BIOL 1550: MICROBIOLOGY (2025)



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Licensing

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CHAPTER OVERVIEW

1: Introduction to Microbiology

From boiling thermal hot springs to deep beneath the Antarctic ice, microorganisms can be found almost everywhere on earth in great quantities. Microorganisms (or microbes, as they are also called) are small organisms. Most are so small that they cannot be seen without a microscope.

Most microorganisms are harmless to humans and, in fact, many are helpful. They play fundamental roles in ecosystems everywhere on earth, forming the backbone of many food webs. People use them to make biofuels, medicines, and even foods. Without microbes, there would be no bread, cheese, or beer. Our bodies are filled with microbes, and our skin alone is home to trillions of them[1]. Some of them we can't live without; others cause diseases that can make us sick or even kill us.



Figure **1.1**: A veterinarian gets ready to clean a sea turtle covered in oil following the Deepwater Horizon oil spill in the Gulf of Mexico in 2010. After the spill, the population of a naturally occurring oil-eating marine bacterium called *Alcanivorax borkumensis* skyrocketed, helping to get rid of the oil. Scientists are working on ways to genetically engineer this bacterium to be more efficient in cleaning up future spills. (credit: modification of work by NOAA's National Ocean Service)

Although much more is known today about microbial life than ever before, the vast majority of this invisible world remains unexplored. Microbiologists continue to identify new ways that microbes benefit and threaten humans.

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Footnotes

1. 1 J. Hulcr et al. "A Jungle in There: Bacteria in Belly Buttons are Highly Diverse, but Predictable." *PLoS ONE* 7 no. 11 (2012): e47712. doi:10.1371/journal.pone.0047712.

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1.1: Introduction to Microbiology

Learning Objectives

- State three harmful effects and four beneficial effects associated with the activities of microorganisms.
- Define microbiota and microbiome.
- Briefly describe two different beneficial things the human microbiome does for the normal function of our body.
- State several diseases associated with a change in our "normal" microbiota.
- List and recognize a description of the each of the 5 basic groups of microbes.

Microorganisms are the dominant life forms on earth, are found in almost every conceivable environment, and are essential to sustaining life on this planet. There are five basic groups of microorganisms:

- **Bacteria** are typically unicellular, microscopic, prokaryotic organisms that reproduce by binary fission.
- **Fungi (**yeasts and molds) are typically unicellular, microscopic, eukaryotic fungi that reproduce asexually by budding. Molds are typically filamentous, eukaryotic fungi that reproduce by producing asexual reproductive spores.
- **Viruses** are typically submicroscopic, acellular infectious particles that can only replicate inside a living host cell. The vast majority of viruses possess either DNA or RNA, but not both.
- **Protozoa** are typically unicellular, microscopic, eukaryotic organisms that lack a cell wall.
- **Algae** are typically eukaryotic microorganisms that carry out photosynthesis.

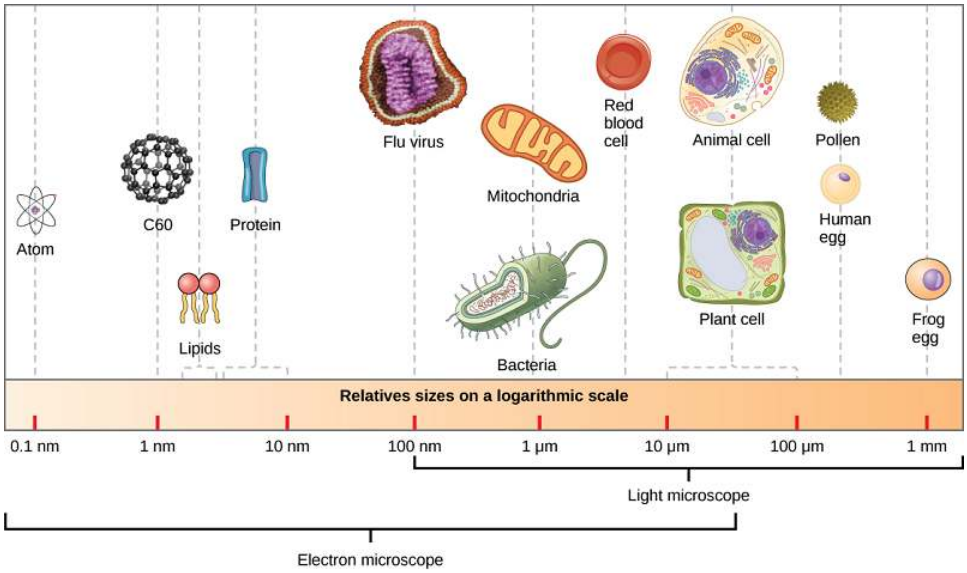


Figure **1.1.1**: The size of a virus is very small relative to the size of cells and organelles.

In this course we will be looking at various fundamental concepts of microbiology, with particular emphasis on their relationships to human health. The overall goal is to better understand the total picture of infectious diseases in terms of host-infectious agent interaction. We will look at various groups of microbes and learn what they might do to establish infection and harm the body, we will look at the body to see the ways in which it defends itself against these microbes, and we will learn what can be done to help the body in its defense efforts.

The Big Picture of Infectious Diseases

One of the most important things in microbiology is learining the "Big Picture of Infectious Diseases," which is the biological basis of host parasite interaction. There are four interlocking parts to this big picture:

A. **The microbe's side of the story - why some microbes have more potential to be harmful:** The overwhelming majority of microbes are harmless to humans and, in fact, many are beneficial, being key players in the recycling of nutrients in nature. We will look at the major groups of microbes, learn what they are composed of chemically and structurally, and see

how how they carry out their metabolism and reproduce. We will learn of a variety of factors some microbes may possess that play a role in increasing their ability to cause disease. Also we will learn how, through mutation, genetic recombination, and natural selection, microbes may adapt to resist our control attempts.

B. **The body's side of the story - ways in which the body is able to defend itself naturally against infectious disease agents:** Here will learn about the phenomenal defenses the body has available to defend itself against infectious disease agents, as well as altered body cells such as cancer cells and infected cells. The body is able to do this through the innate immune system and the adaptive immune system. Innate immune defenses are those you are born with and include anatomical barriers, mechanical removal, cytokines, pattern-recognition receptors, phagocytosis, inflammation, the complement pathways, and fever. The adaptive immune defenses are those you develop throughout your life and include antibody production and cell-mediated immunity.

C. **Ways in which we can artificially help the body defend itself by removing the microbes or enhancing body defenses:** We will learn how we can artificially help ourselves to avoid or reduce the risk of infection. Also we will learn ways in which we are able to artificially remove microbes from the body and its environment using agents such as antiseptics, disinfectants, physical agents such as heat and cold, antimicrobial chemotherapeutic chemicals, and antibiotics. Finally we will learn ways we are currently able to - or potentially in the future will be able to - improve or restore the body's immune responses through such techniques as immunization, adoptive immunotherapy, or immune modulation.

D. **Relationship between the Human Microbiome and Human Health:** The complex mutually beneficial symbiotic relationship between humans and their natural microbes is critical to good health. It is now recognized that the millions of genes associated with the normal flora or microbiota of the human body -especially in the intestinal tract - aid in the digestion of many foods, the regulation of multiple host metabolic pathways, and the regulation the body's immune defenses.

Benefits of Microbial Activity

Most people tend to think of microorganisms as harmful because of their roles in causing infectious diseases in humans and other animals, and agricultural loss as a result of infectious diseases of plants and the spoilage of food. The fact is, however, the vast majority of microorganisms are not harmful but rather beneficial. Without them there would be no life on earth. Therefore, we will start this course by looking at a few of the many benefits from microbial activity on this planet.

1. **Food production:** Many food products employ microorganisms in their production. These include the microbial fermentation processes used to produce yogurt, buttermilk, cheeses, alcoholic beverages, leavened breads, sauerkraut, pickles, and kimchi.
2. **Energy production and cleaning up the environment**: Methane, or natural gas, is a product of *methanogenic microorganisms*. Many aquatic microbes capture light energy and store it in molecules used as food then used by other organisms. Animal wastes, domestic refuse, biomass, and grain can be converted to biofuels such as ethanol and methane by microorganisms. In addition, through a process called bioremediation , some pollutants such pesticides, solvents, and oil spills can be cleaned up with the aid of microbes.
3. **Sustaining agriculture**: Through their roles in recycling nitrogen, carbon, and sulfur, microorganism are able to convert these essential elements into forms that can be used by plants in their growth. They are also essential in enabling ruminant animals such as cows and sheep to digest cellulose from the grasses they eat.
4. **Production of useful natural gene products or products from bioengineering**. Examples include specific enzymes, antibiotics, vaccines, and medications such as human insulin, interferons, and growth hormones.
5. **The human microbiota and microbiome**: Where we be without microorganisms? While the typical human body contains an estimated 37 trillion human cells, it also contains over 100 trillion bacteria and other microbes. The human body has 3 times as many bacterial cells as it does human cells! It is estimated the the mass of the human microbiota is 2.5 pounds.

The complex mutually beneficial symbiotic relationship between humans and their natural microbes is critical to good health. It is now recognized that the millions of genes associated with the microbiota of the human body -especially in the intestinal tract - aid in the digestion of many foods, the regulation of multiple host metabolic pathways, and the regulation the body's immune defenses. These collective microbes and their genes are referred to as the human microbiome. There are currently an estimated 5,000,000 - 10,000,000 genes from over 1000 species that constitute the human microbiome compared to the approximately 20,000 - 23,000 genes that make up the human genome. There are approximately 300 non-human genes in the human body for every human gene.



Key Concepts and Summary

1. Microorganisms are typically too small to be seen with the naked eye.
2. Bacteria, fungi, viruses, protozoa, and algae are the major groups of microorganisms.
3. The vast majority of microorganisms are not harmful but rather beneficial.
4. Microbiota refers to all of the microorganisms that live in a particular environment.
5. A microbiome is the entire collection of genes found in all of the microbes associated with a particular host.
6. The microbiome of the human body - especially in the intestinal tract - aid in the digestion of many foods, the regulation of multiple host metabolic pathways, and the regulation the body's immune defenses.

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1.2: Introduction to Microbiology

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1.3: Cellular Organization - Prokaryotic and Eukaryotic Cells

1.02: Cellular Organization - Prokaryotic and Eukaryotic Cells

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1.4: Classification - The Three Domain System

1.03: Classification - The Three Domain System

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1.5: What Our Ancestors Knew

Learning Objectives

- Describe how our ancestors improved food with the use of invisible microbes
- Describe how the causes of sickness and disease were explained in ancient times, prior to the invention of the microscope
- Describe key historical events associated with the birth of microbiology

Most people today, even those who know very little about microbiology, are familiar with the concept of microbes, or “germs,” and their role in human health. Schoolchildren learn about bacteria, viruses, and other microorganisms, and many even view specimens under a microscope. But a few hundred years ago, before the invention of the microscope, the existence of many types of microbes was impossible to prove. By definition, **microorganisms**, or **microbes**, are very small organisms; many types of microbes are too small to see without a microscope, although some parasites and fungi are visible to the naked eye.

