure 4.5.

Equation (4.3) is a combustion reaction. In fact, if you light a marshmallow—essentially pure sugar—it burns fantas-

tically. (Don't try this at home. Only when roasting marshmallows over a campfire!) The activation energy necessary to get the reaction going is the flame. But although burning sugar releases a lot of energy, a flame-producing chemical reaction obviously cannot occur in all 37 trillion cells of your body. Thus, cells manage Equation (4.3) through a highly controlled, enzyme-mediated series of many chemical reactions. Cellular respiration comprises all of them, many of which are redox reactions.

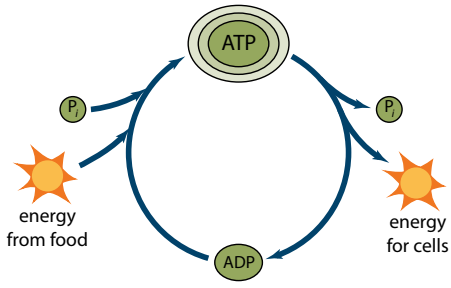


FIGURE 4.5. *The recycling of ATP. Using the energy from food, ADP combines with inorganic phosphate to make new ATP molecules. The ATP molecules then react to liberate energy for powering cells, producing ADP and inorganic phosphate in the process.*

Before cellular respiration can take place, your body must transport fuel and oxygen to each cell. The food you eat is broken down (digested) in the mouth, stomach, and small intestines by a number of hydrolytic enzymes. The resulting small molecules are absorbed through the small intestine into the bloodstream, which delivers them to the cells. Though many molecules can serve as fuel sources (lipids, proteins, and carbohydrates), we focus on glucose here for simplicity. Broken down products of the other types of molecules can all enter the cellular respiration process at one point or another. We consider these in more detail a bit later.

When you breathe in oxygen gas, it diffuses across your lungs into the red blood cells in your bloodstream, where the hemoglobin protein transports it to every cell. The small nature of the O_2 molecule allows its passage across membranes by simple diffusion, both in the lungs and at the receiving cells. Once the cells have glucose and O_2 molecules at hand, cellular respiration can commence.

4.2.2 *The Four Major Stages in Cellular Respiration*

The overall purpose of cellular respiration is to turn food and oxygen into usable energy, ATP. This process, illustrated in Figure 4.6, is tightly controlled and occurs in four identifiable stages:

1. *Glycolysis* Literally meaning "sugar breaking," glycolysis occurs in the cytosol. This stage breaks the six-carbon glucose molecule into two three-carbon pyruvic acid molecules, while releasing two ATP molecules per glucose molecule, and high-energy electrons, carried by the electron carrier molecule NADH, for use later.
2. *Oxidation* of *pyruvate* After moving into the mitochondrial matrix, the pyruvic acid (or pyruvate) is converted into

a two-carbon compound called acetyl coenzyme A (acetyl-CoA). This step also produces one molecule of carbon dioxide and transfers electrons to produce another molecule of NADH. A coenzyme is one of a class of molecules called cofactors—molecules that must be present in addition to a specific enzyme in order to catalyze a particular reaction.

3. *Krebs cycle* Taking place in the mitochondria, this cycle takes acetyl-CoA and transforms it into various other four-, five-, and six-carbon compounds, releasing several NADH, $FADH_2$, and ATP molecules in the process. Carbon dioxide is released as a byproduct. (The waste CO_2 is transported to the lungs and exhaled.) This process is also called the *citric acid cycle* or the *tricarboxylic acid* (*TCA*) *cycle*.

4. *Oxidative phosphorylation* This process takes place on the inner membrane of the mitochondria. Using the energy of electrons stored in carrier molecules such as NADH and $FADH_2$, the electrons are passed through an *electron transport chain* (ETC) that indirectly drives a molecular motor called the *ATP synthase*. This amazing enzyme "charges" used-up ADP molecules, converting them back into ATP that the cell can use. This process cannot proceed without oxygen, which serves as the final electron acceptor.

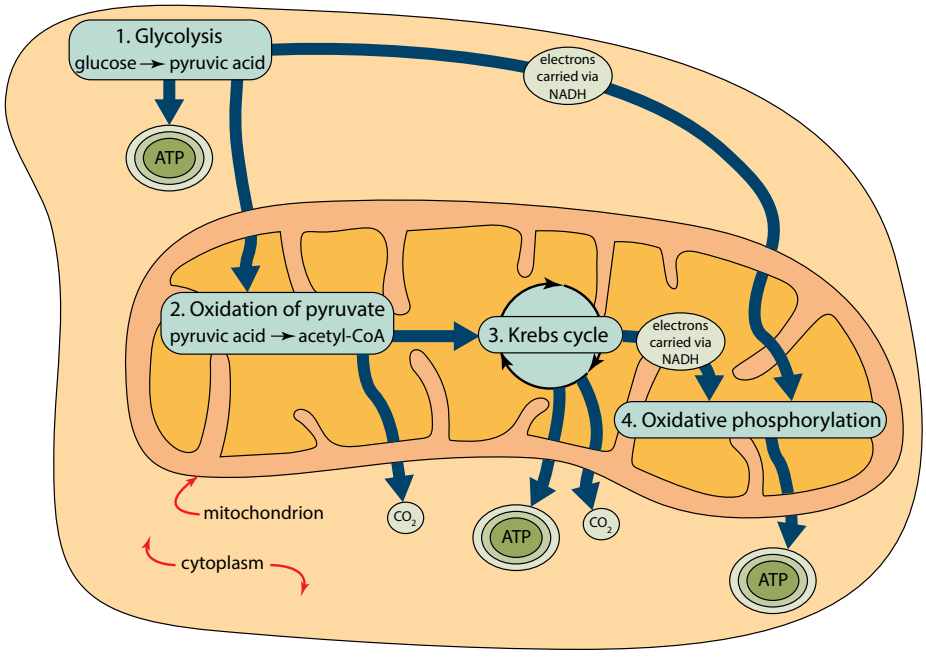


FIGURE 4.6. *The four major stages in cellular respiration.*

If reading those paragraphs makes your head spin, don't worry. We now walk through the process in detail, elaborating on each step. As you grapple with understanding this topic, think about each of these reactions occurring in every one of

your cells. The air you breathe and the food you eat are powering the amazingly complex mitochondrial machinery, yet God knows every molecule and every enzyme and what they are doing in each of your cells, every nanosecond of your life. This process is so essential that if a person's oxygen supply is cut off for just a few minutes, the person dies. As you study this complex topic, let it be an opportunity to worship our great God who knows you in far greater detail than this relatively simple overview can hope to communicate.

1 Glycolysis

Figure 4.7 shows the ten chemical reactions that take place during the process of glycolysis, starting with the six-carbon sugar glucose. Each chemical reaction is catalyzed by its own specific enzyme, indicated in blue. Glycolysis occurs in the cell cytoplasm and converts glucose into pyruvic acid, the compound that enters the mitochondrion.

The first five chemical reactions of glycolysis are collectively called the *energy investment phase*. Let's look at each of these steps more closely. In step one, an ATP molecule is hydrolyzed to ADP. The phosphate is transferred to the glucose molecule, resulting in the molecule glucose-6-phosphate, in which the numeral '6' indicates which carbon atom the phosphate is joined to. In step two, glucose-6-phosphate is rearranged into an *isomer*, fructose-6-phosphate. (Isomers are molecules that have the same chemical formula but different arrangements of the atoms in their molecular structure.) A second ATP molecule is hydrolyzed in step three, again transferring a phosphate, resulting in the molecule fructose-1,6-biphosphate. This is then broken into two three-carbon molecules which are isomers, dihydroxyacetone phosphate and glyceraldehyde-3-phosphate. Step 5 converts the dihydroxyacetone phosphate into glyceraldehyde-3-phosphate so that there are two identical molecules at the end of the energy investment phase. The intermediate product at the end of the energy investment phase is two three-carbon molecules of glyceraldehyde-3-phosphate (G3P).

The next five reactions are part of the *energy payoff phase* of glycolysis, during which the G3P intermediates are transformed into pyruvic acid. In the sixth reaction, each of the two G3P molecules is oxidized: two electrons and two hydrogens are removed from each G3P and used to reduce a carrier molecule known as NAD^+ to make NADH and H^+. This results in a net production of two NADH molecules. These carry the electrons, and their associated energy, to the inner membrane of the mitochondria where they are used in the electron transport chain. You may also notice that an inorganic phosphate is added to each of the G3P molecules in this step as well. The resulting products are two molecules of 1,3-bisphosphoglycerate. In step seven, one of the phosphate molecules is removed from each of the 1,3-bisphosphoglycerates and transferred to ADP to make ATP. These are the first two ATP molecules produced in the pro-

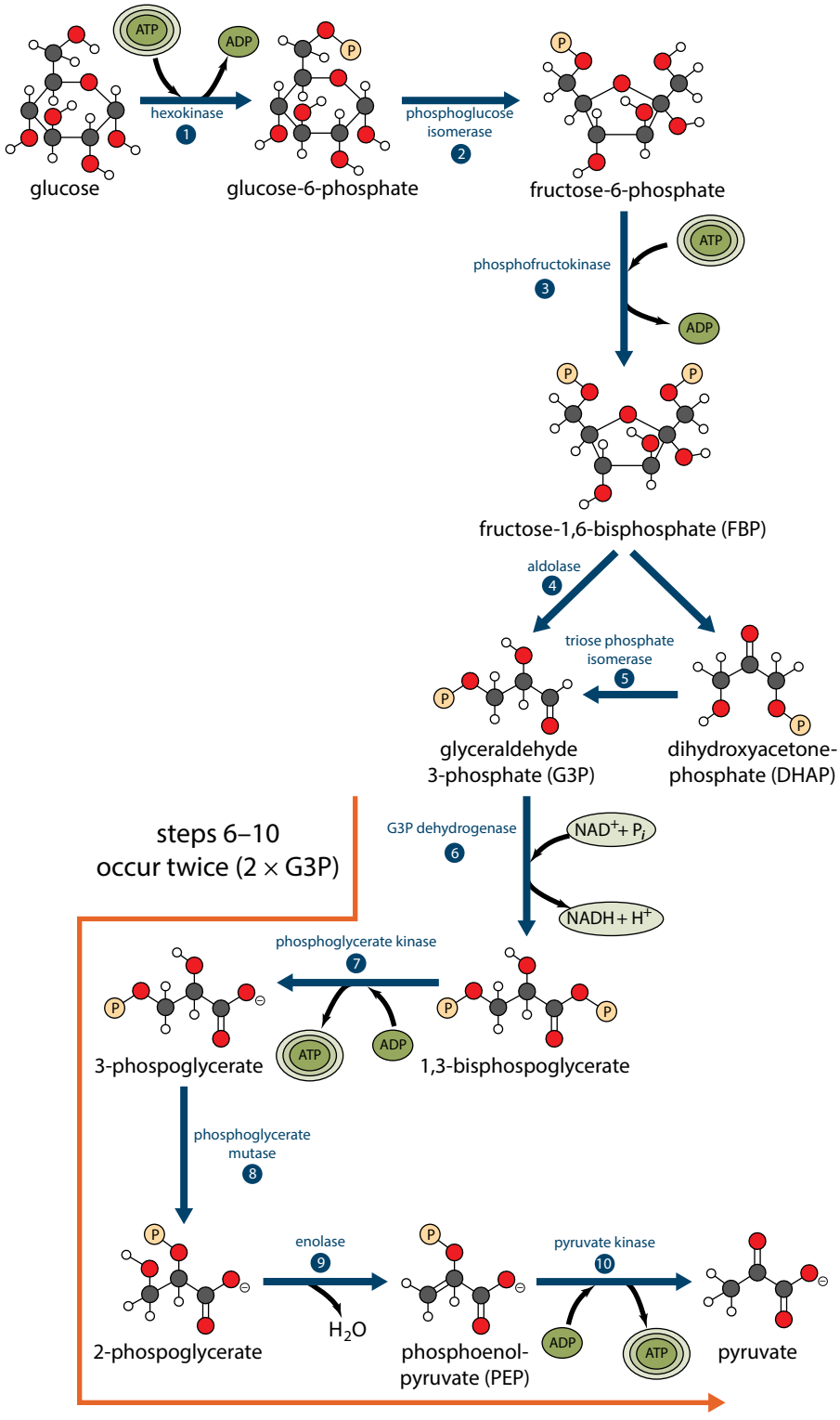


FIGURE 4.7. *The 10 steps of glycolysis.*

cess of glycolysis. The 3-phosphoglycerate generated in step seven is rearranged in step eight into 2-phosphoglycerate. In step nine, a water molecule is removed, and phosphoenolpyruvate is produced. In the final step, step ten, the remaining phosphate is removed and transferred to another ADP to produce two more ATP molecules. This results in the final product of glycolysis, pyruvic acid (or pyruvate). In summary, two ATP molecules are invested in the first phase of glycolysis and four are produced in the second phase, resulting in a net gain of two ATP molecules per glucose that are immediately available to serve the energy needs of the cell. In addition, each glucose also produces two NADH molecules and some water as a byproduct.

Note that glycolysis does not require oxygen and can thus occur in energy processes where there is no oxygen. One of these is fermentation, which we consider later.

The process of glycolysis may be summarized as follows:

1. Converts one glucose molecule into two pyruvic acid molecules.
2. Uses two ATP molecules to generate two G3P intermediates; from these generates four ATP molecules, a net production of two ATPs.
3. Generates two NADH molecules (electron transporters).
4. Occurs in the cytoplasm of cells.
5. Proceeds in the absence of oxygen.

2 Oxidation of Pyruvate

The two molecules of pyruvic acid from glycolysis are transported from the cytoplasm of the cell into the matrix of the mitochondria for the second step in cellular respiration. In this step, each molecule of pyruvic acid reacts with a cofactor molecule called coenzyme A (CoA), as illustrated in Figure 4.8. The products of this reaction are acetyl-CoA, NADH, and carbon dioxide. As mentioned above, a cofactor is a molecule that must be present in addition to a specific enzyme in order to catalyze a particular reaction. This type of molecule is called a coenzyme because, like an enzyme, it is not itself consumed in the chemical pathway and can therefore be reused. However, it is not a protein as enzymes are.

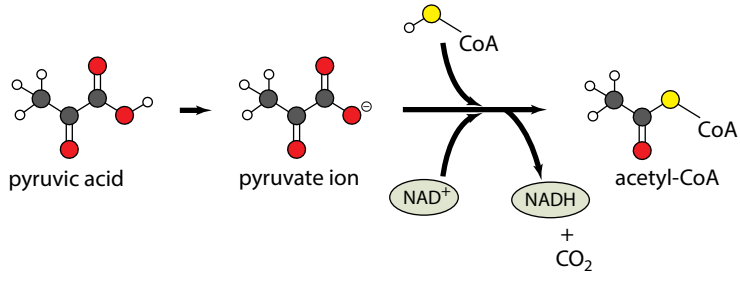


FIGURE 4.8. *The oxidation of pyruvic acid, resulting in acetyl-CoA and generating NADH and CO_2 in the process. Sulfur atoms are shown in yellow.*

In this reaction, the pyruvic acid molecules are first ionized to form pyruvate ions. Then the CoA (containing sulfur and hydrogen atoms) attaches to the pyruvate to form acetyl-CoA. Notice that pyruvic acid contains three carbons, and the acetyl group in acetyl-CoA only contains two carbons. The remaining carbon is released as CO_2. Because this process is a redox reaction, electrons are transferred to NAD^+ to form NADH.

The oxidation of pyruvate may be summarized as follows:

1. Inputs are two pyruvate molecules and two CoA molecules per glucose that enters glycolysis.
2. Outputs are two acetyl-CoA molecules, two NADH molecules, and two CO_2 molecules per glucose.
3. Occurs in the mitochondrial matrix.
4. The acetyl-CoA molecules enter the Krebs cycle.

3 Krebs Cycle

The Krebs cycle is a wonder of biochemistry and its discovery has an interesting history. Preliminary components of the pathway were discovered by Hungarian biochemist Albert Szent-Györgyi (Figure 4.9), for which he received the Nobel Prize in 1937. The entire pathway was elucidated by German biochemist Hans Adolf Krebs (Figure 4.10) in 1937. He won the 1953 Nobel Prize for this important discovery.

FIGURE 4.9. *Hungarian biochemist Albert Szent-Györgyi (1893–1986).*

Interestingly, both men were affected by their status as Jews in Germany. Earlier in his career, Krebs earned repute for his groundbreaking work in the discovery of other metabolic pathways. Despite his accomplishments, he was forced to flee Germany in 1933 because of his Jewish heritage. Recognizing his talent, the University of Cambridge in England quickly recruited him, enabling his research to continue.

During WWII, Szent-Györgyi joined the Hungarian resistance movement, helping his Jewish friends escape from the Axis-aligned country. In 1944, he went to Cairo under the guise of a scientific lecture, but he was actually there to collaborate with the Allies. As a result, Adolf Hitler himself issued a warrant for Szent-Györgyi's arrest. He became a fugitive, eventually emigrating to the United States.

Let's turn now to describing the Krebs cycle. The feature that distinguishes a cyclic reaction pathway from a linear one is that one of the reaction product molecules at the end of the cycle is also one of the initial substrate molecules at the beginning of the cycle. Cycles can either produce energy, as the Krebs cycle does, or consume energy, as the Calvin cycle does (Section 4.3.3). The Krebs cycle is repeated twice for each molecule of glucose that undergoes glycolysis (once for each acetyl-CoA molecule that enters).

FIGURE 4.10. *German biochemist Hans Adolf Krebs (1900–1981).*

Studying Figure 4.11, you see that there are eight different chemical reactions, each regulated by a specific enzyme. Let's walk through each of these reactions.

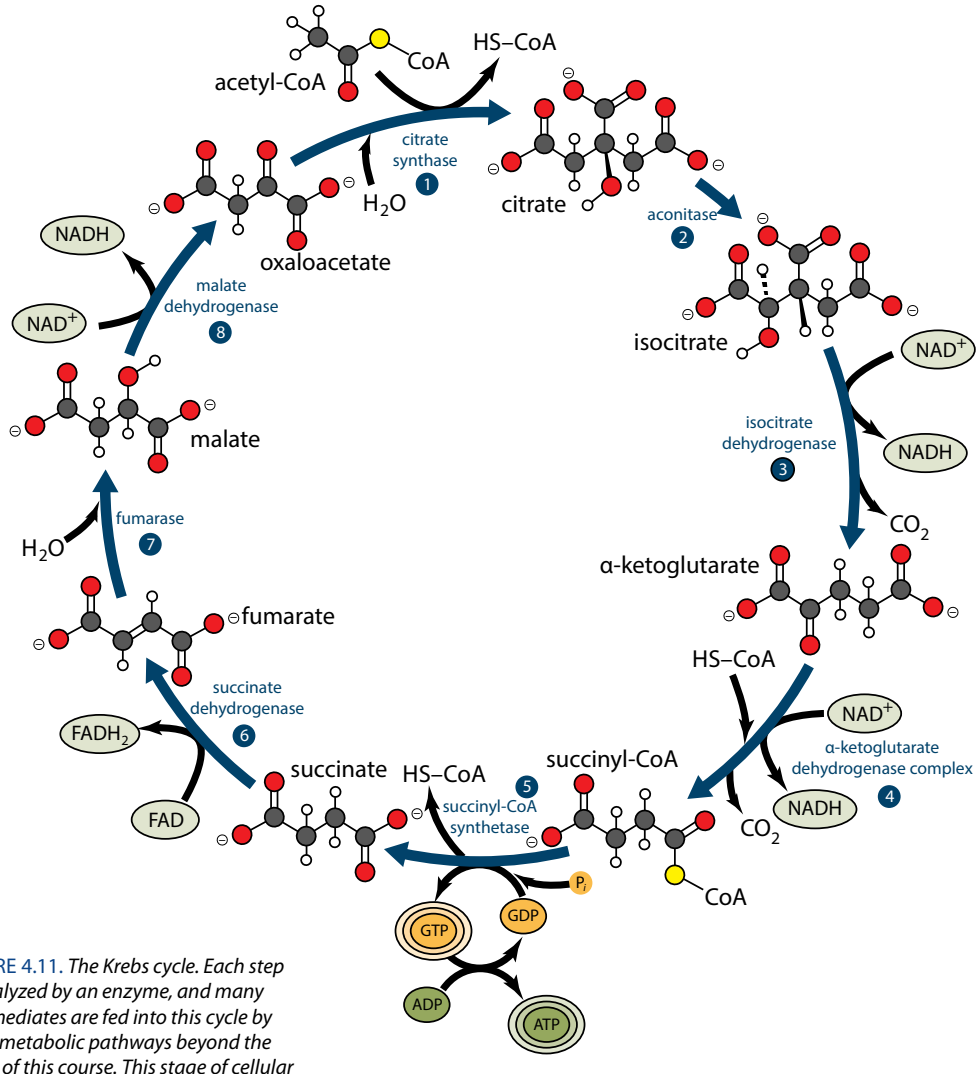


FIGURE 4.11. *The Krebs cycle. Each step is catalyzed by an enzyme, and many intermediates are fed into this cycle by other metabolic pathways beyond the scope of this course. This stage of cellular respiration is cyclical, repeating twice before a particular glucose molecule is completely broken down into carbon dioxide.*

Each turn through the cycle begins with the reception of an acetyl-CoA molecule from the previous pyruvate oxidation step. The acetyl group from the acetyl-CoA joins to a four-carbon molecule called oxaloacetate to form the six-carbon molecule citrate, or citric acid. This is where the cycle gets its name, "citric acid cycle." The CoA then leaves the cycle and becomes available to oxidize another molecule of pyruvic acid. The second reaction is a rearrangement of citrate into its isomer isocitrate. In the third reaction, the isocitrate is oxidized, removing two electrons and hydrogens, transferring them to an NAD^+ molecule to produce NADH. There is also the removal of a carbon atom in the form of CO_2. This produces the molecule alpha-ketoglutarate (α-ketoglutarate). The fourth reaction is another oxidation: more electrons are stripped and transferred to NAD^+, producing another molecule of NADH and another CO_2. The result is the intermediate molecule succinyl-CoA. In the fifth reaction, energy is used to produce a molecule of guanosine triphosphate (GTP) (later converted into ATP),

resulting in a molecule of succinate. Next, succinate is oxidized, losing electrons and hydrogens. However, this time they are transferred to a different electron carrier molecule, FAD, which is reduced to make $FADH_2$. The $FADH_2$ is a slightly less-energetic cousin of NADH. The resulting intermediate is fumarate, which is rearranged in the seventh reaction into the molecule malate. In the final (eighth) reaction, malate is oxidized, losing more electrons, which are transferred to NAD^+, which is reduced, to produce one more molecule of NADH. This transforms malate back into oxaloacetate. This molecule is then ready to receive another molecule of acetyl-CoA to start the cycle over.

In summary, each acetyl-CoA molecule that enters the Krebs cycle produces two CO_2 molecules, three NADH molecules, one $FADH_2$ molecule, and one molecule of ATP. The CO_2 molecules return to the blood to be exhaled or to regulate pH in the bicarbonate buffer system in the blood. The NADH and $FADH_2$ molecules transport electrons to the electron transport chain for use in the next phase of cellular respiration.

Because there are two molecules of acetyl-CoA produced in glycolysis for every glucose, the Krebs cycle is completed twice for each glucose molecule that enters glycolysis. This means that the final products for a glucose molecule are double those listed above. The Krebs cycle may be summarized as follows:

1. Occurs twice per glucose that enters glycolysis.
2. The input molecules are two acetyl-CoA molecules per glucose.
3. The outputs are two ATPs, six NADH, two $FADH_2$, and four CO_2 per glucose.
4. Occurs in the mitochondrial matrix.
5. The NADH and $FADH_2$ deliver electrons to the electron transport chain and the CO_2 is exhaled from the body.

4 Oxidative Phosphorylation

So far, we have walked through glycolysis, the oxidation of pyruvate, and the Krebs cycle in detail. For each glucose molecule, we have broken down the six-carbon glucose molecule into six molecules of carbon dioxide, generated a net of four ATP molecules, and stored high-energy electrons in ten NADH carrier molecules and two $FADH_2$ carrier molecules (two electrons per carrier molecule). In the final stage of cellular respiration, there is a large payoff in terms of additional ATP molecules.

The purpose of oxidative phosphorylation is to use the energy contained in NADH and $FADH_2$ to overcome the high energy cost involved in the chemical reaction that produces ATP, Equation (4.2):

$$\text{ADP} + \text{phosphate} \rightarrow \text{ATP} + H_2O$$

The result is ATP that the cell uses for every process.

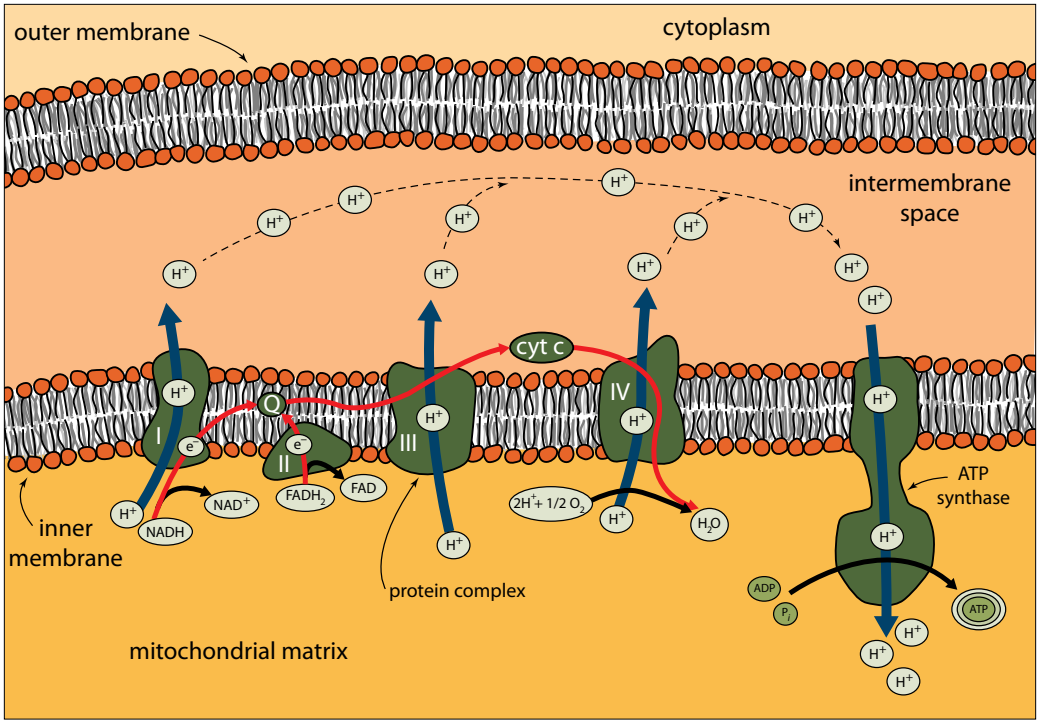


FIGURE 4.12. *Oxidative phosphorylation. Multi-enzyme complexes I, II, III, and IV comprise the electron transport chain. Using the electron carriers (NADH and $FADH_2$) from previous steps, these membrane proteins pass electrons from one protein to the next, releasing a controlled amount of energy at each step that drives the active transport of H^+ ions across the membrane. The final electron acceptor is O_2, producing water as the final product. The rightmost enzyme is the ATP synthase, powered by the passive transport of H^+ back into the mitochondrial matrix. The energy generated by the H^+ "waterfall" spins the ATP synthase, generating ATP.*

The process of oxidative phosphorylation is illustrated in Figure 4.12. The process involves four separate *protein complexes*, identified as I–IV, each comprising proteins, coenzymes, and cofactors. These types of molecules are able to carry electrons and pass them on to the next molecule in the chain. NADH and $FADH_2$ donate their electrons to the first two complexes in the chain, respectively; these electrons are then transferred to the next protein, and the next, in stepwise fashion. With each pass of an electron, a little bit of energy is released.

To understand this concept better, consider an analogy. Imagine you have a raw egg you wish to transport safely from a balcony to the ground, without leaving the balcony yourself. You have plenty of helpers, but no other materials or equipment. You only have two options: (1) drop the egg (which would certainly result in a yolky mess on the ground), or (2) have your helpers line up on the stairs, and then carefully pass the egg from one person to the next until it can be gently set down. Each time the egg is passed to a person standing lower on the stairs, a small amount of gravitational potential energy is released into some other form of energy. The latter scenario is analogous to what happens in the electron transport chain (ETC). If NADH and $FADH_2$ were to release their electrons to O_2 all at once, the result would be an uncontrolled explosion of energy that would be disastrous for the cell, just as dropping an egg from a high balcony would be disastrous for the egg. The gentle release of energy as the electrons are sequentially passed to complexes down an energy "hill" allows for a controlled process.

Looking at the ETC in more detail, we begin at complex I, where NADH is oxidized to NAD^+ by the loss of two electrons, which are then passed to the next molecule. In the process, the negative charge of the electrons pulls up hydrogen ions (protons) into the membrane protein. As the electrons join with the next electron transport molecule, the H^+ are released on the other side of the membrane. In this fashion, the passing of electrons down the ETC results in protons being actively transported from the inner mitochondrial matrix to the intermembrane space.

At complex II, another mechanism is exploited. Here, $FADH_2$ becomes oxidized, releasing its electrons to a coenzyme Q (labeled in Figure 4.12 as Q). Two H^+ join as well to neutralize the charge. Coenzyme Q floats freely through the membrane. It travels to complex III, where the H^+ ions are released to the intermembrane space and the electrons are passed on to the next molecule, cytochrome c (cyt c in the figure). Finally, in complex IV, four molecules of cyt c pass on their electrons, by now comparatively low in energy, to molecular oxygen (O_2), the final electron acceptor in the ETC. In this process, molecular oxygen is reduced to form two molecules of H_2O and four H^+ ions are pumped across the membrane.

Interestingly, the activity of the complex IV enzyme (cytochrome c oxidase) is inhibited by both cyanide (CN^-) and carbon monoxide (CO). What do you think the result is of poisoning by either of these compounds? Suffocation from the inside! The reason we breathe oxygen is that O_2 serves as the final electron acceptor in the ETC. No O_2 means the electrons moving through the ETC have no place to go. As a result, the ETC "backs up." The effect is the NADH and $FADH_2$ molecules cannot donate their electrons and oxidize to their NAD^+ and FAD counterparts. Without NAD^+ and FAD available to accept the electrons, the previous steps of cellular respiration—the Krebs cycle and pyruvate oxidation—cannot occur. In short, without this process, our cells would not have nearly enough ATP to function. In the presence of small molecular inhibitors CN^- or CO, the entire ETC is halted, resulting in death.

Let's continue discussing Figure 4.12. Now that the ETC is complete, all the electrons produced from glycolysis and the citric acid cycle have been spent, resulting in a concentration gradient of H^+ across the membrane. In other words, the intermembrane space has a high H^+ concentration, while the matrix has a relatively low one. To make a physical analogy, the pumping of H^+ out of the matrix is like a water pump that transports water up a hill. The net result is water with a lot of gravitational potential energy that can be harnessed if it falls back to the ground. In fact, this is exactly how hydroelectric power plants work to generate electricity, shown in Figure 4.13. As water falls down through turbines in a dam, the turbines spin an electric

FIGURE 4.13. *A hydroelectric power plant harnesses energy from the height difference in water, just as the ATP synthase harnesses energy from the concentration difference in H^+ across the membrane.*

generator, generating electricity. Thus, gravitational potential energy of the falling water is converted into electrical energy.

In the mitochondria, instead of having water with high gravitational potential energy falling to an area of lower energy, there is high potential energy stored in a chemical gradient, and the H^+ ions "fall" from an area of high concentration to one of low concentration. The chemical term for this phenomenon is *chemiosmosis*. Recall that the force of water across a membrane is what drives osmosis. In chemiosmosis, a species other than water provides this force—H^+ ions, in this case. Instead of falling through the turbines of a hydroelectric power plant, H^+ ions pass through the ATP synthase. This molecule is the rightmost transmembrane protein shown in Figure 4.12, shown also in the model of Figure 4.14. As protons pass through the ATP synthase by means of facilitated passive transport (facilitated diffusion), they interact with portions of the protein that resemble a turbine. In fact, the force of the H^+ ions literally makes the ATP synthase spin! As it spins, the ATP synthase catalyzes the addition of inorganic phosphate to ADP, forming ATP molecules.

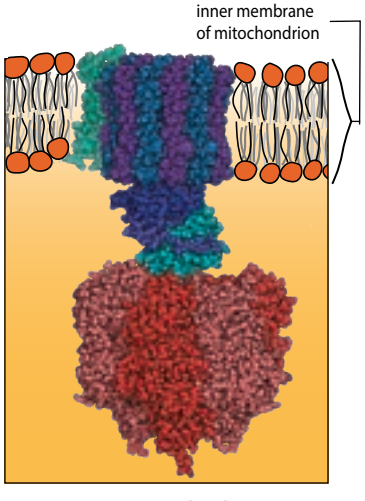


FIGURE 4.14. *3-D molecular structure of the ATP synthase (individual atoms shown). The top portion (green/purple) sits embedded in the inner mitochondrial membrane. The bottom red portion spins as ATP is generated. H^+ ions flow through the enzyme, from top to bottom.*

This elegant system produces quite a number of ATP molecules. For several reasons, the number of ATP molecules produced by cellular respiration is inexact, but the maximum is 36 or 38. One of the reasons these values are inexact is because the ratios of NADH and $FADH_2$ molecules (the electron carriers) to ATP molecules are not whole numbers. Rounded off, one NADH can produce three ATP, but the number ranges from 2.5 to 3.3. There are other considerations as well, but in the best-case scenario, one molecule of glucose produces 36 or 38 molecules of ATP. There are two values because the mitochondrial membrane is impermeable to the NADH molecules produced in the cytosol by glycolysis, so the electrons heading for the ETC inside the mitochondrion must be passed across the mitochondrial membranes to either FAD (in brain cells) or NAD^+ (in liver and heart cells) molecules inside the mitochondrial matrix. When FAD transport is employed, the maximum number of ATP molecules is 36; for NAD^+ transport, the maximum is 38.

It is amazing that the man-made mechanism for generating electricity—exploiting the laws of nature—should so close-

ly resemble the divinely designed mechanism that powers living cells. Both processes are governed by the laws of nature God established, and the biological one provides all the energy an organism needs.