Humans have been living with—and using—microorganisms for much longer than they have been able to see them. Historical evidence suggests that humans have had some notion of microbial life since prehistoric times and have used that knowledge to develop foods as well as prevent and treat disease. In this section, we will explore some of the historical applications of microbiology as well as the early beginnings of microbiology as a science.

Fermented Foods and Beverages

People across the world have enjoyed fermented foods and beverages like beer, wine, bread, yogurt, cheese, and pickled vegetables for all of recorded history. Discoveries from several archeological sites suggest that even prehistoric people took advantage of fermentation to preserve and enhance the taste of food. Archaeologists studying pottery jars from a Neolithic village in China found that people were making a fermented beverage from rice, honey, and fruit as early as 7000 BC.[2]

Production of these foods and beverages requires microbial fermentation, a process that uses bacteria, mold, or yeast to convert sugars (carbohydrates) to alcohol, gases, and organic acids (Figure **1.5.2**). While it is likely that people first learned about fermentation by accident—perhaps by drinking old milk that had curdled or old grape juice that had fermented—they later learned to harness the power of fermentation to make products like bread, cheese, and wine.

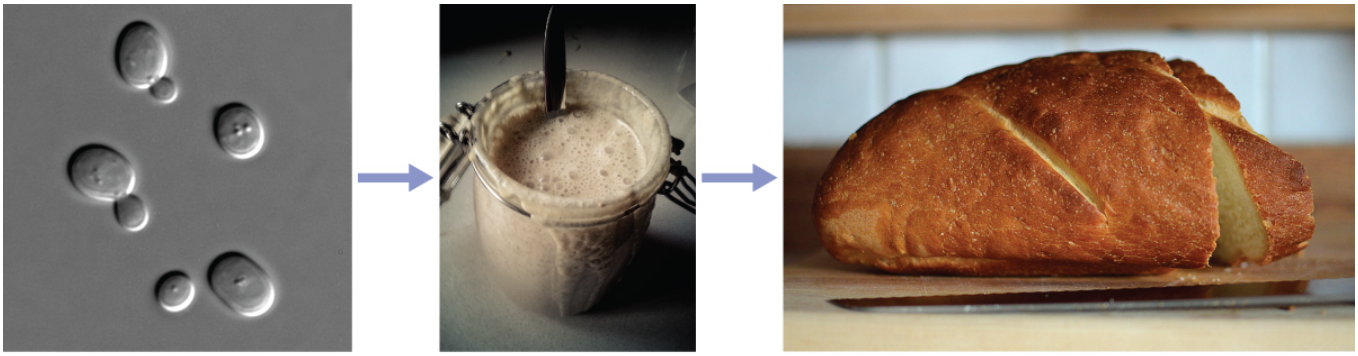


Figure **1.5.2**: A microscopic view of *Saccharomyces cerevisiae*, the yeast responsible for making bread rise (left). Yeast is a microorganism. Its cells metabolize the carbohydrates in flour (middle) and produce carbon dioxide, which causes the bread to rise (right). (credit middle: modification of work by Janus Sandsgaard; credit right: modification of work by “MDreibelbis”/Flickr)

The Iceman Treateth

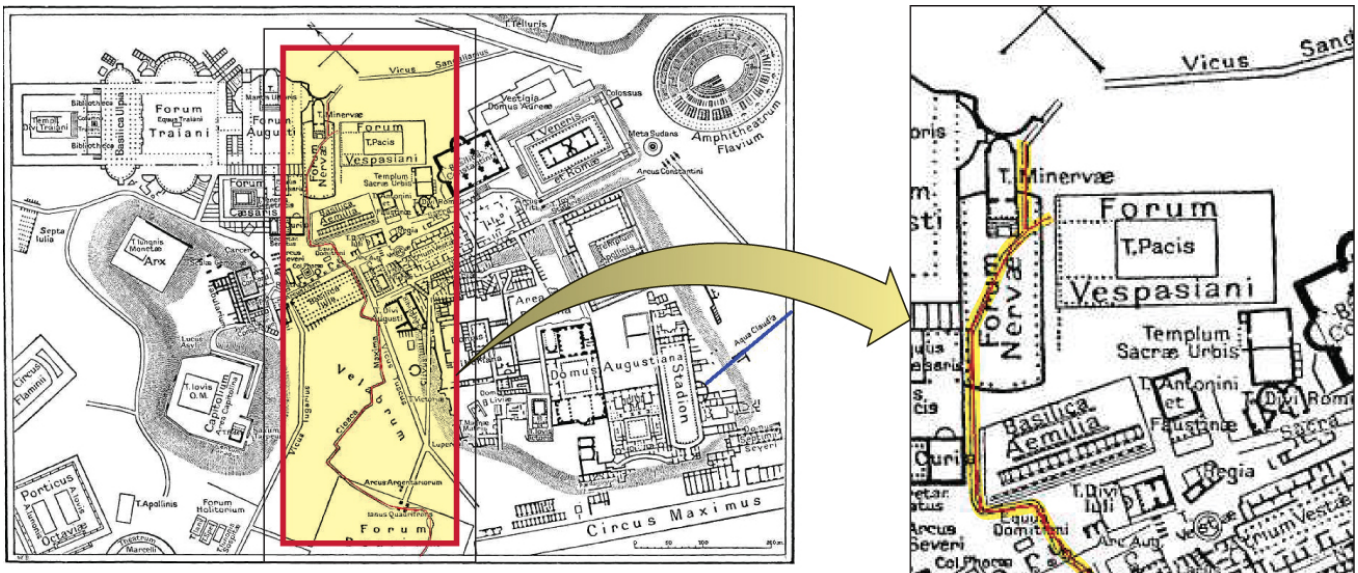
Prehistoric humans had a very limited understanding of the causes of disease, and various cultures developed different beliefs and explanations. While many believed that illness was punishment for angering the gods or was simply the result of fate, archaeological evidence suggests that prehistoric people attempted to treat illnesses and infections. One example of this is Ötzi the Iceman, a 5300-year-old mummy found frozen in the ice of the Ötzal Alps on the Austrian-Italian border in 1991. Because Ötzi was

so well preserved by the ice, researchers discovered that he was infected with the eggs of the parasite *Trichuris trichiura*, which may have caused him to have abdominal pain and anemia. Researchers also found evidence of *Borrelia burgdorferi*, a bacterium that causes Lyme disease.[3] Some researchers think Ötzi may have been trying to treat his infections with the woody fruit of the *Piptoporus betulinus* fungus, which was discovered tied to his belongings.[4] This fungus has both laxative and antibiotic properties. Ötzi was also covered in tattoos that were made by cutting incisions into his skin, filling them with herbs, and then burning the herbs.[5] There is speculation that this may have been another attempt to treat his health ailments.

Early Notions of Disease, Contagion, and Containment

Several ancient civilizations appear to have had some understanding that disease could be transmitted by things they could not see. This is especially evident in historical attempts to contain the spread of disease. For example, the Bible refers to the practice of quarantining people with leprosy and other diseases, suggesting that people understood that diseases could be communicable. Ironically, while leprosy is communicable, it is also a disease that progresses slowly. This means that people were likely quarantined after they had already spread the disease to others.

The ancient Greeks attributed disease to bad air, *mal'aria*, which they called "miasmatic odors." They developed hygiene practices that built on this idea. The Romans also believed in the miasma hypothesis and created a complex sanitation infrastructure to deal with sewage. In Rome, they built aqueducts, which brought fresh water into the city, and a giant sewer, the *Cloaca Maxima*, which carried waste away and into the river Tiber (Figure **1.5.3**). Some researchers believe that this infrastructure helped protect the Romans from epidemics of waterborne illnesses.





(b)

Figure **1.5.3**: (a) The *Cloaca Maxima*, or "Greatest Sewer" (shown in red), ran through ancient Rome. It was an engineering marvel that carried waste away from the city and into the river Tiber. (b) These ancient latrines emptied into the *Cloaca Maxima*.

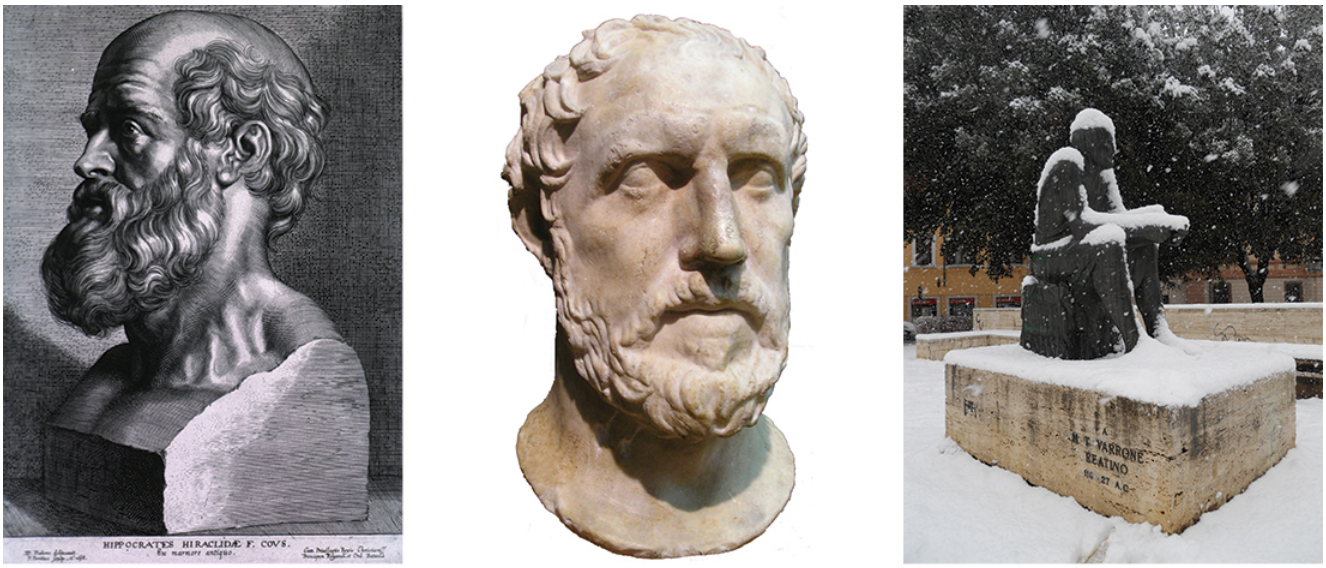
Even before the invention of the microscope, some doctors, philosophers, and scientists made great strides in understanding the invisible forces—what we now know as microbes—that can cause infection, disease, and death.

The Greek physician Hippocrates (460–370 BC) is considered the "father of Western medicine" (Figure **1.5.4*a***). Unlike many of his ancestors and contemporaries, he dismissed the idea that disease was caused by supernatural forces. Instead, he posited that diseases had natural causes from within patients or their environments. Hippocrates and his heirs are believed to have written the *Hippocratic Corpus*, a collection of texts that make up some of the oldest surviving medical books.[6] Hippocrates is also often credited as the author of the Hippocratic Oath, taken by new physicians to pledge their dedication to diagnosing and treating patients without causing harm.

While Hippocrates is considered the father of Western medicine, the Greek philosopher and historian Thucydides (460–395 BC) is considered the father of scientific history because he advocated for evidence-based analysis of cause-and-effect reasoning (Figure

1.5.4b). Among his most important contributions are his observations regarding the Athenian plague that killed one-third of the population of Athens between 430 and 410 BC. Having survived the epidemic himself, Thucydides made the important observation that survivors did not get re-infected with the disease, even when taking care of actively sick people.[7] This observation shows an early understanding of the concept of immunity.

Marcus Terentius Varro (116–27 BC) was a prolific Roman writer who was one of the first people to propose the concept that things we cannot see (what we now call microorganisms) can cause disease (Figure 1.5.4c). In *Res Rusticae* (*On Farming*), published in 36 BC, he said that "precautions must also be taken in neighborhood swamps . . . because certain minute creatures [*animalia minuta*] grow there which cannot be seen by the eye, which float in the air and enter the body through the mouth and nose and there cause serious diseases."[8]



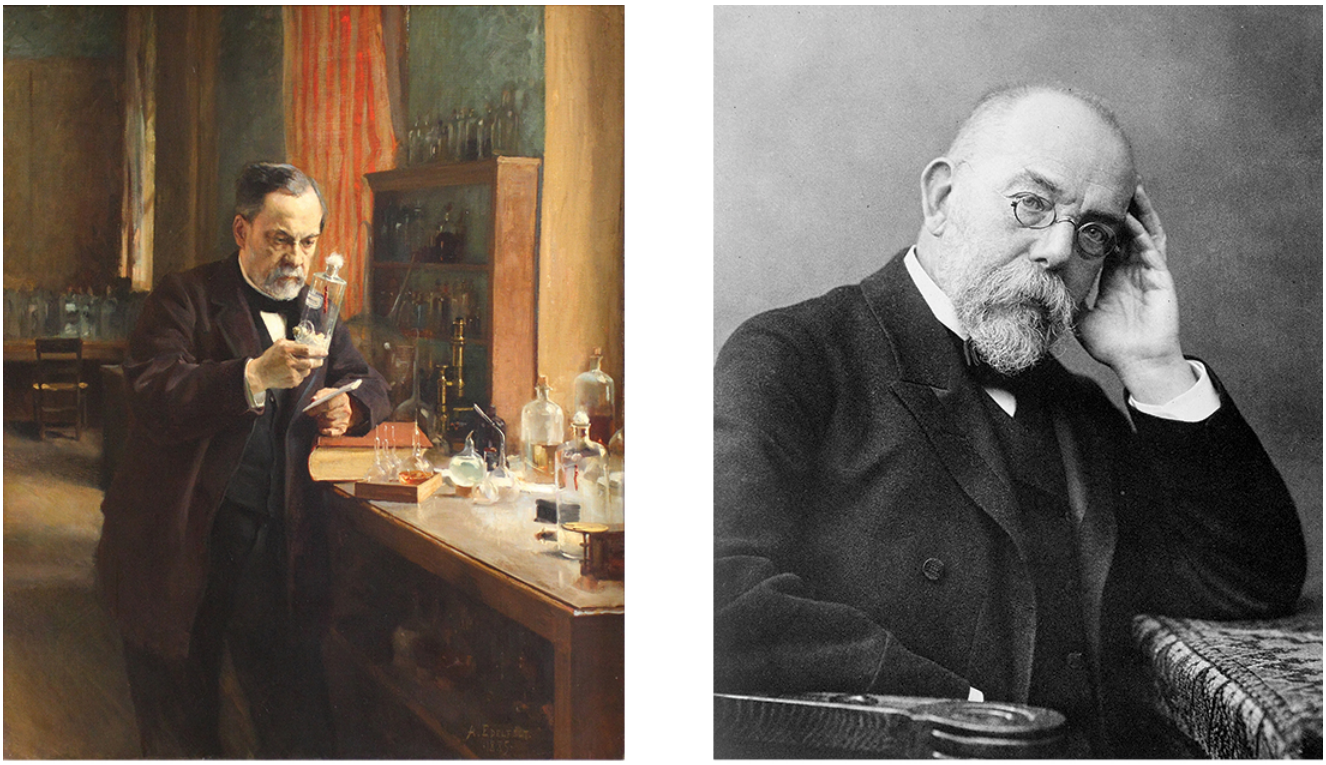
(a) (b) (c)

Figure 1.5.4: (a) Hippocrates, the "father of Western medicine," believed that diseases had natural, not supernatural, causes. (b) The historian Thucydides observed that survivors of the Athenian plague were subsequently immune to the infection. (c) Marcus Terentius Varro proposed that disease could be caused by "certain minute creatures . . . which cannot be seen by the eye." (credit c: modification of work by Alessandro Antonelli)

The Birth of Microbiology

While the ancients may have suspected the existence of invisible "minute creatures," it wasn't until the invention of the microscope that their existence was definitively confirmed. While it is unclear who exactly invented the microscope, a Dutch cloth merchant named Antonie van Leeuwenhoek (1632–1723) was the first to develop a lens powerful enough to view microbes. In 1675, using a simple but powerful microscope, Leeuwenhoek was able to observe single-celled organisms, which he described as "animalcules" or "wee little beasties," swimming in a drop of rain water. From his drawings of these little organisms, we now know he was looking at bacteria and protists. (We will explore Leeuwenhoek's contributions to microscopy further in Chapter 2: How We See the Invisible World.)

Nearly 200 years after van Leeuwenhoek got his first glimpse of microbes, the "Golden Age of Microbiology" spawned a host of new discoveries between 1857 and 1914. Two famous microbiologists, Louis Pasteur and Robert Koch, were especially active in advancing our understanding of the unseen world of microbes (Figure 1.5.5). Pasteur, a French chemist, showed that individual microbial strains had unique properties and demonstrated that fermentation is caused by microorganisms. He also invented pasteurization, a process used to kill microorganisms responsible for spoilage, and developed vaccines for the treatment of diseases, including rabies, in animals and humans. Koch, a German physician, was the first to demonstrate the connection between a single, isolated microbe and a known human disease. For example, he discovered the bacteria that cause anthrax (*Bacillus anthracis*), cholera (*Vibrio cholera*), and tuberculosis (*Mycobacterium tuberculosis*).[9] We will discuss these famous microbiologists, and others, in later chapters.



(a) (b)

Figure **1.5.5**: (a) Louis Pasteur (1822–1895) is credited with numerous innovations that advanced the fields of microbiology and immunology. (b) Robert Koch (1843–1910) identified the specific microbes that cause anthrax, cholera, and tuberculosis.

As microbiology has developed, it has allowed the broader discipline of biology to grow and flourish in previously unimagined ways. Much of what we know about human cells comes from our understanding of microbes, and many of the tools we use today to study cells and their genetics derive from work with microbes.

Microbiology Toolbox

Because individual microbes are generally too small to be seen with the naked eye, the science of microbiology is dependent on technology that can artificially enhance the capacity of our natural senses of perception. Early microbiologists like Pasteur and Koch had fewer tools at their disposal than are found in modern laboratories, making their discoveries and innovations that much more impressive. Later chapters of this text will explore many applications of technology in depth, but for now, here is a brief overview of some of the fundamental tools of the microbiology lab.

- Microscopes produce magnified images of microorganisms, human cells and tissues, and many other types of specimens too small to be observed with the naked eye.
- Stains and dyes are used to add color to microbes so they can be better observed under a microscope. Some dyes can be used on living microbes, whereas others require that the specimens be fixed with chemicals or heat before staining. Some stains only work on certain types of microbes because of differences in their cellular chemical composition.
- Growth media are used to grow microorganisms in a lab setting. Some media are liquids; others are more solid or gel-like. A growth medium provides nutrients, including water, various salts, a source of carbon (like glucose), and a source of nitrogen and amino acids (like yeast extract) so microorganisms can grow and reproduce. Ingredients in a growth medium can be modified to grow unique types of microorganisms.
- A Petri dish is a flat-lidded dish that is typically 10–11 centimeters (cm) in diameter and 1–1.5 cm high. Petri dishes made out of either plastic or glass are used to hold growth media (Figure **1.5.6**).
- Test tubes are cylindrical plastic or glass tubes with rounded bottoms and open tops. They can be used to grow microbes in broth, or semisolid or solid growth media.

- A Bunsen burner is a metal apparatus that creates a flame that can be used to sterilize pieces of equipment. A rubber tube carries gas (fuel) to the burner. In many labs, Bunsen burners are being phased out in favor of infrared microincinerators, which serve a similar purpose without the safety risks of an open flame.
- An inoculation loop is a handheld tool that ends in a small wire loop (Figure **1.5.6**). The loop can be used to streak microorganisms on agar in a Petri dish or to transfer them from one test tube to another. Before each use, the inoculation loop must be sterilized so cultures do not become contaminated.

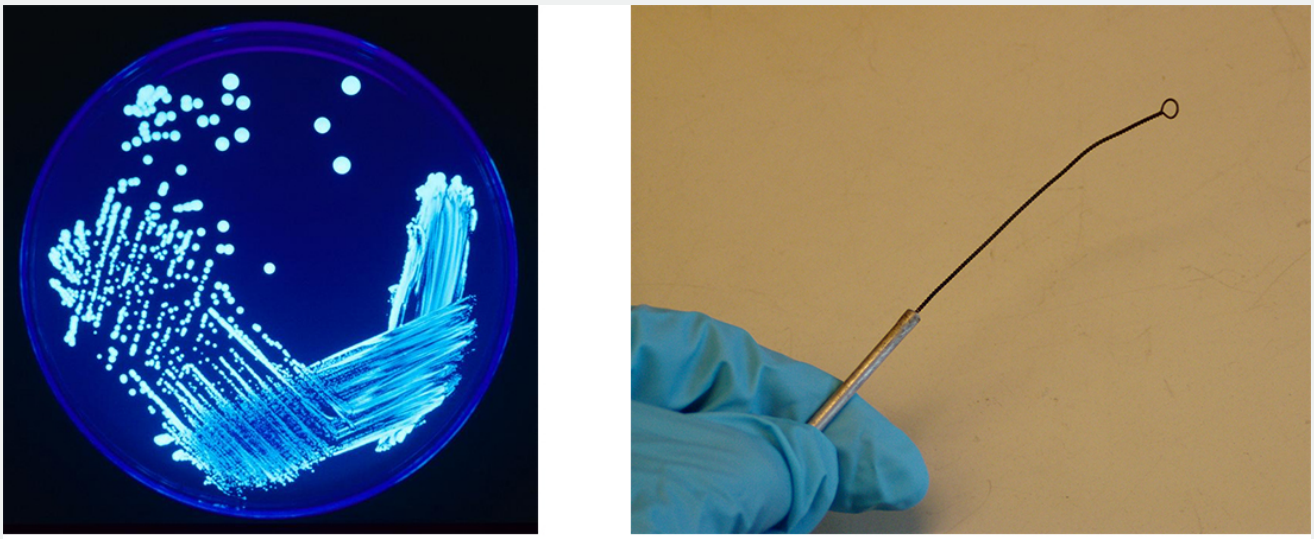


Figure **1.5.6**: (a) This Petri dish filled with agar has been streaked with *Legionella*, the bacterium responsible for causing Legionnaire's disease. (b) An inoculation loop like this one can be used to streak bacteria on agar in a Petri dish. (credit a: modification of work by Centers for Disease Control and Prevention; credit b: modification of work by Jeffrey M. Vinocur)

Key Concepts and Summary

- Microorganisms (or microbes) are living organisms that are generally too small to be seen without a microscope.
- Throughout history, humans have used microbes to make fermented foods such as beer, bread, cheese, and wine.
- Long before the invention of the microscope, some people theorized that infection and disease were spread by living things that were too small to be seen. They also correctly intuited certain principles regarding the spread of disease and immunity.
- Antonie van Leeuwenhoek, using a microscope, was the first to actually describe observations of bacteria, in 1675.
- During the Golden Age of Microbiology (1857–1914), microbiologists, including Louis Pasteur and Robert Koch, discovered many new connections between the fields of microbiology and medicine.