Oxidative phosphorylation may be summarized as follows:

1. Occurs across the inner mitochondrial membrane.
2. Driven by high-energy electrons carried by NADH and $FADH_2$ from previous stages.
3. Electrons are passed along ETC, losing energy as they go.
4. Energy from electrons drives active transport of H^+ into intermembrane space.
5. O_2 is the final electron acceptor in the ETC, forming H_2O.
6. H^+ ions flow back into the matrix through ATP synthase (chemiosmosis), driving ATP synthesis.
7. Approximately 32–34 ATPs are formed in this final stage of cellular respiration (bringing the total for cellular respiration to 36–38 when the two from glycolysis and the two from the citric acid cycle are taken into account).

4.2.3 *Aerobic Respiration, Fermentation, and Anaerobic Respiration*

The cellular respiration process described at length in Section 4.2.2 is only one of three processes cells use to generate ATP energy. In this subsection, we distinguish between these three processes and briefly describe the other two.

As we have seen, when oxygen is available, it is the final electron acceptor in cellular respiration. Cellular respiration is an *aerobic* process. Aerobic means "requiring air," although it is actually the oxygen in the air that is required. Thus, respiration in the presence of oxygen is *aerobic respiration*. Indeed, the role of O_2 as final electron acceptor in the ETC means that oxidative phosphorylation cannot proceed in the absence of oxygen.

The other two energy processes occur in the absence of oxygen. The first is called *fermentation*, illustrated in Figure 4.15. The absence of oxygen is called an *anaerobic* ("without air") condition. Some tissues, such as muscle, use fermentation as a backup plan for generating ATP under anaerobic conditions. Normally, when enough O_2 is available, the pyruvate formed in glycolysis is oxidized by CoA and then proceeds through the citric acid cycle (the Krebs cycle). However, during periods of intense exercise, there may not be enough O_2 to keep up with the demands of the cell. In this instance, pyruvate goes into an alternate pathway, enabling glycolysis to continue. If the alternate pathway did not exist, then NADH could not be recycled into the NAD^+ that the glycolysis pathway needs to continue breaking glucose into smaller organic molecules. In the alternative scenario, NAD^+ is regenerated by coupling the $NAD^+ \rightarrow$ NADH reaction to the conversion of pyruvate into a

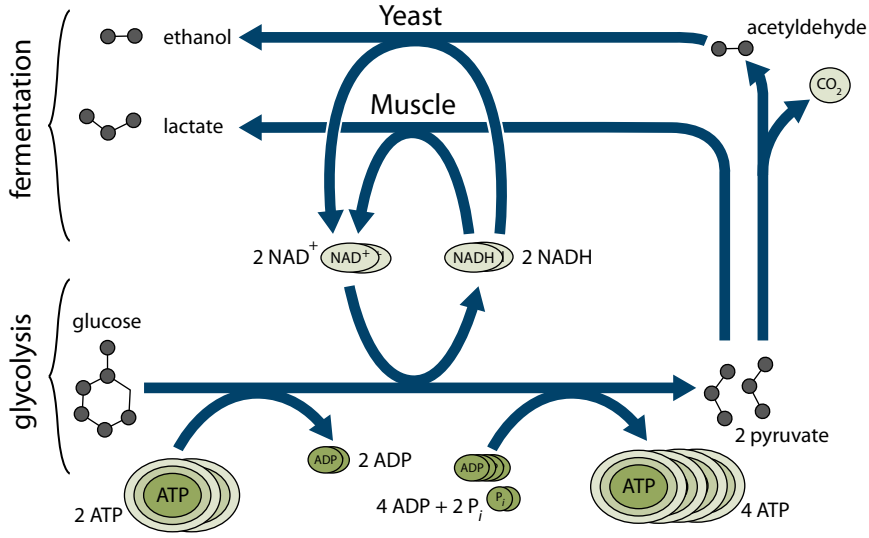


FIGURE 4.15. *Fermentation. In the absence of O_2, glycolysis continues so long as there is a way to recycle NAD^+. Yeast accomplish this by converting pyruvate into ethanol. Muscle cells instead convert pyruvate to lactate.*

molecule called lactate. In this way, muscle cells can keep generating ATP through glycolysis. You may have heard that during intense exercise lactic acid builds up in muscle tissue. This lactic acid is from the lactate generated by fermentation.

As an aside, aerobic exercise is so called because it increases the ability of the cardiovascular and respiratory systems to deliver O_2 to the cells. This enables the muscles to work harder and longer without the build-up of lactic acid.

In addition to its occurrence in muscle tissue, fermentation appears in many other natural processes. A number of species of bacteria and yeast undergo fermentation. In these instances, pyruvate is transformed into either lactate or ethanol, the primary constituent of alcoholic beverages. Beer and wine making involve transformation of glucose into alcohol by fermentation. Barrels are used to provide the oxygen-free environment for this process. Additionally, the yeast used in bread-making undergo a process of fermentation, forming CO_2 bubbles and ethanol. These products cause bread to rise and give bread its characteristically pleasant aroma, as the ethanol is evaporated during baking.

Fermentation is one way species generate ATP in the absence of oxygen. There is another possibility, used by microorgansims living in environments where there is no oxygen. This process, called *anaerobic respiration*, is essentially the same as cellular respiration, except for one main difference. The microorganisms use a different molecule as the final electron acceptor in their electron transport chains. One such molecule is sulfate (SO_4^{2-}), which is reduced into hydrogen sulfide (H_2S). Hydrogen sulfide is easily recognized by its characteristic odor, generally described as a rotten-egg smell. If you've ever been to

the Fountain Paint Pots at Yellowstone National Park, shown in Figure 4.16, you may have smelled this stinky result of anaerobic respiration.

Finally, we should also note that while prokaryotes (which are all microorganisms) do not have mitochondria, they do run the electron transport chain across their plasma membranes.

To recap, there are three methods of ATP generation. Aerobic respiration uses O_2 to accept electrons through the ETC. Fermentation proceeds in the absence of oxygen through glycolysis only, enabling NAD^+ regeneration by producing either ethanol (alcohol) or lactate. Finally, anaerobic respiration occurs in microorganisms that use molecules other than O_2 to accept electrons from their ETC.

FIGURE 4.16. *The Fountain Paint Pots at Yellowstone National Park contain anaerobic bacteria that reduce sulfate rather than oxygen. The resulting H_2S molecule gives this geologic wonder its characteristic smell of rotten eggs.*

4.2.4 *Metabolism of Other Biomolecules*

So far, we have been discussing how the monosaccharide glucose is broken down to produce a lot of energy for the cell in the form of ATP. All carbohydrates are essentially broken down in a similar way. Larger complex polysaccharides are first broken down into their monomers. These monosaccharides then enter glycolysis at different points depending on the type of monosaccharide, as illustrated in Figure 4.7. For example, fructose is converted into fructose-6-phosphate and then pyruvate, while galactose is converted into glucose-6-phosphate.

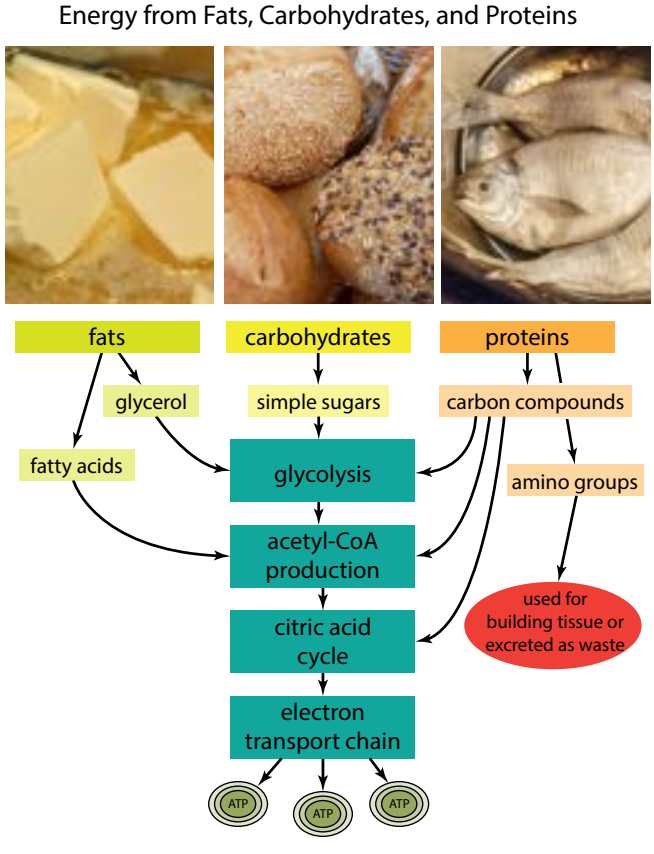


FIGURE 4.17. *Metabolism of biomolecules.*

Lipids undergo a different pathway. Recall that lipids are long chains of hydrocarbons. These hydrocarbons are broken into small segments of two carbons each. These are then converted to acetyl-CoA through a process called beta oxidation (β-oxidation). The acetyl-CoA then enters the Krebs cycle to generate molecules of NADH and $FADH_2$.

Proteins are initially broken down into their individual amino acids. The amine group of the amino acid is removed through the process of *deamination*. Subsequently, the amine group is converted into ammonia, then into urea or uric acid, and removed from the organism as a waste product. The remaining portion of the amino acid is converted into pyruvate, acetyl-CoA, or other intermediates found in the Krebs cycle as indicated in Figure 4.17.

4.3 Photosynthesis

Cellular respiration harnesses energy from organic molecules (food) and O_2, generating ATP for cells to use. Carbon dioxide is a reaction product. In a sense, *photosynthesis* is the reverse process, and the two processes are interdependent, as illustrated in Figure 4.18. Photosynthesis harnesses sunlight to synthesize sugars out of CO_2 and H_2O, with oxygen as a reaction product:

$$6CO_2 + 6H_2O + \text{light} \rightarrow C_6H_{12}O_6 + 6O_2 \quad (4.4)$$

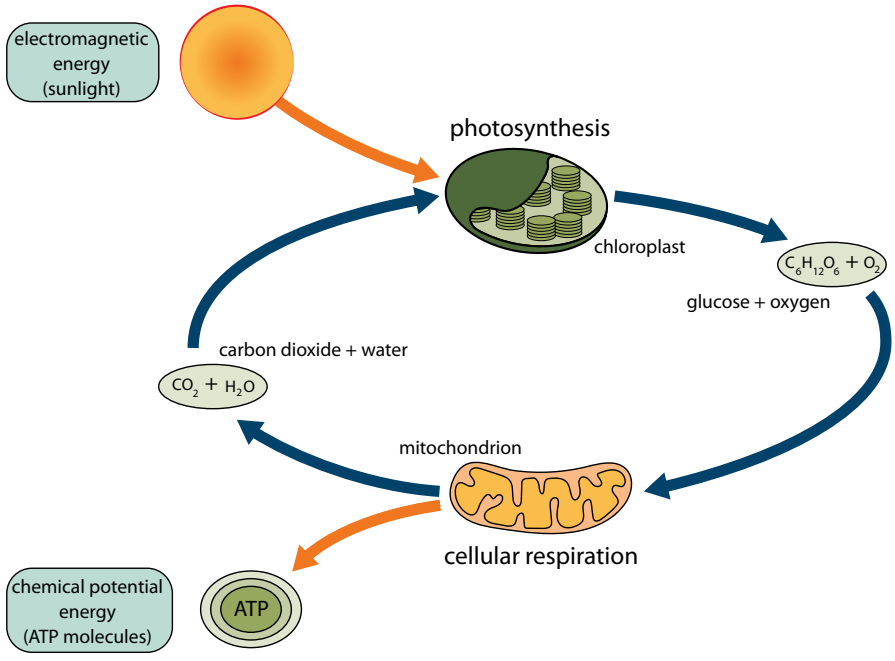


FIGURE 4.18. *The interdependence of cellular respiration and photosynthesis. The products of one process serve to power the other. In this way, all life on Earth is powered by energy from the Sun. Note that although glucose is shown here as the primary sugar produced, simpler sugars are the ones directly made by photosynthesis. These are then precursors to synthesis of many biomolecules in the cell, including glucose.*

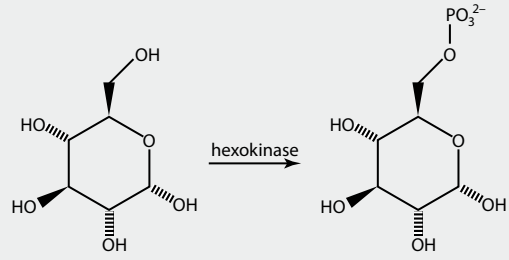
In the Wonderworld of Biology

The Energy Released by ATP Hydrolysis

When we say that the hydrolysis of ATP releases energy that powers cells, you may wonder how this works. What form is that energy in? And how is it used to power processes in the cell?

When ATP is hydrolyzed into ADP and P_i, the P_i is rarely ever just released by itself. Instead, the P_i is almost always transferred to another molecule, activating it for another chemical reaction. An organism's metabolism is a series of interconnected, stepwise chemical reactions, one after the other. For example, in the 10 intermediate steps of glycolysis shown in Figure 4.7, a small bit of energy is released as heat at each step, but most of the energy is converted into chemical potential energy in the next molecule in the series.

The very first step in the glycolysis series, catalyzed by hexokinase (mentioned in Figure 4.3), converts glucose to a molecule called glucose-6-phosphate, as the accompanying figure shows. This reaction, called phosphorylation, pulls a phosphate group from ATP and attaches it to the glucose to yield glucose-6-phosphate. The glucose-6-phosphate now has more chemical potential energy than the glucose did and can successfully undergo the next several reactions in the glycolysis pathway. A few steps later, another phosphorylation reaction occurs using another molecule of ATP.



glucose glucose-6-phosphate

All metabolic reactions are just manifestations of the law of conservation of energy. The ATP molecule is like a roller coaster at the top of a hill, with high gravitational potential energy. Successive chemical reactions represent lower hills on the up-and-down pattern that roller coasters often take, only energy is being transferred into chemical potential energy of different molecules instead of gravitational potential energy of different hills. At each step, little bits of energy are lost to heat, just as little bits of energy on roller coaster are lost to friction. When the roller coaster needs to go up an especially large hill, an additional source of energy (such as a motor with a chain) is required to supply the needed gravitational potential energy. Likewise, an ATP molecule supplies the energy via a phosphorylation reaction when the chemical reaction needs an energy boost.

And what about the heat? The heat lost from metabolism helps maintain body temperature and is eventually released from the body as infrared electromagnetic radiation. This is how snakes sense rats in the dark and is why people can be seen in the dark through the use of night-vision goggles.

Another illustration of the way the energy from ATP hydrolysis is used is in the sodium-potassium pump (Figure 3.47). The pump is a protein embedded in the cell membrane. In its normal configuration, the protein allows sodium ions in the cell to enter and bind to the protein. When a passing ATP molecule phosphorylates an amino acid in the protein, the protein's structure becomes unstable and the protein changes its shape to regain stability, ejecting the sodium ions outside the cell and allowing potassium ions outside the cell to enter and bind to the protein. When the phosphate group is removed from the protein, which happens spontaneously, the protein's structure again becomes unstable and it changes back to its original shape, opening to the inside of the cell and releasing potassium ions inside the cell.

Amazing? I should say so.

Though only certain types of organisms (plants, algae, and cyanobacteria) undergo photosynthesis, the energy they store in sugars powers almost all life on Earth! Non-photosynthetic organisms must consume other organisms to obtain food, whose energy originally came from the Sun. Furthermore, the other product of photosynthesis (O_2) is necessary for other organisms to breathe. Once again, this carefully designed, interconnected system displays God's intricate care for his creatures, evident in the chemical reactions of molecules, the machinery of cells, and in interdependence of different species throughout ecosystems.

4.3.1 *Chlorophyll Molecules*

In the first step of photosynthesis, electromagnetic radiation (light) is captured and converted into chemical potential energy, a process that occurs in the chloroplasts of cells. As you are probably aware, light exhibits both wave-like properties and particle-like properties. When discussing the particle-like properties, we refer to the individual, massless particles of light as *photons*. The real biological solar panels present in the chloroplasts are special photoreactive molecules, whose chemistry allows for photons of light to excite individual electrons into high-energy states. In other words, the electrons absorb the energy of the photons and move to higher-energy orbitals in the atom (or into other processes). The energy is then passed on in photosynthesis as the electrons fall back down to lower-energy states.

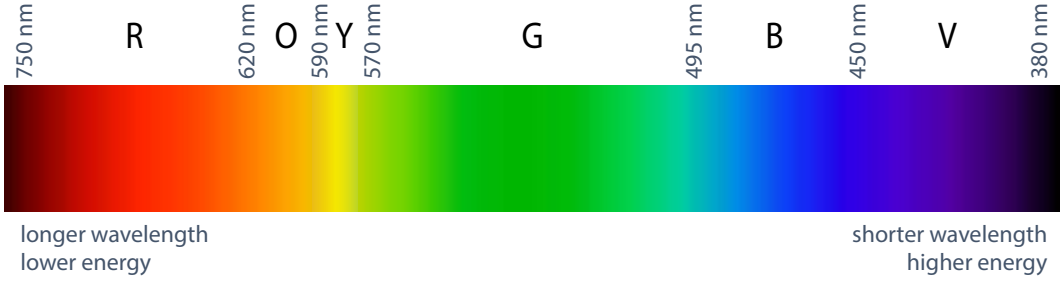


FIGURE 4.19. *The visible light portion of the electromagnetic spectrum showing approximate reference wavelengths for different colors.*

Remember that electromagnetic radiation is a massless form of energy that can propagate through empty space. Electromagnetic radiation includes radio waves, visible light, and X-rays, all as part of the electromagnetic spectrum. These different parts of the electromagnetic spectrum are distinguished one from another by their wavelength. The visible light portion of the electromagnetic spectrum is illustrated in Figure 4.19. Various wavelengths, associated with specific colors, are shown on the diagram.