Footnotes

1. 1 Rebecca Buxton. "Examination of Gram Stains of Spinal Fluid—Bacterial Meningitis." *American Society for Microbiology*. 2007. www.microbelibrary.org/librar...ial-meningitis
2. 2 P.E. McGovern et al. "Fermented Beverages of Pre- and Proto-Historic China." *Proceedings of the National Academy of Sciences of the United States of America* 1 no. 51 (2004):17593–17598. doi:10.1073/pnas.0407921102.
3. 3 A. Keller et al. "New Insights into the Tyrolean Iceman's Origin and Phenotype as Inferred by Whole-Genome Sequencing."*Nature Communications*, 3 (2012): 698. doi:10.1038/ncomms1701.
4. 4 L. Capasso. "5300 Years Ago, the Ice Man Used Natural Laxatives and Antibiotics." *The Lancet*, 352 (1998) 9143: 1864. doi: 10.1016/s0140-6736(05)79939-6.
5. 5 L. Capasso, L. "5300 Years Ago, the Ice Man Used Natural Laxatives and Antibiotics." *The Lancet*, 352 no. 9143 (1998): 1864. doi: 10.1016/s0140-6736(05)79939-6.
6. 6 G. Pappas et al. "Insights Into Infectious Disease in the Era of Hippocrates." *International Journal of Infectious Diseases* 12 (2008) 4:347–350. doi: http://dx.doi.org/10.1016/j.ijid.2007.11.003.

7. 7 Thucydides. *The History of the Peloponnesian War. The Second Book*. 431 BC. Translated by Richard Crawley. http://classics.mit.edu/Thucydides/p....2.second.html.
8. 8 Plinio Prioreschi. *A History of Medicine: Roman Medicine*. Lewiston, NY: Edwin Mellen Press, 1998: p. 215.
9. 9 S.M. Blevins and M.S. Bronze. “Robert Koch and the ‘Golden Age’ of Bacteriology.” *International Journal of Infectious Diseases*. 14 no. 9 (2010): e744-e751. doi:10.1016/j.ijid.2009.12.003.

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1.6: A Systematic Approach

Learning Objecctives

- Describe how microorganisms are classified and distinguished as unique species
- Compare historical and current systems of taxonomy used to classify microorganisms

Once microbes became visible to humans with the help of microscopes, scientists began to realize their enormous diversity. Microorganisms vary in all sorts of ways, including their size, their appearance, and their rates of reproduction. To study this incredibly diverse new array of organisms, researchers needed a way to systematically organize them.

The Science of Taxonomy

Taxonomy is the classification, description, identification, and naming of living organisms. Classification is the practice of organizing organisms into different groups based on their shared characteristics. The most famous early taxonomist was a Swedish botanist, zoologist, and physician named Carolus Linnaeus (1701–1778). In 1735, Linnaeus published *Systema Naturae*, an 11-page booklet in which he proposed the Linnaean taxonomy, a system of categorizing and naming organisms using a standard format so scientists could discuss organisms using consistent terminology. He continued to revise and add to the book, which grew into multiple volumes (Figure **1.6.1**).



Figure **1.6.1**: Swedish botanist, zoologist, and physician Carolus Linnaeus developed a new system for categorizing plants and animals. In this 1853 portrait by Hendrik Hollander, Linnaeus is holding a twinflower, named *Linnaea borealis* in his honor.

In his taxonomy, Linnaeus divided the natural world into three kingdoms: animal, plant, and mineral (the mineral kingdom was later abandoned). Within the animal and plant kingdoms, he grouped organisms using a hierarchy of increasingly specific levels and sublevels based on their similarities. The names of the levels in Linnaeus's original taxonomy were kingdom, class, order, family, genus (plural: genera), and species. Species was, and continues to be, the most specific and basic taxonomic unit.



Evolving Trees of Life (Phylogenies)

With advances in technology, other scientists gradually made refinements to the Linnaean system and eventually created new systems for classifying organisms. In the 1800s, there was a growing interest in developing taxonomies that took into account the evolutionary relationships, or **phylogenies**, of all different species of organisms on earth. One way to depict these relationships is via a diagram called a phylogenetic tree (or tree of life). In these diagrams, groups of organisms are arranged by how closely related they are thought to be. In early phylogenetic trees, the relatedness of organisms was inferred by their visible similarities, such as the presence or absence of hair or the number of limbs. Now, the analysis is more complicated. Today, phylogenic analyses include genetic, biochemical, and embryological comparisons, as will be discussed later in this chapter.

Linnaeus's tree of life contained just two main branches for all living things: the animal and plant kingdoms. In 1866, Ernst Haeckel, a German biologist, philosopher, and physician, proposed another kingdom, Protista, for unicellular organisms (Figure **1.6.2**). He later proposed a fourth kingdom, Monera, for unicellular organisms whose cells lack nuclei, like bacteria.

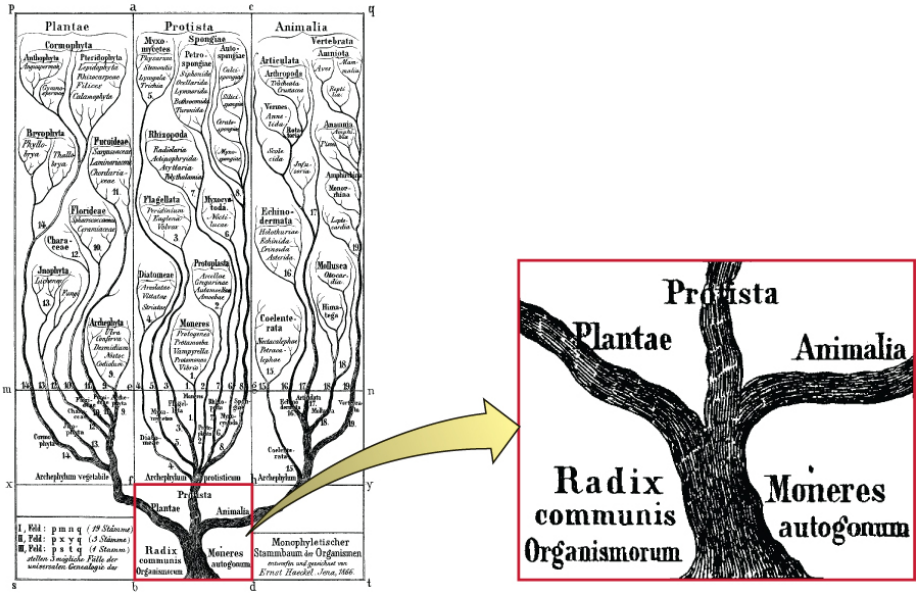


Figure **1.6.2**: Ernst Haeckel's rendering of the tree of life, from his 1866 book *General Morphology of Organisms*, contained three kingdoms: Plantae, Protista, and Animalia. He later added a fourth kingdom, Monera, for unicellular organisms lacking a nucleus.

Nearly 100 years later, in 1969, American ecologist Robert Whittaker (1920–1980) proposed adding another kingdom—Fungi—in his tree of life. Whittaker's tree also contained a level of categorization above the kingdom level—the empire or superkingdom level—to distinguish between organisms that have membrane-bound nuclei in their cells (**eukaryotes**) and those that do not (**prokaryotes**). Empire Prokaryota contained just the Kingdom Monera. The Empire Eukaryota contained the other four kingdoms: Fungi, Protista, Plantae, and Animalia. Whittaker's five-kingdom tree was considered the standard phylogeny for many years.

Figure **1.6.3** shows how the tree of life has changed over time. Note that viruses are not found in any of these trees. That is because they are not made up of cells and thus it is difficult to determine where they would fit into a tree of life.

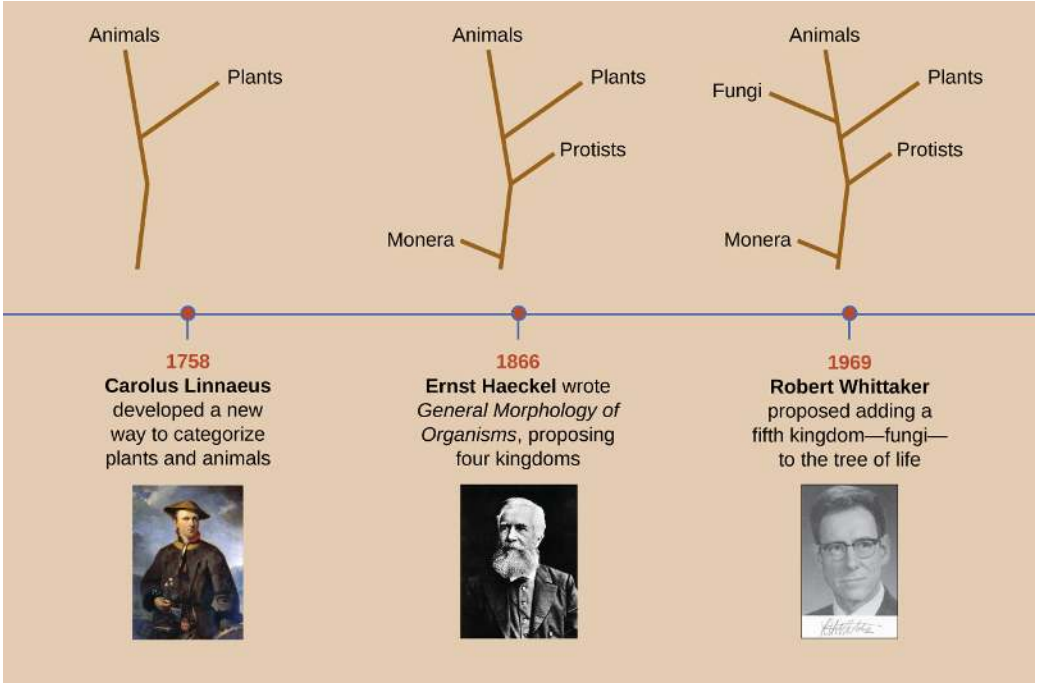


Figure **1.6.3**: This timeline shows how the shape of the tree of life has changed over the centuries. Even today, the taxonomy of living organisms is continually being reevaluated and refined with advances in technology.

The Role of Genetics in Modern Taxonomy

Haeckel's and Whittaker's trees presented hypotheses about the phylogeny of different organisms based on readily observable characteristics. But the advent of molecular genetics in the late 20th century revealed other ways to organize phylogenetic trees. Genetic methods allow for a standardized way to compare all living organisms without relying on observable characteristics that can often be subjective. Modern taxonomy relies heavily on comparing the nucleic acids (deoxyribonucleic acid [DNA] or ribonucleic acid [RNA]) or proteins from different organisms. The more similar the nucleic acids and proteins are between two organisms, the more closely related they are considered to be.

In the 1970s, American microbiologist Carl Woese discovered what appeared to be a "living record" of the evolution of organisms. He and his collaborator George Fox created a genetics-based tree of life based on similarities and differences they observed in the small subunit ribosomal RNA (rRNA) of different organisms. In the process, they discovered that a certain type of bacteria, called archaebacteria (now known simply as archaea), were significantly different from other bacteria and eukaryotes in terms of the sequence of small subunit rRNA. To accommodate this difference, they created a tree with three Domains above the level of Kingdom: Archaea, Bacteria, and Eukarya (Figure **1.6.4**). Genetic analysis of the small subunit rRNA suggests archaea, bacteria, and eukaryotes all evolved from a common ancestral cell type. The tree is skewed to show a closer evolutionary relationship between Archaea and Eukarya than they have to Bacteria.

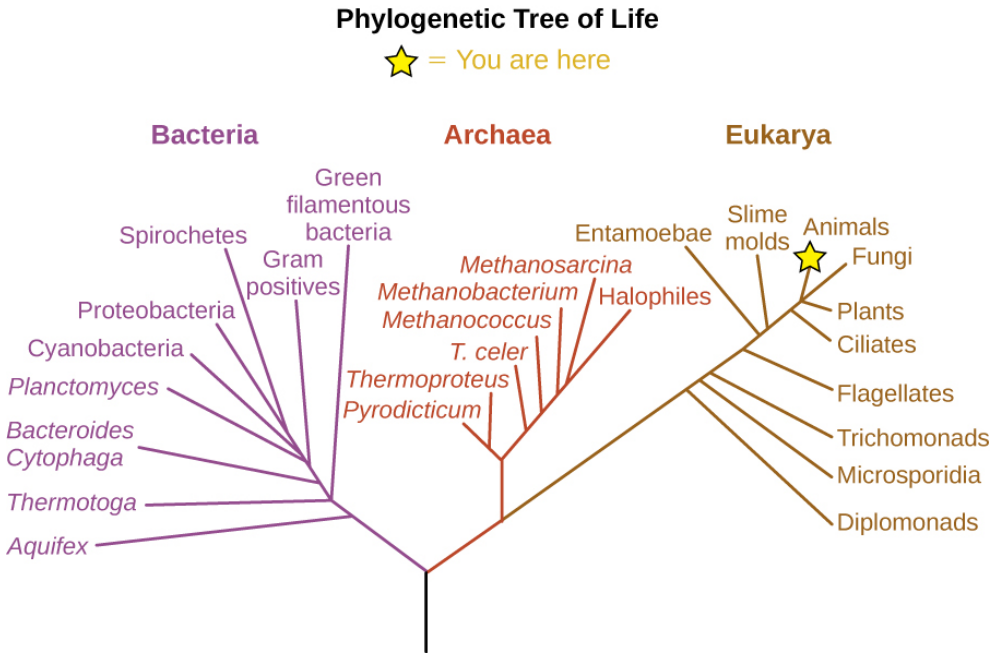


Figure **1.6.4**: Scientists continue to use analysis of RNA, DNA, and proteins to determine how organisms are related. One interesting, and complicating, discovery is that of horizontal gene transfer—when a gene of one species is absorbed into another organism's genome. Horizontal gene transfer is especially common in microorganisms and can make it difficult to determine how organisms are evolutionarily related. Consequently, some scientists now think in terms of "webs of life" rather than "trees of life."

Naming Microbes

In developing his taxonomy, Linnaeus used a system of **binomial nomenclature**, a two-word naming system for identifying organisms by genus and species. For example, modern humans are in the genus *Homo* and have the species name *sapiens*, so their scientific name in binomial nomenclature is *Homo sapiens*. In binomial nomenclature, the genus part of the name is always capitalized; it is followed by the species name, which is not capitalized. Both names are italicized.

Taxonomic names in the 18th through 20th centuries were typically derived from Latin, since that was the common language used by scientists when taxonomic systems were first created. Today, newly discovered organisms can be given names derived from Latin, Greek, or English. Sometimes these names reflect some distinctive trait of the organism; in other cases, microorganisms are named after the scientists who discovered them. The archaeon *Haloquadratum walsbyi* is an example of both of these naming schemes. The genus, *Haloquadratum*, describes the microorganism's saltwater habitat (*halo* is derived from the Greek word for "salt") as well as the arrangement of its square cells, which are arranged in square clusters of four cells (*quadratum* is Latin for "foursquare"). The species, *walsbyi*, is named after Anthony Edward Walsby, the microbiologist who discovered *Haloquadratum walsbyi* in in 1980. While it might seem easier to give an organism a common descriptive name—like a red-headed woodpecker—we can imagine how that could become problematic. What happens when another species of woodpecker with red head coloring is discovered? The systematic nomenclature scientists use eliminates this potential problem by assigning each organism a single, unique two-word name that is recognized by scientists all over the world.

In this text, we will typically abbreviate an organism's genus and species after its first mention. The abbreviated form is simply the first initial of the genus, followed by a period and the full name of the species. For example, the bacterium *Escherichia coli* is shortened to *E. coli* in its abbreviated form. You will encounter this same convention in other scientific texts as well.

Bergey's Manuals

Whether in a tree or a web, microbes can be difficult to identify and classify. Without easily observable macroscopic features like feathers, feet, or fur, scientists must capture, grow, and devise ways to study their biochemical properties to differentiate and classify microbes. Despite these hurdles, a group of microbiologists created and updated a set of manuals for identifying and classifying microorganisms. First published in 1923 and since updated many times, *Bergey's Manual of Determinative Bacteriology*

and *Bergey's Manual of Systematic Bacteriology* are the standard references for identifying and classifying different prokaryotes. (Appendix D of this textbook is partly based on Bergey's manuals; it shows how the organisms that appear in this textbook are classified.) Because so many bacteria look identical, methods based on nonvisual characteristics must be used to identify them. For example, biochemical tests can be used to identify chemicals unique to certain species. Likewise, serological tests can be used to identify specific antibodies that will react against the proteins found in certain species. Ultimately, DNA and rRNA sequencing can be used both for identifying a particular bacterial species and for classifying newly discovered species.

Same Name, Different Strain

Within one species of microorganism, there can be several subtypes called strains. While different strains may be nearly identical genetically, they can have very different attributes. The bacterium*Escherichia coli* is infamous for causing food poisoning and traveler's diarrhea. However, there are actually many different strains of *E. coli*, and they vary in their ability to cause disease.

One pathogenic (disease-causing) *E. coli* strain that you may have heard of is *E. coli* O157:H7. In humans, infection from *E. coli* O157:H7 can cause abdominal cramps and diarrhea. Infection usually originates from contaminated water or food, particularly raw vegetables and undercooked meat. In the 1990s, there were several large outbreaks of *E. coli* O157:H7 thought to have originated in undercooked hamburgers.

While *E. coli* O157:H7 and some other strains have given *E. coli* a bad name, most *E. coli* strains do not cause disease. In fact, some can be helpful. Different strains of *E. coli* found naturally in our gut help us digest our food, provide us with some needed chemicals, and fight against pathogenic microbes.

Key Concepts and Summary

- Carolus Linnaeus developed a taxonomic system for categorizing organisms into related groups.
- Binomial nomenclature assigns organisms Latinized scientific names with a genus and species designation.
- A phylogenetic tree is a way of showing how different organisms are thought to be related to one another from an evolutionary standpoint.
- The first phylogenetic tree contained kingdoms for plants and animals; Ernst Haeckel proposed adding a kingdom for protists.
- Robert Whittaker's tree contained five kingdoms: Animalia, Plantae, Protista, Fungi, and Monera.
- Carl Woese used small subunit ribosomal RNA to create a phylogenetic tree that groups organisms into three domains based on their genetic similarity.
- Bergey's manuals of determinative and systemic bacteriology are the standard references for identifying and classifying bacteria, respectively.
- Bacteria can be identified through biochemical tests, DNA/RNA analysis, and serological testing methods.

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1.7: Types of Microorganisms

Learning Objectives

- List the various types of microorganisms and describe their defining characteristics
- Give examples of different types of cellular and viral microorganisms and infectious agents
- Describe the similarities and differences between archaea and bacteria
- Provide an overview of the field of microbiology

Most microbes are unicellular and small enough that they require artificial magnification to be seen. However, there are some unicellular microbes that are visible to the naked eye, and some multicellular organisms that are microscopic. An object must measure about 100 micrometers (µm) to be visible without a microscope, but most microorganisms are many times smaller than that. For some perspective, consider that a typical animal cell measures roughly 10 µm across but is still microscopic. Bacterial cells are typically about 1 µm, and viruses can be 10 times smaller than bacteria (Figure 1.7.1). See Table 1.7.1 for units of length used in microbiology.

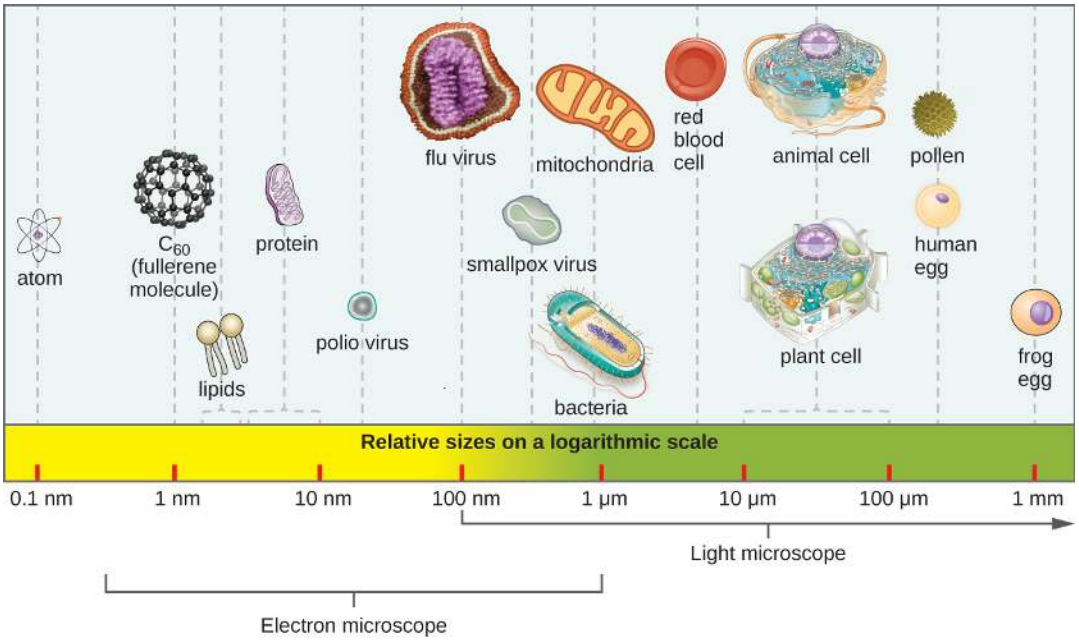


Figure 1.7.1: The relative sizes of various microscopic and nonmicroscopic objects. Note that a typical virus measures about 100 nm, 10 times smaller than a typical bacterium (~1 µm), which is at least 10 times smaller than a typical plant or animal cell (~10–100 µm). An object must measure about 100 µm to be visible without a microscope.