Figure 4.20 illustrates the most important of the photoreactive molecules—*chlorophyll a*. The chemical bonds of this molecule allow its electrons to have only specific energies. Recall that electrons in atoms are only allowed certain energies and the orbitals they are in depend on the energy they have. One consequence of these specific energies is that each electron in an atom can only absorb electromagnetic radiation (pho-

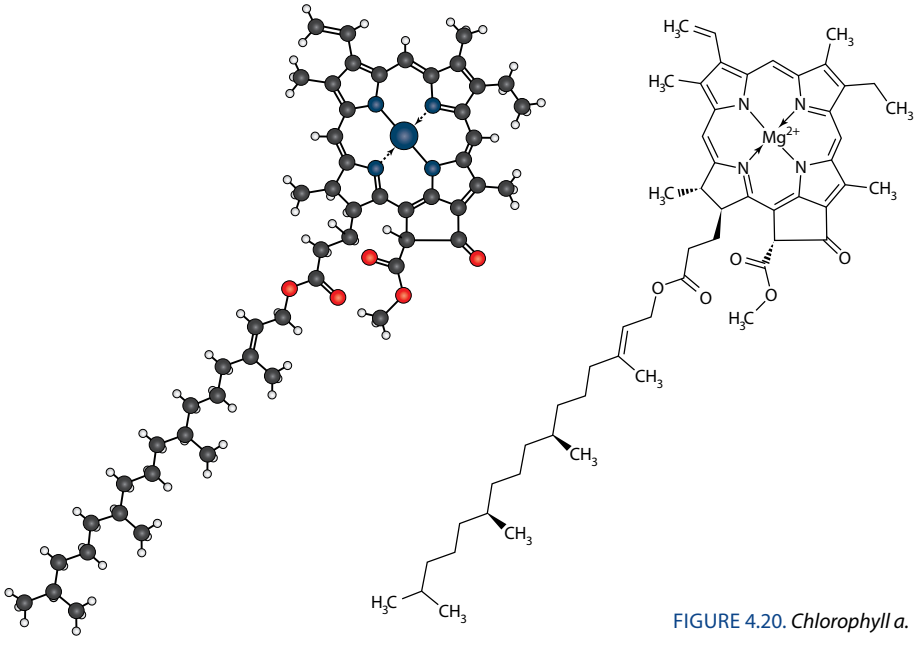


FIGURE 4.20. *Chlorophyll a.*

tons) of a specific energy, and thus specific wavelength. (The energy and wavelength of a photon are correlated by the Planck relation, $E = h \cdot f$, where E is the energy, h is the Planck constant ($h \cong 6.626 \cdot 10^{-34}$ J·s), and f is the frequency.) If the wavelength of light matches the energy required for an electron to jump to an excited state, the electron absorbs the energy of the light and is excited to a higher-energy orbital or freed to leave an atom and into another chemical process altogether. If the wavelength of light doesn't match the energy required for the electron to change energy states, the light is reflected or simply passed through (transmitted). The reflected wavelengths of light give objects the colors we see—after reflecting, rays of light travel to our eyes where they are detected and signals are sent to our brains, which we interpret as particular colors.

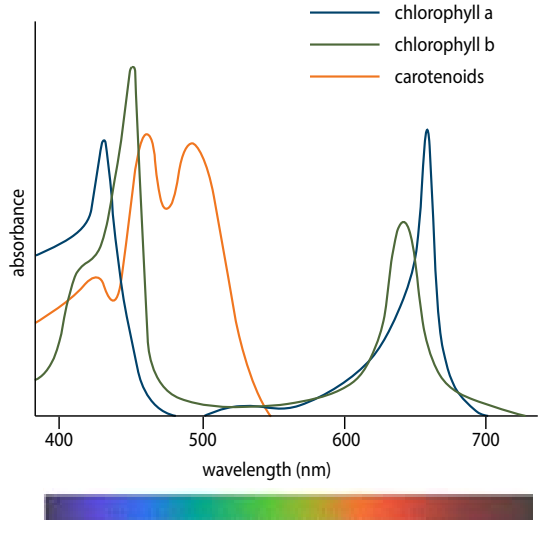


FIGURE 4.21. *Absorption spectra of chlorophylls a and b and the carotenoid pigments.*

The phenomena of absorption and reflection explain why plants appear to be green. The *absorption spectrum* of a molecule is represented by a graph showing the strength of the molecule's light absorbance versus wavelength. Figure 4.21 shows the absorption spectra of the molecules chlorophyll a and chlorophyll b. Where the curve is high in this graph, light is strongly absorbed by the molecule; where the curve is low, light is mostly, or all reflected. Notice that both molecules absorb strongly in the blue and red regions, but they absorb almost nothing in the green region of the spectrum. Consequently, red and blue wavelengths are absorbed and green wavelengths are reflected, causing us to see green when we look at the leaves of plants. Figure 4.22 shows the chloroplasts in plant cells. The chloroplasts are green because of the chlorophyll molecules they contain.

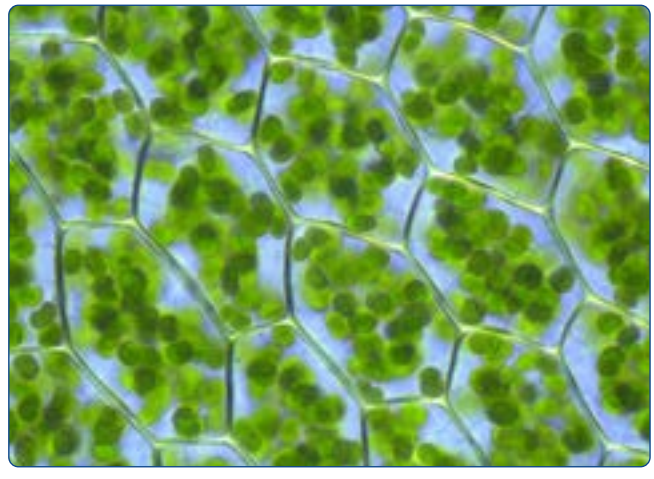


FIGURE 4.22. *Chlorophyll molecules are concentrated in the chloroplasts of plant cells. In this light microscope image, you can see that only the chloroplast organelles (small green circles) appear green, not the other organelles (which appear to be transparent). Notice the clearly visible cell walls.*

While we are on the subject of the colors of leaves, Figure 4.21 also shows the absorption spectrum of the carotenoids, one of the many pigments in plant leaves. Carotenoids are responsible for the yellow colors in many different plants and animals. In plants, the yellows of the carotenoids are masked by the presence of green chlorophyll. In the autumn, they provide one of the dominant colors in the leaves of deciduous trees.

4.3.2 *The Light-Dependent Reactions*

By absorbing energy in sunlight, the chlorophyll molecules power the process of photosynthesis. Figure 4.23 shows a schematic overview of this process. The overall purpose of photosynthesis is to convert CO_2 into sugars, using energy from the Sun. The byproduct of this series of chemical reactions is oxygen. The chlorophyll molecules are embedded in the thylakoid membrane of the chloroplast as part of a large protein complex. Here, the first phase of photosynthesis—the light-dependent reactions—occurs. The purpose of the *light-dependent reactions* is to synthesize ATP and NADPH, using energy from light and releasing O_2 as a waste product. NADPH is an electron carrier molecule similar to NADH, differing only by the presence of an extra phosphate group.

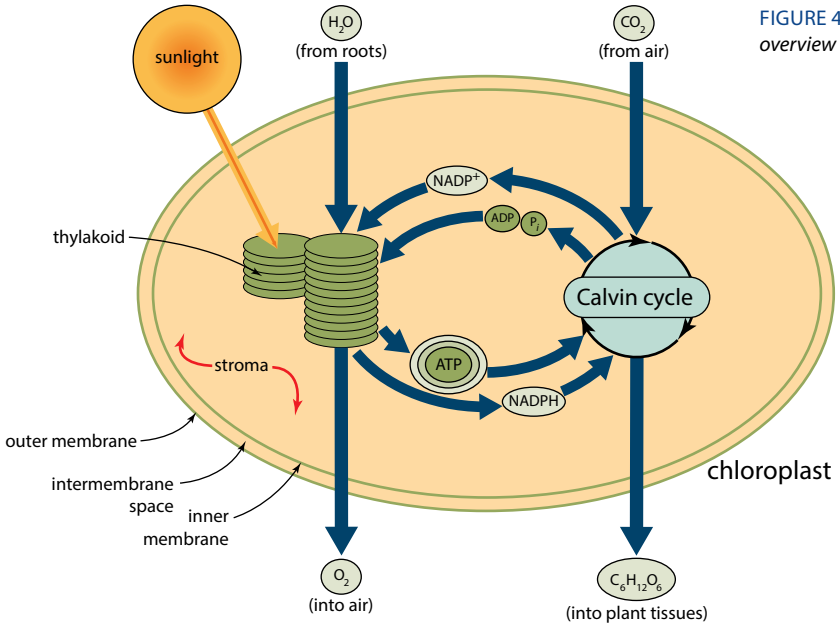


FIGURE 4.23. *A schematic overview of photosynthesis.*

The light-dependent reaction can be expressed as follows:

$$ADP + NADP^+ + P_i + H_2O + light \rightarrow O_2 + ATP + NADPH \quad (4.5)$$

As illustrated in Figure 4.24, the light-dependent phase of photosynthesis uses an electron transport chain mechanism to drive chemiosmosis of H^+ through an ATP synthase, in a manner similar to the process that occurs in cellular respiration. The first protein complex embedded in the thylakoid membrane, shown on the left side of the figure, is Photosystem II (PS II), also known as P680 because the chlorophyll-a pigments absorb wavelengths near 680 nm most effectively. (Don't be confused

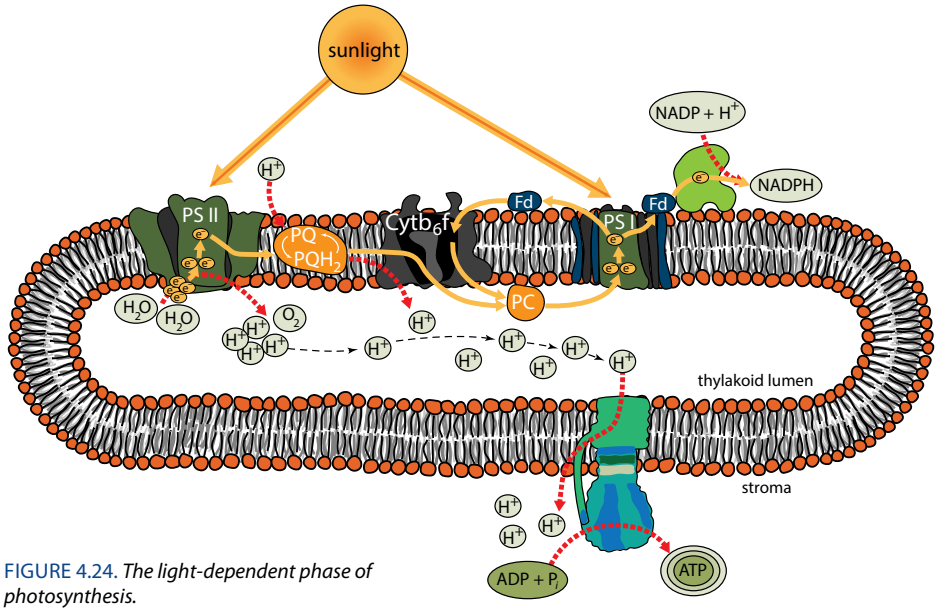


FIGURE 4.24. *The light-dependent phase of photosynthesis.*

by the name. PS II is so called because it was discovered after Photosystem I, even though it is the first step in the pathway). A photosystem is a complex of proteins and other organic molecules. Its structure consists of a central reaction-center complex surrounded by light-harvesting complexes. The light-harvesting complexes contain several different types of pigment molecules. This allows the photosystem to capture a wider range of light wavelengths than it would if it contained only one type of pigment molecule. When light strikes one of the pigment molecules, its electrons are excited to a higher energy level. When the electrons fall back to their ground state, the energy is passed to electrons in a neighboring pigment molecule. As a result, the light energy is transferred from one pigment molecule to another inside the light-harvesting complex. The light energy is eventually transferred to a pair of chlorophyll-a molecules embedded in the reaction-center complex. The energy excites electrons within the chlorophyll molecules, which pass the high-energy electrons to a primary electron acceptor located within the reaction-center complex (PS II). It is therefore the reaction-center complex that converts the light energy into chemical energy. The excited electron is then passed out of the photosystem and enters the electron transport chain (the path indicated by the yellow arrows) embedded in the thylakoid membrane.

The electrons located in the chlorophyll molecules of the reaction-center complex are replaced by electrons provided by splitting water. An enzyme within the photosystem splits two water molecules into four electrons (e^-), four hydrogen ions (H^+), and a molecule of oxygen (O_2).

The electrons proceed down the electron transport chain, which is composed of three molecular complexes, plastoquinone (PQ), a cytochrome c complex (b6f), and plastocyanin (PC). Each of these molecules carries out redox reactions as the electrons are passed from one to the other. As electrons are passed from the electron carrier PQ to cytochrome c, the energy provided pulls H^+ ions from the stroma and actively transports them into the thylakoid lumen. The electron carrier PC carries the electrons from cytochrome c to the second photosystem in the thylakoid membrane, Photosystem I (PS I) also known as P700. In PS I, the electrons are once again excited by a photon of light, just as they are in PS II. The excited electrons are passed to a second, short, electron transport chain through the protein ferredoxin (Fd) and then on to the enzyme $NADP^+$ reductase. This enzyme facilitates the reduction of $NADP^+$ to NADPH by transferring a pair of electrons and one H^+ to $NADP^+$. This results in the production of the high-energy molecule NADPH, which is used in the second part of photosynthesis. An important difference in this second electron transport chain is that no H^+ are moved across the thylakoid membrane.

A large concentration gradient of H^+ builds up in the thylakoid lumen, due to active transport of H^+ and as a byproduct of splitting water. These H^+ ions travel through the ATP synthase

out into the stroma, catalyzing the formation of ATP, just as in cellular respiration.

With the formation of ATP and NADPH, we now have the high-energy molecules needed to carry out the second phase of photosynthesis, the Calvin cycle.

4.3.3 *The Calvin Cycle*

The second phase of *photosynthesis*, called the *light-independent* phase or *Calvin cycle*, uses the energy stored in ATP and NADPH to run this reaction:

$$ATP + NADPH + CO_2 \rightarrow \text{sugars} + ADP + NADP^+ \quad (4.6)$$

The Calvin cycle is named for American biochemist Melvin Calvin (Figure 4.25), who studied the pathway along with colleagues Andrew Benson and James Bassham. Their technique involved labeling or marking compounds with atoms of carbon-14. The radioisotope-labeling technique allowed Calvin to trace a particular carbon atom through the entire pathway. For this work, Calvin was awarded the 1961 Nobel Prize in Chemistry.

FIGURE 4.25. *Melvin Calvin (1911–1997).*

The purpose of the Calvin cycle is to synthesize sugars out of CO_2, using energy supplied by ATP and NADPH from the light-dependent phase of photosynthesis. The main sugar produced in photosynthesis comes from glyceraldehyde 3-phosphate (G3P), a molecule we saw earlier as an intermediate in glycolysis. The Calvin cycle takes place in the stroma of the chloroplast and can be broken down into three basic phases.

Figure 4.26 shows each phase of the Calvin cycle in more detail. In phase one (carbon fixation), three CO_2 molecules from the atmosphere join with three five-carbon molecules of ribulose 1,5-bisphosphate (RuBP) already in the cycle. This step is catalyzed by an enzyme called ribulose 1,5-bisphosphate carboxylase-oxygenase, and known by the catchy nickname RuBisCO (the most abundant enzyme in the world!). One CO_2 combines with one RuBP to form a short-lived six-carbon molecule, which splits into two three-carbon entities called 3-phosphoglycerate (3-PGA), for a total of six molecules.

In the next phase (reduction), ATP and NADPH from the light-dependent reactions are used. First, six molecules of ATP are used to phosphorylate (add a phosphate group) to each of the 3-PGA molecules producing six molecules of 1,3-bisphosphoglycerate. The 1,3-bisphosphoglycerate are reduced into glyceraldehyde 3-phosphate (G3P) with the input of six NADPH molecules. G3P is the primary product of the Calvin cycle. It is the precursor to a number of larger molecules, including glucose, that the cell synthesizes. Because only one molecule of G3P is exported for each turn of the Calvin cycle, it takes two turns of the Calvin cycle to make one glucose molecule. This also means it takes 18 molecules of ATP and 12 molecules of NADPH to make one glucose.