Table 1.7.1: Units of Length Commonly Used in Microbiology

Metric Unit	Meaning of Prefix	Metric Equivalent
meter (m)	—	1 m = 10^0 m
decimeter (dm)	1/10	1 dm = 0.1 m = 10^{-1} m
centimeter (cm)	1/100	1 cm = 0.01 m = 10^{-2} m
millimeter (mm)	1/1000	1 mm = 0.001 m = 10^{-3} m
micrometer (µm)	1/1,000,000	1 µm = 0.000001 m = 10^{-6} m
nanometer (nm)	1/1,000,000,000	1 nm = 0.000000001 m = 10^{-9} m

Microorganisms differ from each other not only in size, but also in structure, habitat, metabolism, and many other characteristics. While we typically think of microorganisms as being unicellular, there are also many multicellular organisms that are too small to

be seen without a microscope. Some microbes, such as viruses, are even **acellular** (not composed of cells).

Microorganisms are found in each of the three domains of life: Archaea, Bacteria, and Eukarya. Microbes within the domains Bacteria and Archaea are all prokaryotes (their cells lack a nucleus), whereas microbes in the domain Eukarya are eukaryotes (their cells have a nucleus). Some microorganisms, such as viruses, do not fall within any of the three domains of life. In this section, we will briefly introduce each of the broad groups of microbes. Later chapters will go into greater depth about the diverse species within each group.

Prokaryotic Microorganisms

Bacteria are found in nearly every habitat on earth, including within and on humans. Most bacteria are harmless or helpful, but some are **pathogens**, causing disease in humans and other animals. Bacteria are prokaryotic because their genetic material (DNA) is not housed within a true nucleus. Most bacteria have cell walls that contain peptidoglycan.

Bacteria are often described in terms of their general shape. Common shapes include spherical (coccus), rod-shaped (bacillus), or curved (spirillum, spirochete, or vibrio). Figure **1.7.2** shows examples of these shapes.

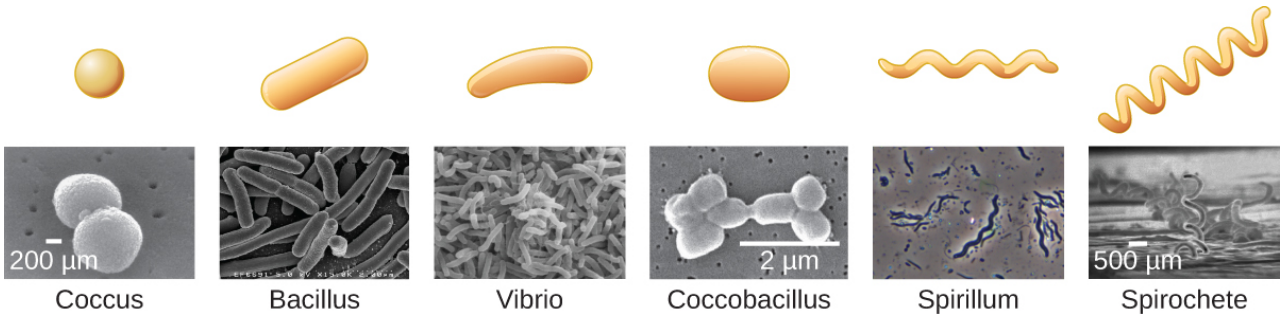


Figure **1.7.2**: Common bacterial shapes. Note how coccobacillus is a combination of spherical (coccus) and rod-shaped (bacillus). (credit “Coccus”: modification of work by Janice Haney Carr, Dr. Richard Facklam, Centers for Disease Control and Prevention; credit “Bacillus”: modification of work by “Elapied”/Wikimedia Commons)

They have a wide range of metabolic capabilities and can grow in a variety of environments, using different combinations of nutrients. Some bacteria are photosynthetic, such as oxygenic cyanobacteria and anoxygenic green sulfur and green nonsulfur bacteria; these bacteria use energy derived from sunlight, and fix carbon dioxide for growth. Other types of bacteria are nonphotosynthetic, obtaining their energy from organic or inorganic compounds in their environment.

Archaea are also unicellular prokaryotic organisms. Archaea and bacteria have different evolutionary histories, as well as significant differences in genetics, metabolic pathways, and the composition of their cell walls and membranes. Unlike most bacteria, archaeal cell walls do not contain peptidoglycan, but their cell walls are often composed of a similar substance called pseudopeptidoglycan. Like bacteria, archaea are found in nearly every habitat on earth, even extreme environments that are very cold, very hot, very basic, or very acidic (Figure **1.7.3**). Some archaea live in the human body, but none have been shown to be human pathogens.



Figure **1.7.3**: Some archaea live in extreme environments, such as the Morning Glory pool, a hot spring in Yellowstone National Park. The color differences in the pool result from the different communities of microbes that are able to thrive at various water temperatures.

Eukaryotic Microorganisms

The domain Eukarya contains all eukaryotes, including uni- or multicellular eukaryotes such as protists, fungi, plants, and animals. The major defining characteristic of eukaryotes is that their cells contain a nucleus.

Protists

Protists are unicellular eukaryotes that are not plants, animals, or fungi. Algae and protozoa are examples of protists.

Algae (singular: alga) are plant-like protists that can be either unicellular or multicellular (Figure **1.7.4**). Their cells are surrounded by cell walls made of cellulose, a type of carbohydrate. Algae are photosynthetic organisms that extract energy from the sun and release oxygen and carbohydrates into their environment. Because other organisms can use their waste products for energy, algae are important parts of many ecosystems. Many consumer products contain ingredients derived from algae, such as carrageenan or alginic acid, which are found in some brands of ice cream, salad dressing, beverages, lipstick, and toothpaste. A derivative of algae also plays a prominent role in the microbiology laboratory. Agar, a gel derived from algae, can be mixed with various nutrients and used to grow microorganisms in a Petri dish. Algae are also being developed as a possible source for biofuels.

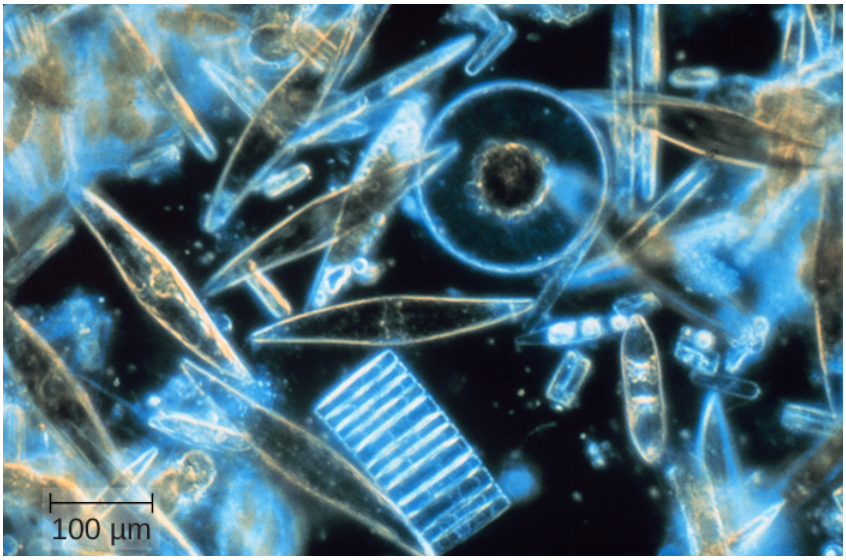


Figure **1.7.4**: Assorted diatoms, a kind of algae, live in annual sea ice in McMurdo Sound, Antarctica. Diatoms range in size from 2 µm to 200 µm and are visualized here using light microscopy. (credit: National Oceanic and Atmospheric Administration)

Protozoa (singular: protozoan) are protists that make up the backbone of many food webs by providing nutrients for other organisms. Protozoa are very diverse. Some protozoa move with help from hair-like structures called cilia or whip-like structures called flagella. Others extend part of their cell membrane and cytoplasm to propel themselves forward. These cytoplasmic extensions are called pseudopods ("false feet"). Some protozoa are photosynthetic; others feed on organic material. Some are free-living, whereas others are parasitic, only able to survive by extracting nutrients from a host organism. Most protozoa are harmless, but some are pathogens that can cause disease in animals or humans (Figure 1.7.5).



Figure 1.7.5: Giardia lamblia, an intestinal protozoan parasite that infects humans and other mammals, causing severe diarrhea. (credit: modification of work by Centers for Disease Control and Prevention)

Fungi

Fungi (singular: fungus) are also eukaryotes. Some multicellular fungi, such as mushrooms, resemble plants, but they are actually quite different. Fungi are not photosynthetic, and their cell walls are usually made out of chitin rather than cellulose.

Unicellular fungi—yeasts—are included within the study of microbiology. There are more than 1000 known species. Yeasts are found in many different environments, from the deep sea to the human navel. Some yeasts have beneficial uses, such as causing bread to rise and beverages to ferment; but yeasts can also cause food to spoil. Some even cause diseases, such as vaginal yeast infections and oral thrush (Figure 1.7.6).

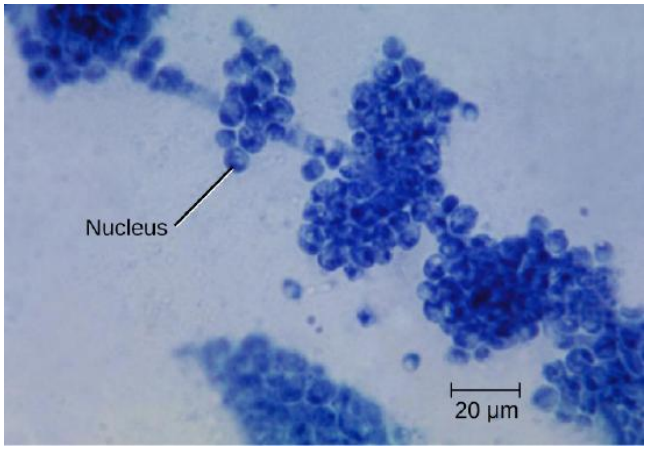


Figure 1.7.5: Candida albicans is a unicellular fungus, or yeast. It is the causative agent of vaginal yeast infections as well as oral thrush, a yeast infection of the mouth that commonly afflicts infants. *C. albicans* has a morphology similar to that of coccus bacteria; however, yeast is a eukaryotic organism (note the nuclei) and is much larger. (credit: modification of work by Centers for Disease Control and Prevention)

Other fungi of interest to microbiologists are multicellular organisms called molds. Molds are made up of long filaments that form visible colonies (Figure 1.7.6). Molds are found in many different environments, from soil to rotting food to dank bathroom corners. Molds play a critical role in the decomposition of dead plants and animals. Some molds can cause allergies, and others produce

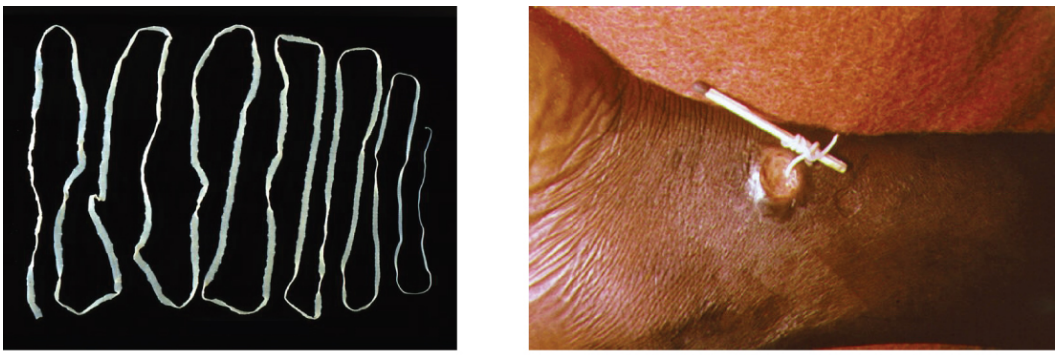
disease-causing metabolites called mycotoxins. Molds have been used to make pharmaceuticals, including penicillin, which is one of the most commonly prescribed antibiotics, and cyclosporine, used to prevent organ rejection following a transplant.