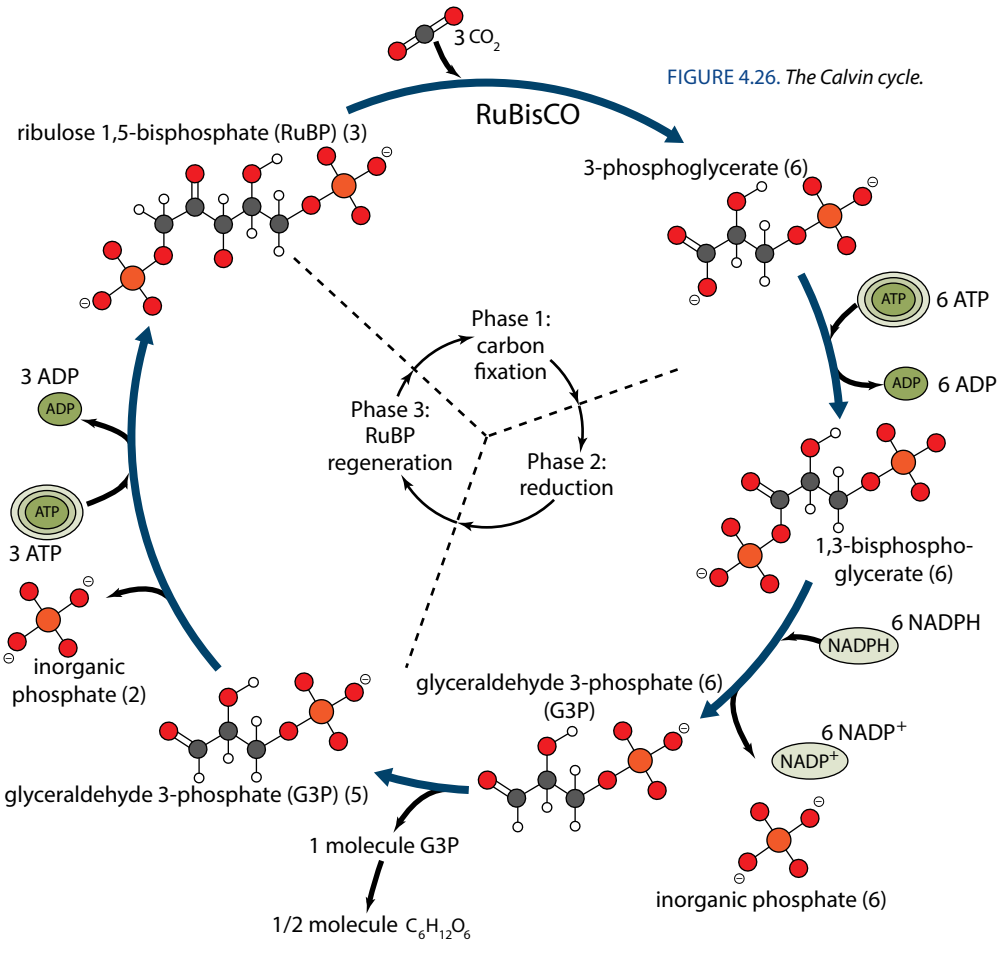


FIGURE 4.26. *The Calvin cycle.*

Of the six G3P molecules synthesized, only one goes on toward other metabolic pathways. The remaining five move into the third and final phase, regeneration of RuBP. In this phase, the five molecules of G3P use three molecules of ATP and are rearranged (through a number of steps not shown here) back into three molecules of RuBP to begin the cycle again.

The Calvin cycle may be summarized as follows:

1. Three CO_2 molecules enter from the environment (cytoplasm and stroma)
2. These are converted into the sugar precursor G3P.
3. Energy for this process comes from ATP and NADPH generated by the light-dependent photosynthetic reactions.

4.3.4 *Photorespiration and Photosynthetic Adaptations*

RuBisCO is the most abundant protein on earth. As we have seen, the enzyme RuBisCO is responsible for attaching a molecule of CO_2 to a RuBP molecule to begin the Calvin cycle. This process results in the production of sugar molecules for the

In the Wonderworld of Biology

Light and Humans

Photosynthesis does not occur in humans, so we are not able to make our own food. However, special cells in our eyes and skin do render many of our physiological processes sensitive to light.

In addition to the cells in our eyes responsible for vision, we have photosensitive cells that detect current levels of light exposure (even through closed eyelids) and send signals to the universal timekeeper of our bodies—a region of our brain called the SCN. In low-light or dark conditions, the SCN signals for melatonin production. Melatonin is a hormone that causes us to feel sleepy. During daylight hours, melatonin production stops.

Under normal conditions, light exposure leads to a 24-hour sleep/wake cycle (circadian rhythm) that is tied into many other aspects of our physiology, including metabolism, mood, learning, and memory. When the time periods of light and dark exposure are disrupted—such as when flying to another part of the world—the body takes a few days to adjust to the new location, causing the body to experience the fatigue and irregular sleep patterns of jetlag. Shift workers often have trouble keeping a consistent circadian rhythm when they must sleep during the day and work at night. In far northern regions, where winter days are very short, the depressive symptoms of SAD (seasonal affective disorder) are a common problem.

Research indicates that short-wavelength light is especially activating to the SCN. Commonly called "blue light," it is a major component of the light emitted from screens, such as those on smartphones. Perhaps, then, excessive use of one's smartphone right before bed isn't the best way to keep one's circadian rhythm in sync. Lots of light exposure during the day (go outside!) and limited exposure at night is the pattern that best maintains your circadian rhythm.

plant. Without RuBisCO, plants would be unable to make their own food. We can see how very important this enzyme is for plant life as well as all life on Earth. Interestingly, the active site on RuBisCO that binds CO_2 also has a strong affinity for O_2. In mild conditions, this is not generally a problem. However, when conditions become hot, many plants close their stomata (defined below) in order to prevent the loss of H_2O molecules due to evaporation. When this happens, the O_2 produced in the light reactions of photosynthesis is unable to escape out of the leaves and builds up. At the same time, the leaf is unable to bring in more molecules of CO_2. The effect is a higher concentration of O_2 than CO_2 in the leaf cells. The excess concentration of O_2 causes it to bind to RuBisCO instead of CO_2. The result is a two-carbon molecule called 2-phosphoglycolate. This

FIGURE 4.27. *Rice, a staple crop for many populations, is a C3 plant.*

molecule is rearranged within peroxisomes and mitochondria of the plant cell to regenerate RuBP, a process that uses ATP and NADPH, but does not produce any sugar molecules. Instead, it releases CO_2 as a product. This process is referred to as *photorespiration*. In short, photorespiration uses up the plant's energy and makes it harder for the plant to grow in hot and arid environments.

Plants that undergo regular photosynthesis and are more subject to photorespiration are referred to as C3 plants. An example of a C3 plant is rice, shown in Figure 4.27, a major food source for human beings. In order to counter photorespiration, some groups of plants have developed adaptations that reduce the amount of photorespiration that occurs. There are two major alternative strategies that we mention here: C4 photosynthesis and CAM photosynthesis.

As illustrated in Figure 4.28, leaves in plants have passages—called stomata—that open to allow environmental CO_2 to enter and H_2O molecules to escape during transpiration (Figure 2.7). The cells surrounding the stomata—called guard cells—enable them to open when cells have abundant water (so that the water may evaporate) and close when cell vacuoles do not have enough water (to prevent too much water loss).

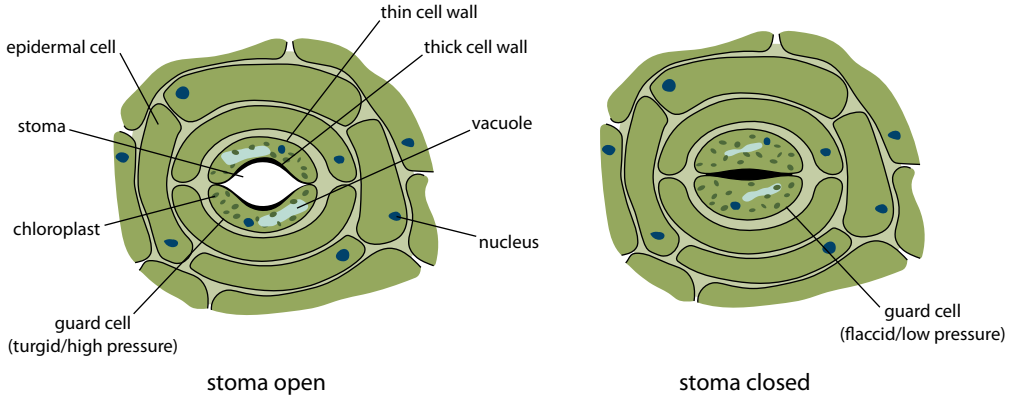


FIGURE 4.28. *Stomata, openings in leaves that allow for entrance of CO_2 and exit of H_2O. The stomata open when water is abundant and close when water is scarce.*

In hot, dry climates, plants close their stomata to prevent too much water loss. The side effect of stomata closure is that not enough CO_2 is present for photosynthesis, causing the Calvin cycle to use O_2 instead of CO_2. This wastes energy but helps keep the plant alive.

C4 plants, such as the corn and sugarcane shown in Figures 4.29 and 4.30, avoid this wasteful scenario by separating parts of the Calvin cycle into different cell types. In this way, CO_2 is collected and stored in one particular cell type at high concentration, enabling the Calvin cycle to run normally.

Another strategy is utilized by CAM plants, such as pineapple, shown in Figure 4.31. CAM plants separate the metabolic processes by night and day. To prevent excessive water loss, the plants open their stomata at night, allowing entrance

of CO_2, which is stored by reaction with organic carrier molecules. During the daytime, the stomata close, but the stored CO_2 is still available to run the rest of the Calvin cycle.

FIGURE 4.29. *Corn is a C4 plant, separating steps of the Calvin cycle into different cell types so as to concentrate CO_2 while retaining water.*

FIGURE 4.30. *Sugarcane, another C4 plant.*

FIGURE 4.31. *Pineapple uses another photosynthetic strategy, the CAM pathway, in which metabolic processes are separated by night and day.*

Chapter 4 Exercises

SECTION 4.1

1. Explain the role of a catalyst in a chemical reaction.
2. Compare and contrast combustion, redox, and hydrolysis reactions.
3. Classify the following chemical reactions as combustion, redox, or hydrolysis:
   a. sucrose + water → glucose + fructose
   b. $C_3H_8 + 5O_2 \rightarrow 3CO_2 + 4H_2O$
   c. $4Fe + 6O_2 \rightarrow 3Fe_2O_3$
   d. $CH_4 + 2O_2 \rightarrow CO_2 + 2H_2O$
   e. triglyceride + base + water → soap
4. Explain why ATP is the ideal energy currency for the cell.
5. Distinguish between endergonic and exergonic chemical reactions.
6. Draw and label two reaction coordinate diagrams: one for an exergonic reaction and one for an endergonic one. Include the pathways with and without an enzyme.
7. Describe in detail how enzymes facilitate biological chemical reactions.
8. Explain the process of feedback inhibition in a series of enzyme-catalyzed chemical reactions.

SECTION 4.2

9. What is the overall purpose of cellular respiration?
10. Make a table that lists the inputs, lists the outputs, and describes the overall results for each of the four stages of cellular respiration.
11. Using your table from the previous exercise, describe in words how each of the four stages of cellular respiration contribute to the overall purpose. (Describe the process and intermediate results accomplished by each phase.)

12. During glycolysis, how many ATPs are invested and how many are generated?
13. Make a flow chart showing the catabolism of glucose into pyruvate in glycolysis.
14. Describe the role of coenzyme A in cellular respiration. How does this prepare pyruvate for entrance into the citric acid cycle?
15. Describe the function of NADH and $FADH_2$ in glycolysis and the citric acid cycle.
16. Name two major scientists who contributed to our understanding of the citric acid cycle and describe their findings.
17. What happens to the CO_2 generated in the citric acid cycle?
18. Create a diagram depicting the transformation of citric acid back into oxaloacetate in the citric acid cycle. Indicate where NADH, $FADH_2$, and ATP are generated within the cycle.
19. Where in the cell do glycolysis and the citric acid cycle take place?
20. Write paragraphs that summarize the overall chemical transformations that occur in glycolysis and the citric acid cycle.
21. How is active transport used in the electron transport chain?
22. Describe what happens to the potential energy of each electron as it is passed down the electron transport chain.
23. Describe the mechanism by which coenzyme Q transmits electrons.
24. Track the path of an electron from NADH through the ETC to its final destination.
25. What is the final electron acceptor of the ETC? What happens to energy production in its absence?
26. Describe how poisons such as cyanide and carbon monoxide affect cytochrome c oxidase (the final enzyme in the electron transport chain of cellular respiration).
27. Compare the chemical gradient of H^+ ions across the inner mitochondrial membrane to the process by which hydroelectric power is generated.
28. Describe the mechanism of the ATP synthase.
29. Summarize the steps that occur in oxidative phosphorylation. How do these steps contribute to the overall goal of oxidative phosphorylation?
30. Compare and contrast aerobic respiration, anaerobic respiration, and fermentation.
31. Describe the process of beta oxidation and its role in fat metabolism.
32. Explain how proteins are broken down to provide energy for the cell.

SECTION 4.3

33. What is the overall goal of photosynthesis?
34. Explain why photosynthesis and cellular respiration are interdependent.
35. Name the colors of light that are most absorbed by plant pigments such as chlorophyll.
36. Suppose a person decides to grow some plants in a basement under green-light emitting lamps. Describe the results one should expect from this project.
37. Some foods, such as carrots and squash, appear to be orange/yellow. Based on what you know about absorption and pigment molecules, form a hypothesis as to why these vegetables display these colors.

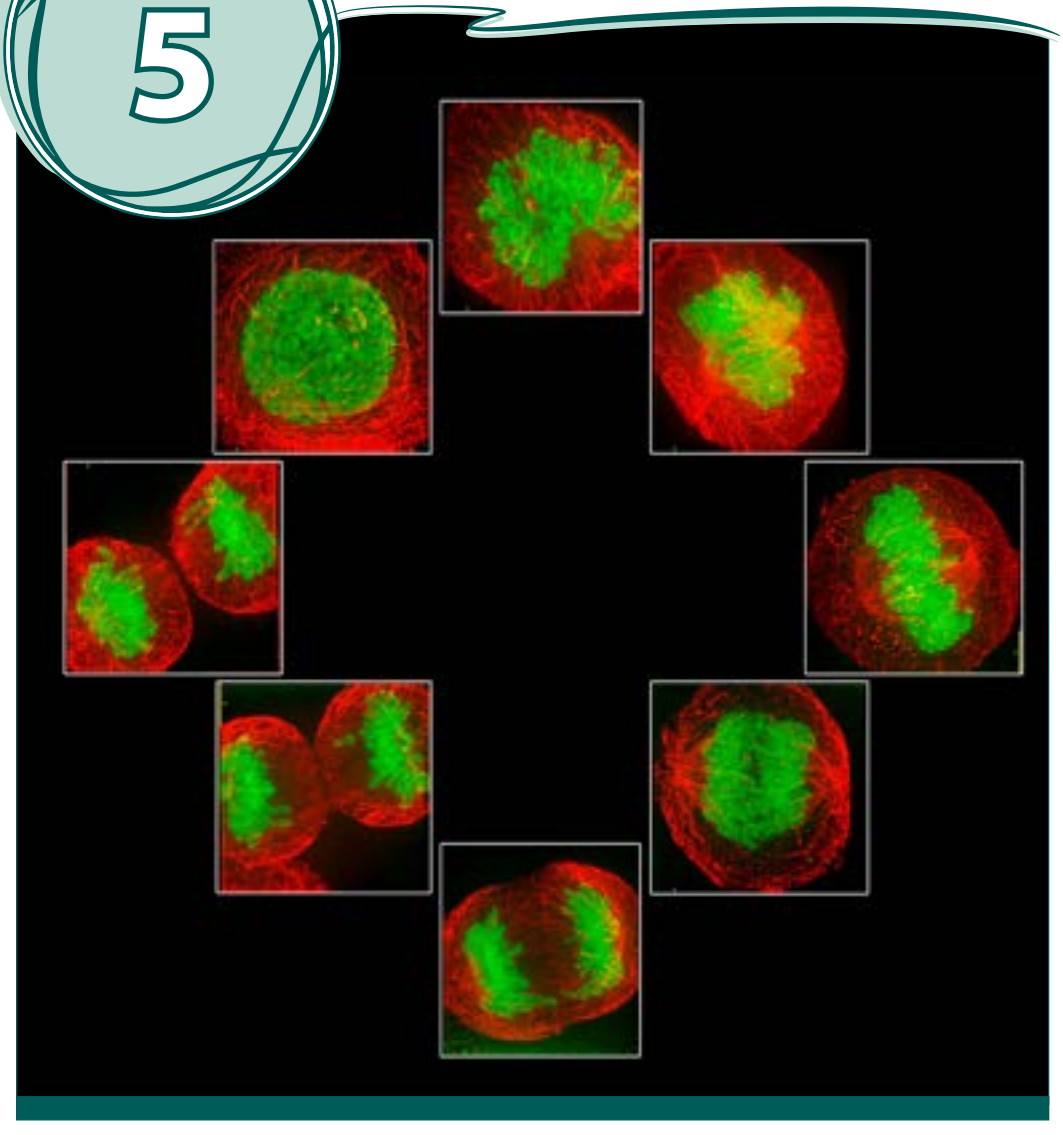
38. How does the arrangement of electrons in atoms relate in general to the spectrum of colored light that a particular compound absorbs? How does this arrangement determine the color that a compound appears?
39. Briefly describe how the light-dependent and light-independent phases of photosynthesis contribute to its overall goal.
40. Explain how light energy is converted into chemical energy in the photosystems of the chloroplasts.
41. Compare and contrast the electron transport chain of photosynthesis to that of cellular respiration.
42. Name and describe two mechanisms by which the H^+ concentration gradient is created during the light-dependent phase of photosynthesis.
43. Briefly describe the mechanism of the Calvin cycle.
44. Compare and contrast the Krebs cycle and the Calvin cycle.
45. Explain two strategies used by plants in dry climates to minimize water loss during photosynthesis.