Figure **1.7.6**: Large colonies of microscopic fungi can often be observed with the naked eye, as seen on the surface of these moldy oranges.

Helminths

Multicellular parasitic worms called **helminths** are not technically microorganisms, as most are large enough to see without a microscope. However, these worms fall within the field of microbiology because diseases caused by helminths involve microscopic eggs and larvae. One example of a helminth is the guinea worm, or *Dracunculus medinensis*, which causes dizziness, vomiting, diarrhea, and painful ulcers on the legs and feet when the worm works its way out of the skin (Figure **1.7.7**). Infection typically occurs after a person drinks water containing water fleas infected by guinea-worm larvae. In the mid-1980s, there were an estimated 3.5 million cases of guinea-worm disease, but the disease has been largely eradicated. In 2014, there were only 126 cases reported, thanks to the coordinated efforts of the World Health Organization (WHO) and other groups committed to improvements in drinking water sanitation.[12]

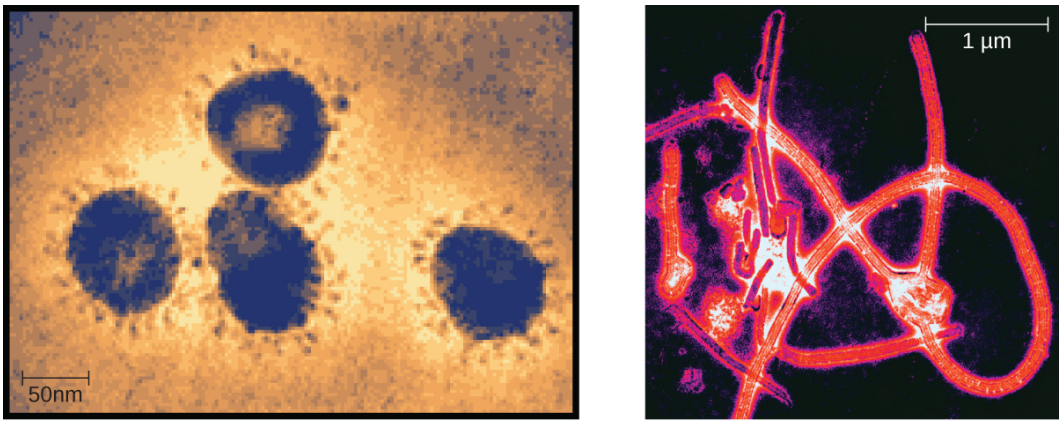


(a) (b)

Figure **1.7.7**: The beef tapeworm, *Taenia saginata*, infects both cattle and humans. *T. saginata* eggs are microscopic (around 50 µm), but adult worms like the one shown here can reach 4–10 m, taking up residence in the digestive system. (b) An adult guinea worm, *Dracunculus medinensis*, is removed through a lesion in the patient's skin by winding it around a matchstick. (credit b: modification of work by Centers for Disease Control and Prevention)

Viruses

Viruses are **acellular** microorganisms, which means they are not composed of cells. Essentially, a virus consists of proteins and genetic material—either DNA or RNA, but never both—that are inert outside of a host organism. However, by incorporating themselves into a host cell, viruses are able to co-opt the host's cellular mechanisms to multiply and infect other hosts. Viruses can infect all types of cells, from human cells to the cells of other microorganisms. In humans, viruses are responsible for numerous diseases, from the common cold to deadly Ebola (Figure **1.7.8**). However, many viruses do not cause disease.



(a) (b)

Figure **1.7.8**: (a) Members of the Coronavirus family can cause respiratory infections like the common cold, severe acute respiratory syndrome (SARS), and Middle East respiratory syndrome (MERS). Here they are viewed under a transmission electron microscope (TEM). (b) Ebolavirus, a member of the Filovirus family, as visualized using a TEM. (credit b: modification of work by Thomas W. Geisbert).

Microbiology as a Field of Study

Microbiology is a broad term that encompasses the study of all different types of microorganisms. But in practice, microbiologists tend to specialize in one of several subfields. For example, bacteriology is the study of bacteria; mycology is the study of fungi;protozoology is the study of protozoa; parasitology is the study of helminths and other parasites; and virology is the study of viruses (Figure **1.7.9**). **Immunology**, the study of the immune system, is often included in the study of microbiology because host–pathogen interactions are central to our understanding of infectious disease processes. Microbiologists can also specialize in certain areas of microbiology, such as clinical microbiology, environmental microbiology, applied microbiology, or food microbiology.

In this textbook, we are primarily concerned with clinical applications of microbiology, but since the various subfields of microbiology are highly interrelated, we will often discuss applications that are not strictly clinical.



Figure **1.7.9**: A virologist samples eggs from this nest to be tested for the influenza A virus, which causes avian flu in birds. (credit: Don Becker)

Bioethics in Microbiology

In the 1940s, the U.S. government was looking for a solution to a medical problem: the prevalence of sexually transmitted diseases (STDs) among soldiers. Several now-infamous government-funded studies used human subjects to research common STDs and treatments. In one such study, American researchers intentionally exposed more than 1300 human subjects in Guatemala to syphilis, gonorrhea, and chancroid to determine the ability of penicillin and other antibiotics to combat these diseases. Subjects of the study included Guatemalan soldiers, prisoners, prostitutes, and psychiatric patients—none of whom were informed that they were taking part in the study. Researchers exposed subjects to STDs by various methods, from facilitating intercourse with infected prostitutes to inoculating subjects with the bacteria known to cause the diseases. This latter method involved making a small wound on the subject's genitals or elsewhere on the body, and then putting bacteria directly into the wound.[3] In 2011, a U.S. government commission tasked with investigating the experiment revealed that only some of the subjects were treated with penicillin, and 83 subjects died by 1953, likely as a result of the study.[4]

Unfortunately, this is one of many horrific examples of microbiology experiments that have violated basic ethical standards. Even if this study had led to a life-saving medical breakthrough (it did not), few would argue that its methods were ethically sound or morally justifiable. But not every case is so clear cut. Professionals working in clinical settings are frequently confronted with ethical dilemmas, such as working with patients who decline a vaccine or life-saving blood transfusion. These are just two examples of life-and-death decisions that may intersect with the religious and philosophical beliefs of both the patient and the health-care professional.

No matter how noble the goal, microbiology studies and clinical practice must be guided by a certain set of ethical principles. Studies must be done with integrity. Patients and research subjects provide informed consent (not only agreeing to be treated or studied but demonstrating an understanding of the purpose of the study and any risks involved). Patients' rights must be respected. Procedures must be approved by an institutional review board. When working with patients, accurate record-keeping, honest communication, and confidentiality are paramount. Animals used for research must be treated humanely, and all protocols must be approved by an institutional animal care and use committee. These are just a few of the ethical principles explored in the *Eye on Ethics* boxes throughout this book.

Key Concepts and Summary

- Microorganisms are very diverse and are found in all three domains of life: Archaea, Bacteria, and Eukarya.
- Archaea and bacteria are classified as prokaryotes because they lack a cellular nucleus. Archaea differ from bacteria in evolutionary history, genetics, metabolic pathways, and cell wall and membrane composition.
- Archaea inhabit nearly every environment on earth, but no archaea have been identified as human pathogens.
- Eukaryotes studied in microbiology include algae, protozoa, fungi, and helminths.
- Algae are plant-like organisms that can be either unicellular or multicellular, and derive energy via photosynthesis.
- Protozoa are unicellular organisms with complex cell structures; most are motile.
- Microscopic fungi include molds and yeasts.
- Helminths are multicellular parasitic worms. They are included in the field of microbiology because their eggs and larvae are often microscopic.
- Viruses are acellular microorganisms that require a host to reproduce.
- The field of microbiology is extremely broad. Microbiologists typically specialize in one of many subfields, but all health professionals need a solid foundation in clinical microbiology.

Footnotes

1. 1 C. Greenaway “Dracunculiasis (Guinea Worm Disease).” *Canadian Medical Association Journal* 170 no. 4 (2004):495–500.
2. 2 World Health Organization. “Dracunculiasis (Guinea-Worm Disease).” *WHO*. 2015. http://www.who.int/mediacentre/factsheets/fs359/en/. Accessed October 2, 2015.
3. 3 Kara Rogers. “Guatemala Syphilis Experiment: American Medical Research Project”. *Encylopaedia Britannica*. www.britannica.com/event/Guat...lis-experiment. Accessed June 24, 2015.
4. 4 Susan Donaldson James. “Syphilis Experiments Shock, But So Do Third-World Drug Trials.” *ABC World News*. August 30, 2011. http://abcnews.go.com/Health/guatema...ry?id=14414902. Accessed June 24, 2015.
5. 5 Greg Botelho. “Case of Creutzfeldt-Jakob Disease Confirmed in New Hampshire.” *CNN*. 2013. http://www.cnn.com/2013/09/20/health...brain-disease/.
6. 6 P. Rudge et al. “Iatrogenic CJD Due to Pituitary-Derived Growth Hormone With Genetically Determined Incubation Times of Up to 40 Years.” *Brain* 138 no. 11 (2015): 3386–3399.
7. 7 J.G. Heckmann et al. “Transmission of Creutzfeldt-Jakob Disease via a Corneal Transplant.” *Journal of Neurology, Neurosurgery & Psychiatry* 63 no. 3 (1997): 388–390.
8. 8 National Institute of Neurological Disorders and Stroke. “Creutzfeldt-Jakob Disease Fact Sheet.” *NIH*. 2015. http://www.ninds.nih.gov/disorders/c....htm#288133058.
9. 9 National Institute of Neurological Disorders and Stroke. “Creutzfeldt-Jakob Disease Fact Sheet.” *NIH*. 2015. http://www.ninds.nih.gov/disorders/c....htm#288133058. Accessed June 22, 2015.