REVIEW QUESTIONS

46. Briefly describe the six requirements for life.
47. How does the capacity to cycle matter and energy (a characteristic of life discussed in this chapter in detail) interrelate with the other characteristics of life?
48. Of the classes of biomolecules discussed in Chapter 2 (carbohydrates, lipids, proteins, and nucleic acids), which types do you see at play in cellular respiration and photosynthesis? Identify as many as you can.
49. List as many cell organelles as you can. Identify the ones involved in photosynthesis and those involved in cellular respiration. For those involved in one or the other of these processes, describe their roles.
50. Define active transport and discuss several ways this can be accomplished across a cell membrane. Which of these modes are involved in photosynthesis and cellular respiration?
51. Define passive transport and discuss several ways this can be accomplished across a cell membrane. Which of these modes are involved in photosynthesis and cellular respiration?

CHAPTER 5

Cell Signaling and the Cell Cycle



How do cells grow, divide, and transmit genetic information to the next generation? These stunning fluorescence microscope images show a HeLa cell doing just that. (See the opening image for Chapter 3 for the origin of HeLa cells). The process of cell division, or mitosis, *proceeds in distinct stages, shown clockwise from the top: prophase, prometaphase, metaphase, anaphase, telophase, cytokinesis, late cytokinesis and interphase. In these images, spindle fibers are stained red and the genetic material (chromosomes) are fluorescing green. In this chapter, we describe how cells communicate with each other and their environment. In addition, we consider how cells copy their DNA and divide, faithfully passing DNA on to daughter cells.*

OBJECTIVES

After studying this chapter and completing the exercises, you should be able to do each of the following tasks, using supporting terms and principles as necessary.

SECTION 5.1

1. Describe cell communication.
2. List several examples of how cell communication is used.
3. Describe Earl Sutherland's contributions to our understanding of cell communication.

SECTION 5.2

4. List and briefly describe the three stages of cell signaling.
5. List and describe four different examples of signal reception.
6. Explain how a cell signal is relayed inside a cell.
7. Describe two mechanisms for signal transduction.
8. Explain why different cells have varied responses to the same signal.

SECTION 5.3

9. Describe the stages of the cell cycle.
10. Describe the chemical reaction that occurs as a new nucleotide is added to a growing DNA strand.
11. List and describe the roles of five enzymes involved in DNA replication.
12. Describe how the replication process begins.
13. Distinguish between leading strands and lagging strands during DNA replication and describe the process and direction of replication for each one.
14. Draw, list, and describe the events that occur at each stage of mitosis.
15. Differentiate between the process of cytokinesis for animal and plant cells.
16. Describe the roles of the three cell-cycle checkpoints, including the consequences of the criteria not being met.
17. Explain how cyclins and Cdks regulate the cell cycle.
18. Describe the cellular malfunctions that lead to cancer.
19. Describe the role of telomeres in the cell.

5.1 Cell Signaling: Introduction

In Chapter 1, we describe several characteristics of living things. One of these characteristics is the ability to grow, develop, and reproduce. In this chapter, we explore how cells are able to reproduce and—more importantly—how they know when to *commence* reproduction. Reproduction at the wrong time or too often is associated with diseases such as cancer. The signal for cells to reproduce comes through a complex mechanism known as *cell communication*.

Cell communication is the method by which cells communicate with each other—they send messages by means of chemicals. A cell receives a chemical signal released from either the environment or another cell, and internalizes the signal. This leads to a cellular response, such as undergoing reproduction (cell division). This process of going from signal reception,

to transduction of the signal, to a cellular response is the *signal-transduction pathway.*

Cell communication is used in virtually every living organism. Bacteria cells use cell communication to monitor the cell density—the number of bacteria cells present in the environment. If there are few bacteria cells, the bacteria respond by undergoing cell division. If the bacteria population starts to get too large to be supported by the resources in the environment, cell communication enables the bacteria to communicate with each other to modify or slow the growth of the bacteria colony. We see examples in eukaryotic cells as well. Yeast cells use cell communication to identify the proper cell for mating. There are different sexes of yeast, *a* and α. Mating requires an *a* strain to find an α strain, and this is accomplished using cell communication. Once the signal is received, the yeast cell generates a projection that reaches out towards the opposite yeast sex. This projection, depicted in Figure 5.1, is known as a *shmoo.* (Yes, sometimes biology is comic.) Multicellular organisms also use cell communication. Plant cells release chemicals known as *cytokines*. This group of plant hormones is responsible for regulating plant growth in the roots and stems of plants. Animals use hormones to send messages throughout the organism. One example is growth hormone. This hormone is released from the brain and acts on bone and muscle cells, causing them to undergo cell division so that the bone and muscle grow.

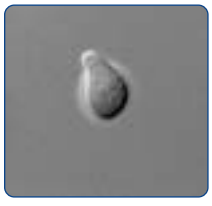


FIGURE 5.1. *Yeast shmoo.*

FIGURE 5.2. *American pharmacologist and biochemist Earl Sutherland (1915–1974).*

The first scientific encounter with cell signaling was described by the American biochemist Earl Sutherland, pictured in Figure 5.2. He was studying glycogen catabolism, and specifically how it is affected by the epinephrine and glucagon hormones. Recall that the metabolism of polysaccharides requires the use of specific enzymes. For breaking down glycogen into glucose, the enzyme is phosphorylase. Sutherland's experiments led him to conclude that epinephrine does not interact with the enzyme directly, but first binds to a receptor on the cell membrane. The receptor stimulates the formation of a second molecule (cyclic AMP, or cAMP) that activates the phosphorylase. Thus, the "second messenger" is the compound that carries information into the cell, resulting in a cellular response. Dr. Sutherland was awarded the Nobel Prize in Physiology or Medicine in 1971 for his work on understanding the mechanism of hormones acting through a messenger.

5.2 Stages of Cell Signaling

Cell signaling, or *signal transduction*, can be broken down into three basic stages: reception, signal transduction, and cellular response, illustrated in Figure 5.3. Reception is the event by which a signal molecule, also known as a *ligand,* binds to a protein receptor on the membrane of the cell. The ligand-receptor interaction triggers the next step, signal transduction, which converts the signal into a cell response. There are numerous ways a cell can respond to signal transduction. Some

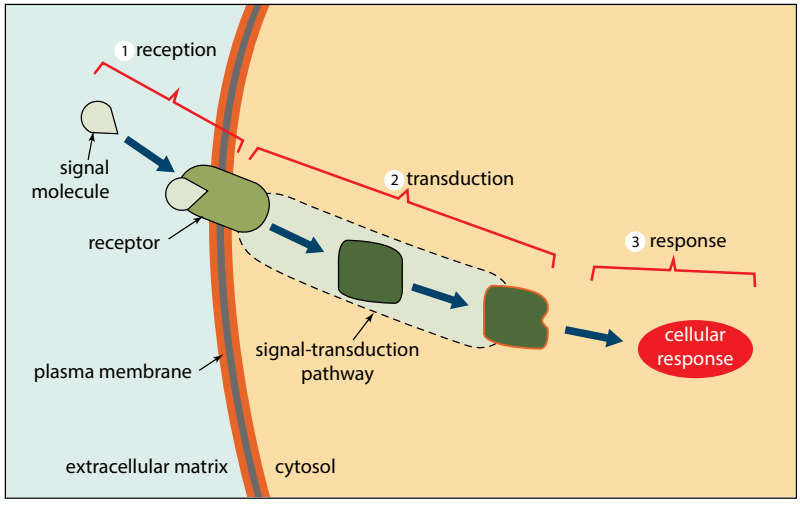


FIGURE 5.3. *Three stages of signal transduction.*

examples include the activation of enzymes, such as in the case of epinephrine and phosphorylase, the activation or "turning on" of genes, or the rearrangement of the cytoskeleton. In the following pages, we explore these three stages in more detail.

5.2.1 *Reception*

The first step in the cell-signaling pathway is reception of the ligand by a receptor molecule. Generally, the receptor molecule is embedded in the plasma membrane, and thus is referred to as a cell *transmembrane receptor*. There are also instances when the receptor molecule is within the cell, an *intracellular receptor*. Each ligand binds to a specific receptor, like the fit we observe between an enzyme and its substrate. Once the ligand binds to the receptor, one of two things occurs. Either the receptor molecule changes shape, or two or more receptor proteins join together. Some examples of common cell receptors will illustrate the process.

The G protein-coupled receptor (GPCR) is one of the most important for medicine because it is the target for approximately half the drugs currently on the market. The GPCR is a transmembrane protein that binds to a protein complex called G protein, consisting of α, β, and γ (alpha, beta, and gamma) subunits. As illustrated in Figure 5.4, in its inactive state, the G protein is bound to a GDP molecule (similar to ADP) and is separate from the receptor protein. When a ligand binds to the receptor, it results in a change to the receptor's shape, enabling the G protein to bind to the GPCR. Upon binding, a GTP molecule binds to the G protein, replacing the GDP molecule, and thereby activating the G protein. The activated G protein dissociates; the separated subunits move through the membrane to bind to other enzymes, activating the enzymes (that is, switching them to an active conformation), and causing the enzymes to catalyze reactions that transmit the signal into the cell. Finally, GDP replaces GTP in the

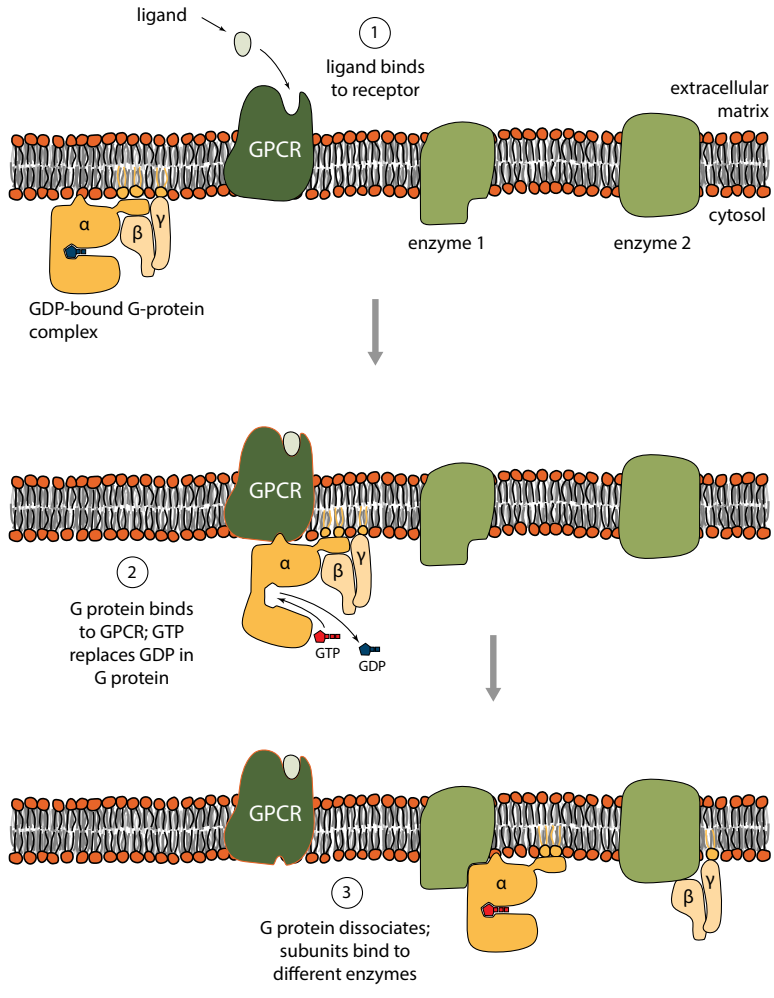


FIGURE 5.4. *G protein-coupled receptor.*

G protein, the subunits bind back together, and the G protein returns to its inactive state.

Another common membrane receptor is receptor tyrosine kinase (RTK), illustrated in Figure 5.5. This is an example of a receptor that joins with another molecule. A kinase is an enzyme that transfers a phosphate group from an ATP molecule to another protein. In the case of RTK, these receptor molecules work in pairs; a single RTK is incapable of receiving and transferring a signal. Instead, two ligands come and bind to two separate RTK molecules. Once this occurs, the receptors bind together to form a dimer—two identical molecules bound together. The dimer is then activated by phosphorylation involving six ATP molecules at the tyrosine amino acid sites. The six dimer phosphate sites each bind to a relay protein, activating it with the phosphate, which triggers a structural change. The multiple activated relay proteins trigger signal transduction pathways, thus beginning the relay of the message into the cell, leading to cellular responses.

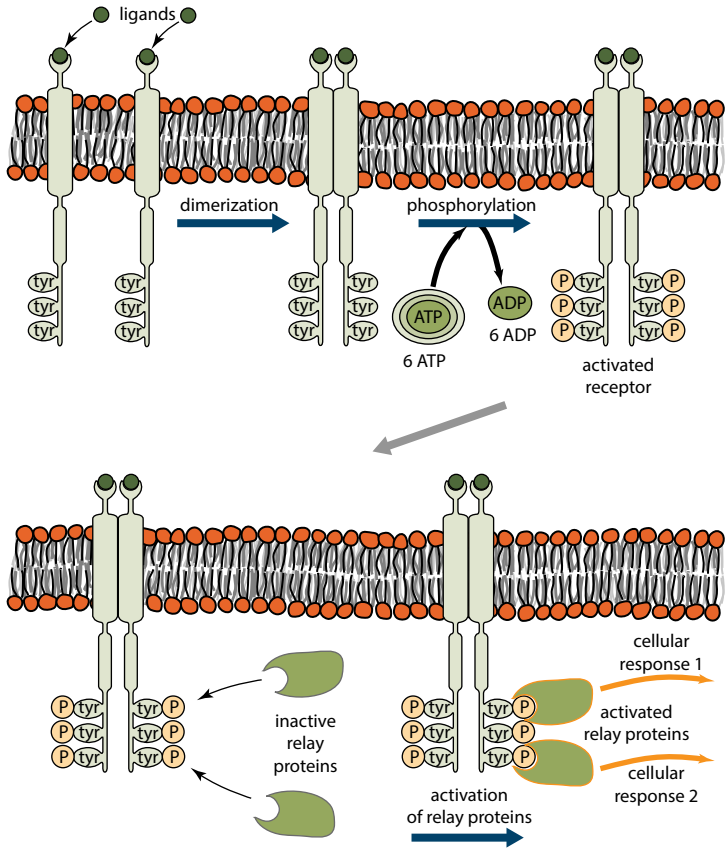


FIGURE 5.5. *Receptor tyrosine kinase.*

A simpler type of membrane receptor is the ligand-gated ion channel, illustrated in Figure 5.6. These proteins look much like a channel or pore, but they have a receptor site. When inactive, they are closed, preventing molecules from moving across the membrane. Activation occurs when a ligand binds to the receptor, causing the protein to change its shape and open. This allows ions to enter the cell, leading to a cellular response. A well-understood example of this is found between nerve cells at what is known as the *synaptic cleft*.

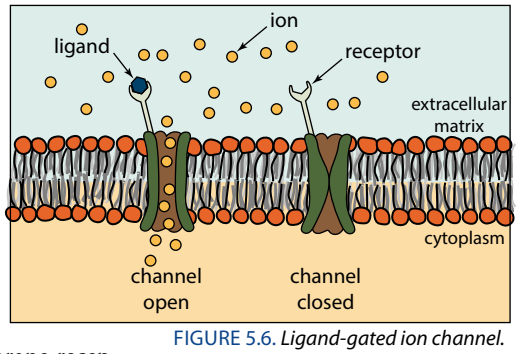


FIGURE 5.6. *Ligand-gated ion channel.*

We have considered three different transmembrane receptors. Next, let's look at an example of an intracellular receptor. As the name suggests, intracellular receptors are found inside the cell rather than embedded in the cell membrane. In some cases, they may even be inside the nucleus of the cell. For this reason, the ligand must be able to pass through the cell membrane to gain access to the receptor. These ligands are usually steroids—lipid-based signaling molecules. As depicted in Figure 5.7, once inside the cell, the ligand binds directly to the intracellular protein receptor, forming a ligand-receptor complex. This complex then goes on to cause a cellular response. One of

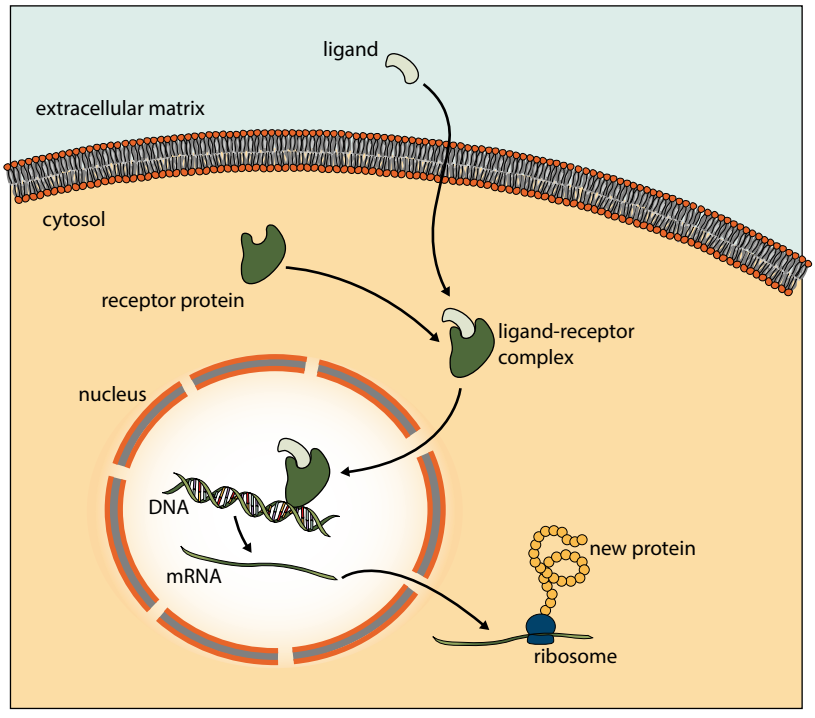


FIGURE 5.7. *Intracellular receptor.*

the most common functions is for it to move into the nucleus and operate as a *transcription factor*. We address these in more detail in a later chapter, but, in brief, they tell the cell to "turn on" a gene so that it is used to make a protein.

5.2.2 *Signal Transduction*

After a signal is received by a cell receptor, the signal is passed into the cell through a multi-step pathway. These steps are usually the result of proteins interacting with other proteins in some way. The message gets passed from one molecule to another, like a relay baton being passed from runner to runner in a relay race. The relay proteins are usually held in place by other structural proteins called scaffolding proteins, illustrated in Figure 5.8. This results in a controlled and efficient relay of information into the cell. Another benefit of a multi-step pathway is that it enables the signal to be passed deeply into the cell. This happens by each protein activating multiple proteins after it, as discussed next.

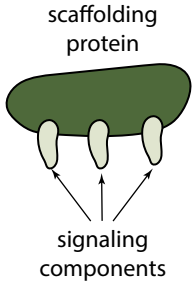


FIGURE 5.8. *Scaffolding protein.*

There are two main mechanisms cells use to relay a message into the cell. The first of these is a *phosphorylation cascade*. We have already described kinases, proteins that transfer a phosphate group from one molecule to another molecule. The addition of a phosphate group (phosphorylation) can activate subsequent proteins—thousands of them—thereby passing the message further into the cell, as depicted in Figure 5.9. Cells also contain protein *phosphatases*. These are enzymes that catalyze the removal of phosphates (*dephosphorylation*). If phos-

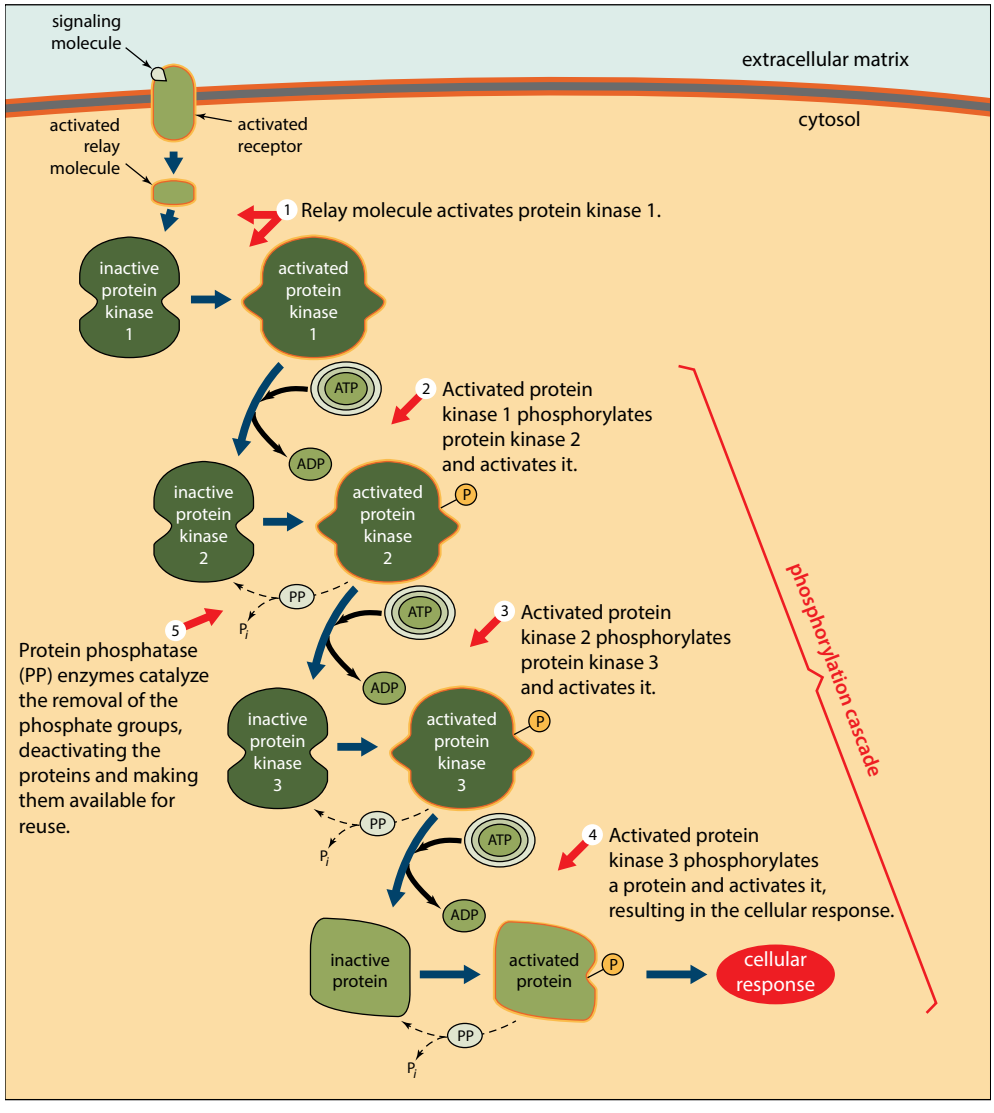


FIGURE 5.9. *Phosphorylation cascade. Although only three proteins are shown in the cascade, such cascades can involve thousands of proteins.*

phorylation activates a molecule, dephosphorylation inactivates the molecule. In this way, the cell regulates the transfer of the signal in the cell.

A second common mechanism for signal transduction is for a cell to utilize a *second messenger*. Second messengers are usually small, non-protein molecules that can diffuse quickly throughout the cell. The two most common second-messenger molecules are cyclic AMP (cAMP) and the calcium ion (Ca^{2+}). The second messenger studied by Earl Sutherland was cAMP. The enzyme adenylyl cyclase transforms a molecule of ATP into cAMP in response to a signal. Many cAMP molecules are made and diffused throughout the cell, resulting in a cellular response, such as the breakdown of glycogen observed by Sutherland.

Examples of Ca^{2+} as a second messenger are found in many cellular processes, such as muscle contractions, exocytosis, and cell division. Normally, the levels of Ca^{2+} inside a cell are relatively low. When a signal is received, channels open to enable Ca^{2+} to flood the interior of the cell. The Ca^{2+} ions may come from outside the cell in the extracellular matrix, or they may be released by the endoplasmic reticulum where they are stored. The rapid increase in cytosolic Ca^{2+} results in various cellular responses.

5.2.3 *Cellular Response and Regulation*

There are many different responses a cell may undergo after receiving a signal and these depend on many different factors. For example, a common signal found in mammals is the hormone epinephrine, also known as adrenaline. But different organs and tissues respond differently to this hormone, depending on the type of cell receptor present on the various tissues and organs. Heart cells have a special receptor called a beta-adrenergic receptor. When epinephrine binds to this receptor, it is activated and tells the heart muscle to contract more strongly and more frequently. In contrast, when epinephrine binds to alpha receptors on liver cells, it signals the cell to begin breaking down glycogen, thereby releasing glucose into the blood stream. These different cell responses are the result of the same ligand binding to different receptors, illustrating how different receptor proteins play an important role in the specific response of each cell type. The response initiated by the cell continues so long as the ligand remains bound to the receptor. Once the ligand is released, the signal is terminated.

5.3 The Cell Cycle

For an organism to grow and develop requires the formation of new cells. We know from our study of cell theory that all cells come from preexisting cells. Thus, one of the responses a cell may exhibit in response to a signal is to undergo cell division, resulting in one cell becoming two. In this section, we examine the details of the *cell cycle*—alternations between periods of growth and periods of division. As you read and study Section 5.3, take time to pause and marvel at the wonderful processes being described. Their elegance and rationality speak strongly to us of the Creator's wisdom!

5.3.1 *Overview of the Cell Cycle*

Just as humans live according to a day-night cycle, alternating sleep and wakefulness, the cell has a cycle of its own. The cell cycle is depicted in Figure 5.10. The cell cycle can be divided into two parts: *interphase* and *mitosis*, shown in the outer circle of the figure. Most of a cell's life is spent in interphase. This is when the cell carries out its normal functions, such as cellular respiration, photosynthesis, and gene expression, along

with more specific functions, such as contraction in a muscle cell or conduction of an electrical impulse by a nerve cell.

There are three stages of interphase, shown in the inner circle of Figure 5.10. The first stage is the *first gap phase*, or G_1. During G_1, the cell carries out its normal functions. G_1 is followed by the *synthesis phase*, or S. At this point, the cell makes a complete copy of its entire genome, a process called *DNA replication*. The third phase is the *second gap phase*, or G_2. During G_2, the cell prepares to divide. After interphase, the cell enters the process of mitosis, when the duplicated chromosomes are separated so that each new cell will receive a complete copy of the genome. Mitosis ends with an elongated cell that is ready to divide. The ending of mitosis occurs in parallel with the onset of *cytokinesis*, the division of the cytoplasm into two separate cells. The result is two daughter cells, each genetically identical to the original cell.

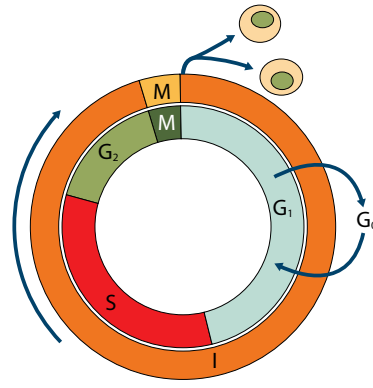


FIGURE 5.10. *Overview of the cell cycle. Outer circle: I = interphase, M = mitosis. Inner circle: G_1 = gap phase, S = synthesis, G_2 = second gap phase, M = mitosis. Some cells stop dividing and enter a separate phase called G_0.*

5.3.2 *DNA Structure and Bonding Details*

In the next section, we discuss how DNA is copied, a process that is part of the cell cycle. But first, we need to add some detail to our description in Chapter 2 of the structure of DNA.

Let's begin by reviewing what we already know about the structure of DNA. DNA is a polymer composed of nucleotide monomers. Each nucleotide consists of three chemical components:

- one of these four nitrogenous bases—adenine (A), thymine (T), cytosine (C), and guanine (G)
- one deoxyribose sugar molecule
- one phosphate group

Ball-and-stick models of the DNA molecule and of one of the nucleotides are shown in Figures 2.35 and 2.36. Figure 5.11 shows a schematic representation of the DNA double helix, with the bases of the nucleotides in pairs running down the middle. Figure 5.12 (and also Figure 2.37) shows how the four nucleotides pair with one another. As mentioned in Chapter 2, a nucleotide with an adenine (A) base always pairs with a nucleotide containing thymine (T), and a nucleotide with cytosine (C) always pairs with a nucleotide containing guanine (G).

Note that the adenine and guanine bases each have two rings bonded together, while thymine and cytosine each have one ring. A DNA base with two rings is called a *purine*; a base with one ring is called a *pyrimidine*. Each DNA base pair includes one of each. The A–T base pair is connected by two hydrogen bonds, indicated by the dashed lines in Figure 5.12. The G—C base pair is connected by three hydrogen bonds. In this way, two long DNA strands are connected to each other by hundreds of thousands of hydrogen bonds.

At the ends of the sugar-phosphate backbones in Figures 5.11 and 5.12 are the labels 3′ and 5′. These designations (pronounced "three prime" and "five prime") are used to indicate

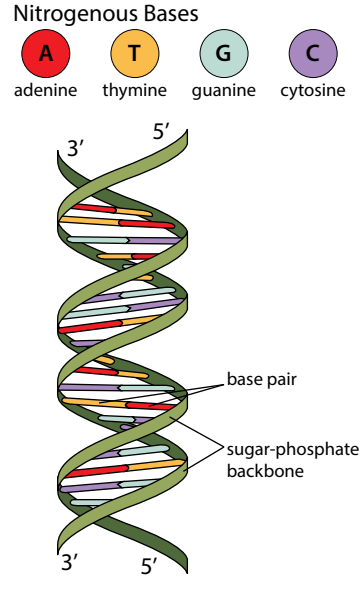


FIGURE 5.11. *DNA schematic structure.*

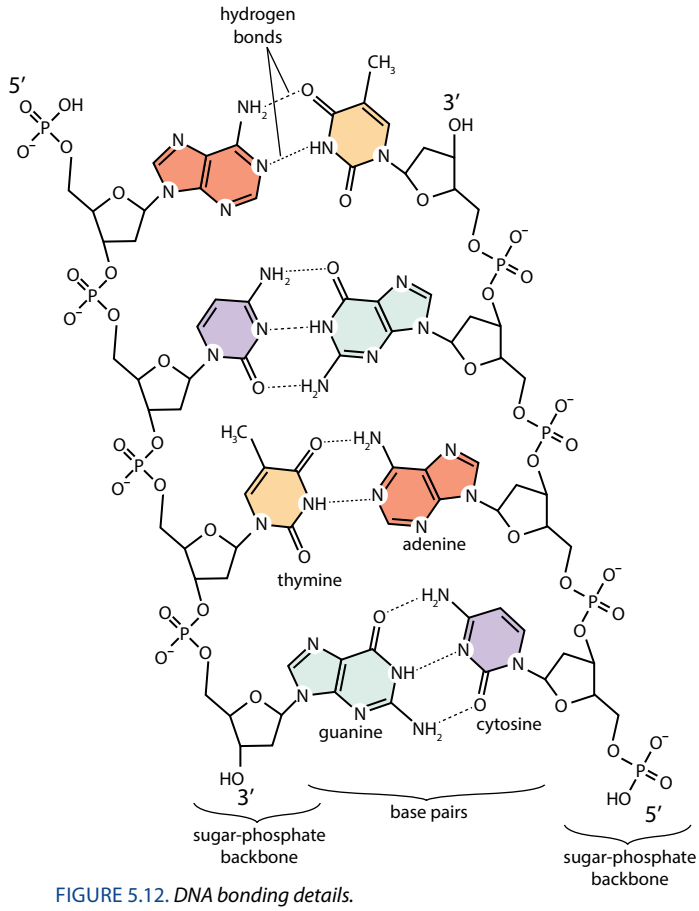


FIGURE 5.12. *DNA bonding details.*

the directionality of the strand. At the 3′ position is a hydroxyl group, at the 5′ position is a phosphate group, and the ends where these occur are in opposite locations on the two strands. The labels 3′ and 5′ are based on the positions of the carbon atoms in the deoxyribose sugar ring. The standard numbering for these carbon atoms is shown in Figure 5.13.

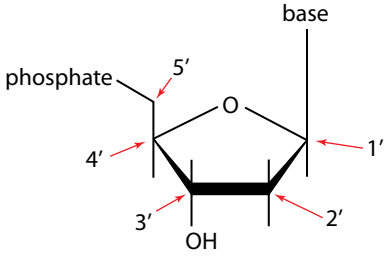


FIGURE 5.13. *Numbering convention for the carbons on the deoxyribose sugar of DNA. Beginning with the carbon attached to the base, we number them 1′ (one-prime), 2′ (two-prime) and so on. The 3′ OH (hydroxyl) group and the 5′ phosphate group are the attachment points for other nucleotides, and thus the terms 3′ and 5′ indicate the direction of the DNA strand.*

Figure 5.12 shows how the 5′ phosphate group is bonded at the location of the 3′ hydroxyl group of the next nucleotide to build a DNA strand. This is called a *photodiester bond*. On the left side, you see the 5′ phosphate at the top end. The complementary strand on the right side runs in the opposite direction. Here, the 3′ hydroxyl group is at the top, while the 5′ phosphate group is at the bottom. Because these complementary DNA strands run in opposite directions, they are said to be *antiparallel*. We use the designations 5′ and 3′ to indicate the direction of the DNA strand, just as we distinguish the east end of a street in a town from the west end.

To illustrate how complementary strands are sequenced, take as an example a strand of DNA with the following sequence:

5′-ATCAGTCAGGGTCACTT-3′

To determine the sequence of the complementary strand, first notice that the given sequence is written in the 5′⟶3′ direction by default. However, due to the antiparallel nature of double stranded DNA, the complementary strand runs in the opposite direction, 3′⟶5′. Also recall the base pairing rules: G pairs with C, while A pairs with T. Using this information, our complementary strand is as follows:

3′-TAGTCAGTCCCAGTGAA-5′

5.3.3 *Synthesis (DNA Replication)*

We return now to the cell cycle and a description of the synthesis phase, during which DNA replication takes place. Before a cell can divide, it must make a full copy of its DNA so that each daughter cell receives a full set of genetic information.

When James Watson and Francis Crick first described the structure of DNA in 1953, they recognized that the structure of DNA provides a mechanism for DNA to be replicated. Since guanine always pairs with cytosine and adenine always pairs with thymine, to make a copy of the DNA the cell separates the two strands, which when separated are called *templates*, lines up free nucleotides to pair with the nucleotides in each template (A bases to T bases, G to C, etc.), and then links the new nucleotides together covalently to form a new strand complementary to the template. Together, a new strand and the template to which it is joined form a new double-stranded DNA molecule.

As we go through the description, you will note that a number of enzymes are involved. Keep track of them all—your ability to describe DNA replication depends on identifying the different enzymes and their roles in the process.

Polymerization Reaction

We begin with the reaction that joins nucleotides together as a new DNA strand is assembled, shown in Figure 5.14.

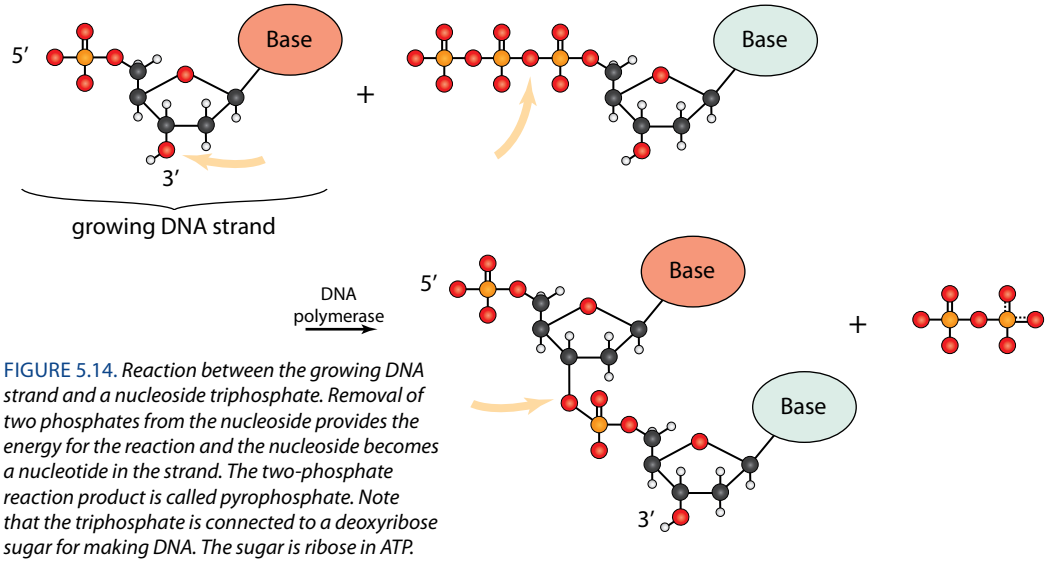


FIGURE 5.14. *Reaction between the growing DNA strand and a nucleoside triphosphate. Removal of two phosphates from the nucleoside provides the energy for the reaction and the nucleoside becomes a nucleotide in the strand. The two-phosphate reaction product is called pyrophosphate. Note that the triphosphate is connected to a deoxyribose sugar for making DNA. The sugar is ribose in ATP.*

Before the reaction, the building blocks that become the nucleotide monomers in the strand are free *nucleoside triphosphates*, molecules containing three phosphate groups, a deoxyribose sugar, and one of the four nucleobases (guanine, cytosine, adenine, and thymine). In the same way that ATP provides energy in the cell by the hydrolysis of one phosphate to become ADP, the energy to accomplish the reaction in Figure 5.14 is provided by the hydrolysis of two of the phosphates from the nucleoside, which creates a nucleotide joined to the growing DNA strand by a phosphodiester bond. This reaction is catalyzed by an enzyme called *DNA polymerase*, about which more in a moment. As the figure indicates, the nucleotide joins the growing strand at the 3′ end, taking the place of the hydroxyl group there. For the description that follows, it is important to note that assembly of a new DNA strand always takes place by joining new nucleotides to the 3′ end of the molecule. This means that the sequence of reactions involved in making new DNA strands always proceeds in the 5′⟶3′ direction along the new strand.

There are numerous types of DNA polymerase. Some of these, denoted Pol I, Pol II, etc., catalyze replication in prokaryotes. Others, denoted Pol α, Pol δ, Pol ε, etc., catalyze replication in eukaryotes. And even within one of these contexts (prokaryote or eukaryote replication), different polymerases (α, δ, ε, etc.) function in various roles in the replication process. It is not necessary for our purposes in this text to distinguish between the many types of polymerase involved, so we refer to them all simply as DNA polymerase.

Getting Replication Started

The process of initiating DNA replication is outlined in the schematic diagram shown in Figure 5.15 for (A) bacteria and (B) eukaryotes. Bacteria use a single *replication origin* to

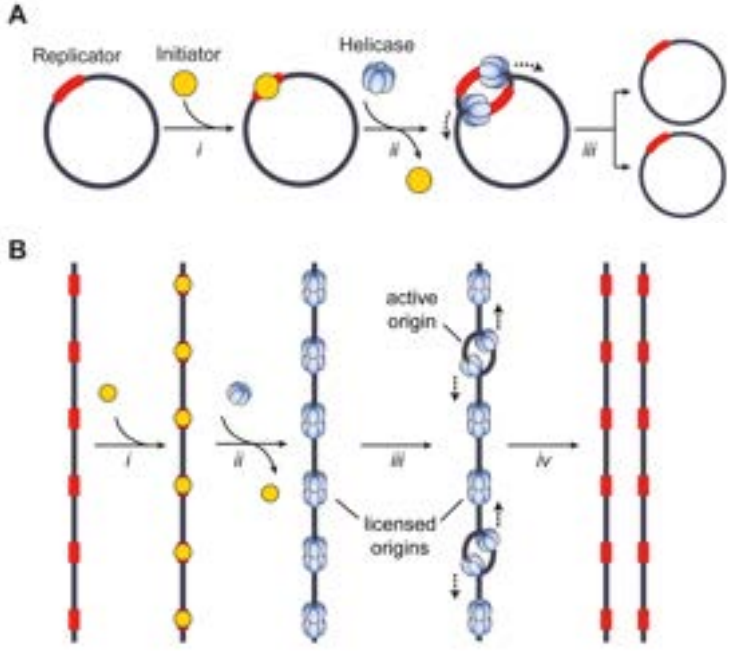


FIGURE 5.15. *Initiating DNA replication in (A) bacteria and (B) eukaryotes. Bacteria use a single replication origin to replicate their circular DNA. Many origins are used to replicate linear eukaryote DNA. Initiator proteins (yellow) bind to the origins (red) and recruit helicase enzymes that bind to the origins in pairs. The single pair of helicases for bacterial DNA is activated; for eukaryotic DNA, only some of the helicase pairs are activated. The activated helicase enzymes separate the DNA strands to form replication "bubbles" and replication begins.*

replicate their circular DNA. Many replication origins (hundreds or thousands) are used to replicate linear eukaryote DNA. Replication origins are sections of DNA, a few hundred base pairs long, that are rich in A–T base pairs. These pairs only use two hydrogen bonds instead of the three bonds in G–C pairs, so the strands are easier to pull apart in these regions. The initiator proteins (yellow) are attracted to the replication origins (red), bind there, and then recruit *helicase* enzymes that bind to the origins in pairs. These sites are now said to be "licensed." The single helicase pair for bacterial DNA is activated; for eukaryotic DNA, only some of the helicase pairs are activated. The activated helicase enzymes separate the DNA strands to form a replication "bubble" and replication begins. As suggested by the figure and described further below, the helicase enzymes remain in the bubble after the initial separation—they are the enzymes that continue to separate the DNA strands, working in both directions, outward from the replication origin.

Overview

DNA replication is so complex it is mind-boggling—another fingerprint of an infinite Creator whose works are surpassingly elegant and wonderful. This complexity will be more easily approached if we ease into it with an overview before getting into some of the finer details. Figure 5.16 shows a schematic of how the process proceeds at a single replication origin. If you first glance through all four sections of this diagram, you can get an idea of what happens in the overall process: origin, attachment of *RNA primers*, assembly of new DNA strands (shown in green) inside the bubble that are complementary to the strands of the parent DNA (shown in blue), and enlargement of the bubble with continued assembly of new DNA until we end up with two separate, complete, and identical sections of DNA. Backing up, let's add in a few more high-level details. After the parent DNA strands are separated to form the bubble, *primase* enzymes synthesize short sections of RNA called *RNA primers* and attach them to both parent strands. As mentioned above, the two strands of parent DNA are called templates. The RNA primers, shown as wiggly blue-green segments, are the starting points for assembling the two new strands, one for each template. The primers have 3′ and 5′ ends, just as DNA strands have, and the assembly of the new strands begins at the 3′ end of a primer and proceeds from there in the 5′⟶3′ direction along the new strand.

We still have our two helicase enzymes (not shown in Figure 5.16) separating the template strands on both ends of the bubble and enlarging

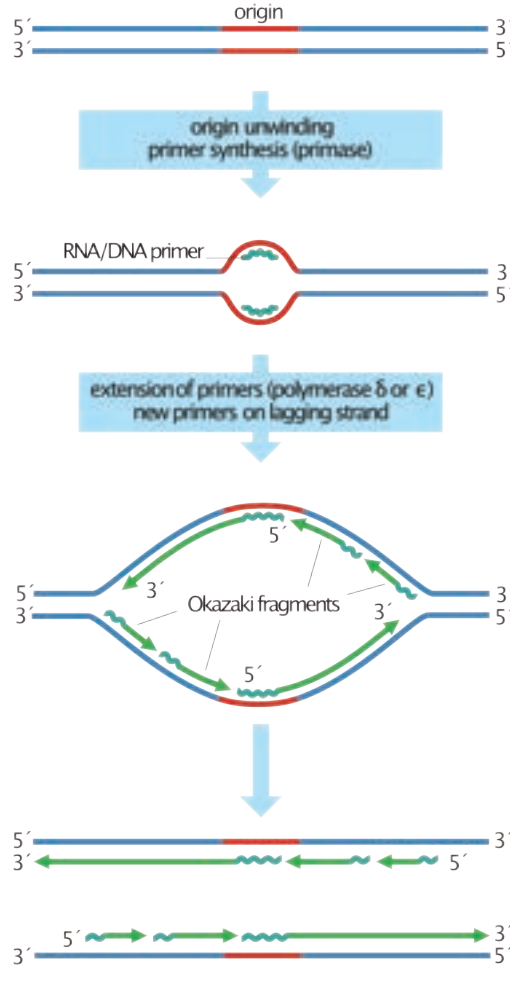


FIGURE 5.16. *DNA replication overview.*

it. The location of the helicase, where the parent strands are being separated, is called the *replication fork*. The green arrows represent the new strands of DNA being assembled by the DNA polymerase enzymes. The two long, new strands pointing toward the replication forks are called *leading strands*, and their assembly is continuous, following along behind the helicase toward the replication fork as the helicase opens the bubble more and more. Notice the shorter green arrows point away from the replication forks. These are called *lagging strands*. As the bubble enlarges new sections of template DNA are opening up between the replication fork and the origin locations where the two initial RNA primers are. For the lagging strands, new RNA primers are planted over and over on the template as the bubble enlarges. New DNA polymerase enzymes show up at every one of these primers, latch onto its 3′ end, and begin assembling a DNA strand, continuing until they run into an RNA primer in front of them. So the assembly of the lagging strands is non-continuous. The short strands made from all these additional RNA primers are also called *Okazaki fragments*.

From our discussion of the reaction creating the phosphodiester bonds, it should be clear why the leading strand can be continuously assembled, following along behind the helicase enzyme, while the lagging strands must be assembled piecemeal. New nucleotides can only be connected to the 3′ end of the strand. In the case of the leading strand, the 3′ end is the one nearest to the helicase, so as the helicase moves along unzipping the parent DNA, the DNA polymerase just goes along behind it adding one new nucleotide after another. But the new regions of template DNA opening behind where the initial RNA primers are cannot be continuously assembled this way because the new nucleotides would have to be joined to the 5′ end of the strand, which cannot happen. So, the enzymes manage this by planting new RNA primers as more template DNA opens up and bringing in DNA polymerase enzymes as needed to work from each one in the 5′⟶3′ direction.

Enzyme and Procedural Details

Figure 5.17 gives us our most detailed look at the DNA replication process, where we zoom in on the replication fork at one end of one bubble. (There is far more detail beyond this, but those details are beyond this course.) Every part of this process makes use of enzymes. Look toward the right in the figure and locate the helicase in the replication fork. The helicase is shown attached to the side where the leading strand is being assembled. This is the case in eukaryotes; in prokaryotes, the helicase attaches to the other side. To the right of the helicase is another enzyme, *topoisomerase*. The function of topoisomerase can be understood by considering the separation of the DNA strands as analogous to separating the strands in the middle of a rope. It is not so hard at first, but to open the separation more and more puts increasing amounts of stress on the remaining twisted section of rope. Relieving this tension is the function

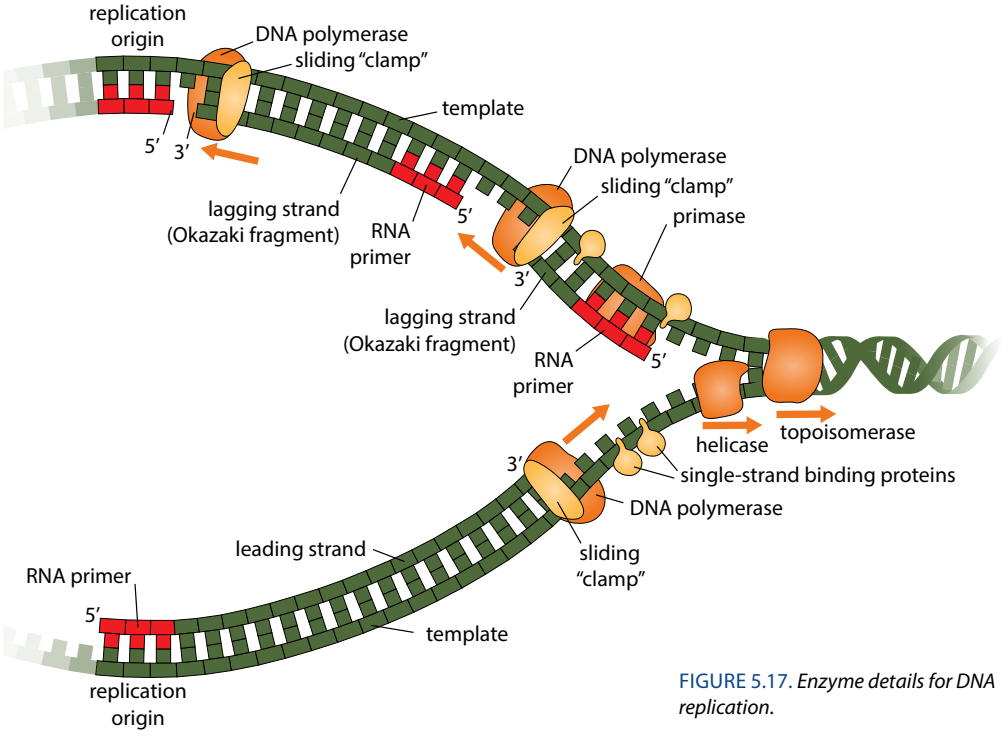


FIGURE 5.17. *Enzyme details for DNA replication.*

of the topoisomerase. Topoisomerase actually goes along ahead of the helicase, breaking and rejoining one or both of the parent DNA strands as necessary to relieve the increasing tension caused by the helicase unwinding the strands. This is sort of like an electric cord that has become over-twisted near where it is plugged in: one can unplug it, take the excess twist out and plug the cord back in. After topoisomerase rejoins the strands, the DNA molecule contains the same atoms as before, but the amount of twist in the molecule has changed. It has become an isomer of what it was before and its topology has changed, hence the name of the enzyme.

Moving to the left, we have the helicase unzipping (aka unwinding) the DNA by breaking the hydrogen bonds that hold the base pairs of the two strands together. This process results in the replication fork, the place where the parent DNA molecule becomes two single-stranded DNA molecules with their nitrogenous bases exposed. Small *single-strand binding proteins* come in to help hold the two DNA strands apart while DNA replication takes place. The assembly process is performed by molecules of DNA polymerase. As described above, the process of assembling complimentary strands for the two templates is different. The polymerase brings in a nucleoside triphosphate (base, sugar, and three phosphates) that is complementary to the corresponding base on the template. Then the polymerase catalyzes its attachment to the 3′ end of the growing chain (and the removal of two of the phosphate groups). Because the nucleotide can only be added to the 3′ end of the growing DNA molecule, DNA replication always proceeds in the 5′⟶3′ di-