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1.8: Classification - The Three Domain System

1.05: Classification - The Three Domain System

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1.9: How We See the Invisible World

1.06: How We See the Invisible World

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1.E: An Invisible World (Exercises)

1.1: What Our Ancestors Knew

Microorganisms (or microbes) are living organisms that are generally too small to be seen without a microscope. Throughout history, humans have used microbes to make fermented foods such as beer, bread, cheese, and wine. Long before the invention of the microscope, some people theorized that infection and disease were spread by living things that were too small to be seen. They also correctly intuited certain principles regarding the spread of disease and immunity.

Multiple Choice

Which of the following foods is NOT made by fermentation?

A. beer
B. bread
C. cheese
D. orange juice

Answer

D

Who is considered the “father of Western medicine”?

A. Marcus Terentius Varro
B. Thucydides
C. Antonie van Leeuwenhoek
D. Hippocrates

Answer

D

Who was the first to observe “animalcules” under the microscope?

A. Antonie van Leeuwenhoek
B. Ötzi the Iceman
C. Marcus Terentius Varro
D. Robert Koch

Answer

A

Who proposed that swamps might harbor tiny, disease-causing animals too small to see?

A. Thucydides
B. Marcus Terentius Varro
C. Hippocrates
D. Louis Pasteur

Answer

B

Fill in the Blank

Thucydides is known as the father of _____.

Answer

scientific history

Researchers think that Ötzi the Iceman may have been infected with _____ disease.

Answer

Lyme

The process by which microbes turn grape juice into wine is called _____.

Answer

fermentation

Short Answer

What did Thucydides learn by observing the Athenian plague?

Why was the invention of the microscope important for microbiology?

What are some ways people use microbes?

Critical Thinking

Explain how the discovery of fermented foods likely benefited our ancestors.

What evidence would you use to support this statement: Ancient people thought that disease was transmitted by things they could not see.

1.2: A Systematic Approach

Carolus Linnaeus developed a taxonomic system for categorizing organisms into related groups. Binomial nomenclature assigns organisms Latinized scientific names with a genus and species designation. A phylogenetic tree is a way of showing how different organisms are thought to be related to one another from an evolutionary standpoint. The first phylogenetic tree contained kingdoms for plants and animals; Ernst Haeckel proposed adding a kingdom for protists.

Multiple Choice

Which of the following was NOT a kingdom in Linnaeus's taxonomy?

A. animal
B. mineral
C. protist
D. plant

Answer

C

Which of the following is a correct usage of binomial nomenclature?

A. Homo Sapiens
B. *homo sapiens*
C. *Homo sapiens*
D. *Homo Sapiens*

Answer

C

Which scientist proposed adding a kingdom for protists?

A. Carolus Linnaeus

B. Carl Woese
C. Robert Whittaker
D. Ernst Haeckel

Answer

D

Which of the following is NOT a domain in Woese and Fox's phylogenetic tree?

A. Plantae
B. Bacteria
C. Archaea
D. Eukarya

Answer

A

Which of the following is the standard resource for identifying bacteria?

A. *Systema Naturae*
B. Bergey's *Manual of Determinative Bacteriology*
C. Woese and Fox's phylogenetic tree
D. Haeckel's *General Morphology of Organisms*

Answer

B

Short Answer

What is a phylogenetic tree?

Which of the five kingdoms in Whittaker's phylogenetic tree are prokaryotic, and which are eukaryotic?

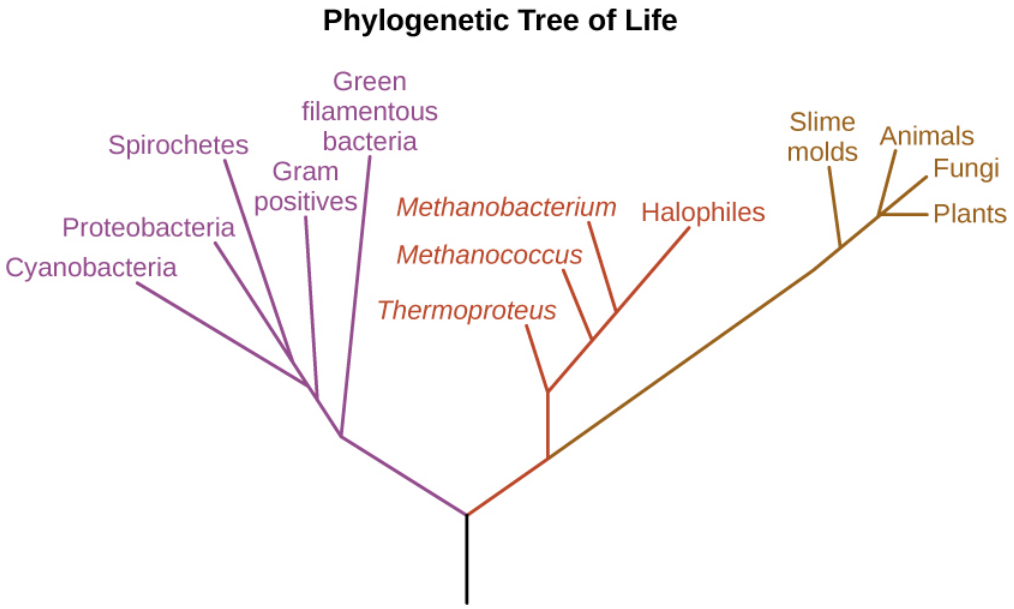
What molecule did Woese and Fox use to construct their phylogenetic tree?

Name some techniques that can be used to identify and differentiate species of bacteria.

Critical Thinking

Why is using binomial nomenclature more useful than using common names?

Label the three Domains found on modern phylogenetic trees.



1.3: Types of Microorganisms

Microorganisms are very diverse and are found in all three domains of life: Archaea, Bacteria, and Eukarya. Archaea and bacteria are classified as prokaryotes because they lack a cellular nucleus. Archaea differ from bacteria in evolutionary history, genetics, metabolic pathways, and cell wall and membrane composition. Archaea inhabit nearly every environment on earth, but no archaea have been identified as human pathogens.

Multiple Choice

Which of the following types of microorganisms is photosynthetic?

A. yeast
B. virus
C. helminth
D. alga

Answer

D

Which of the following is a prokaryotic microorganism?

A. helminth
B. protozoan
C. cyanobacterium
D. mold

Answer

C

Which of the following is acellular?

A. virus
B. bacterium
C. fungus
D. protozoan

Answer

A

Which of the following is a type of fungal microorganism?

A. bacterium
B. protozoan
C. alga
D. yeast

Answer

D

Which of the following is not a subfield of microbiology?

A. bacteriology
B. botany
C. clinical microbiology
D. virology

Answer

B

Fill in the Blank

A _____ is a disease-causing microorganism.

Answer

pathogen

Multicellular parasitic worms studied by microbiologists are called _____.

Answer

helminths

The study of viruses is _____.

Answer

virology

The cells of prokaryotic organisms lack a _____.

Answer

nucleus

Short Answer

Describe the differences between bacteria and archaea.

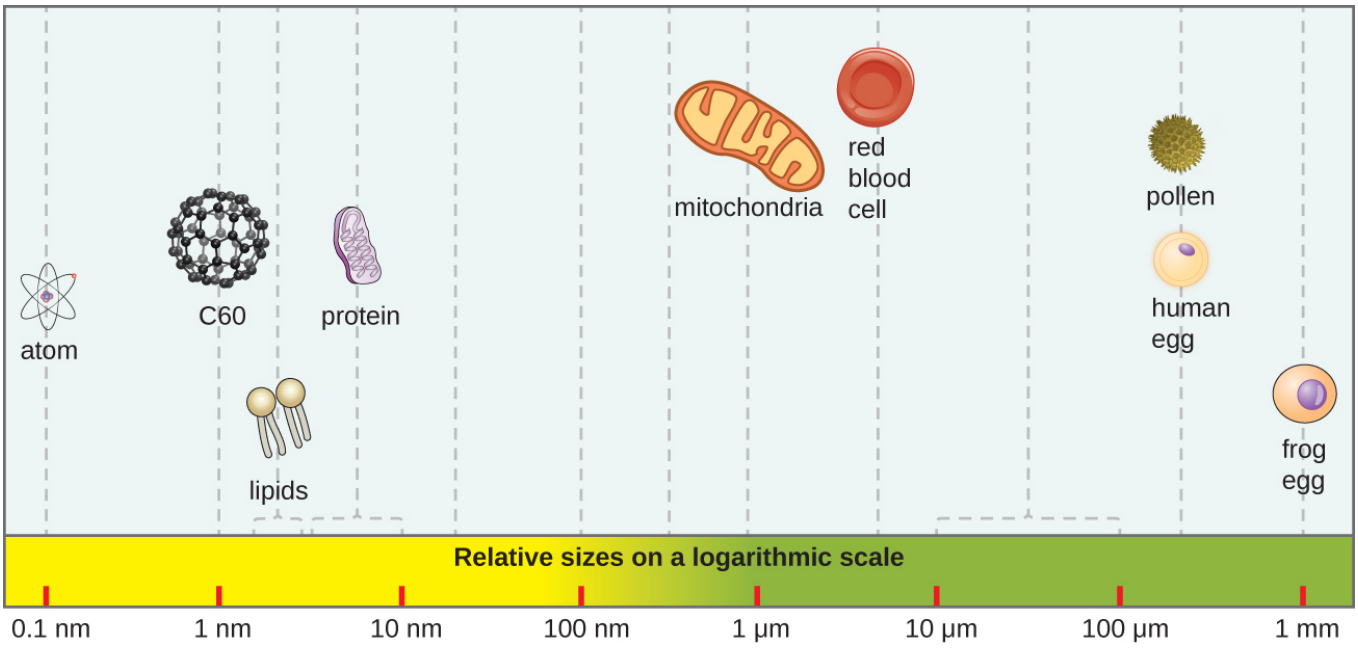
Name three structures that various protozoa use for locomotion.

Describe the actual and relative sizes of a virus, a bacterium, and a plant or animal cell.

Critical Thinking

Contrast the behavior of a virus outside versus inside a cell.

Where would a virus, bacterium, animal cell, and a prion belong on this chart?



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1.E: Fundamentals of Microbiology (Exercises)

1.E: Fundamentals of Microbiology (Exercises)

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CHAPTER OVERVIEW

2: Microscopes

When we look at a rainbow, its colors span the full spectrum of light that the human eye can detect and differentiate. Each hue represents a different frequency of visible light, processed by our eyes and brains and rendered as red, orange, yellow, green, or one of the many other familiar colors that have always been a part of the human experience. But only recently have humans developed an understanding of the properties of light that allow us to see images in color.

Over the past several centuries, we have learned to manipulate light to peer into previously invisible worlds—those too small or too far away to be seen by the naked eye. Through a microscope, we can examine microbial cells and colonies, using various techniques to manipulate color, size, and contrast in ways that help us identify species and diagnose disease.

Figure **2.1** illustrates how we can apply the properties of light to visualize and magnify images; but these stunning micrographs are just two examples of the numerous types of images we are now able to produce with different microscopic technologies. This chapter explores how various types of microscopes manipulate light in order to provide a window into the world of microorganisms. By understanding how various kinds of microscopes work, we can produce highly detailed images of microbes that can be useful for both research and clinical applications.

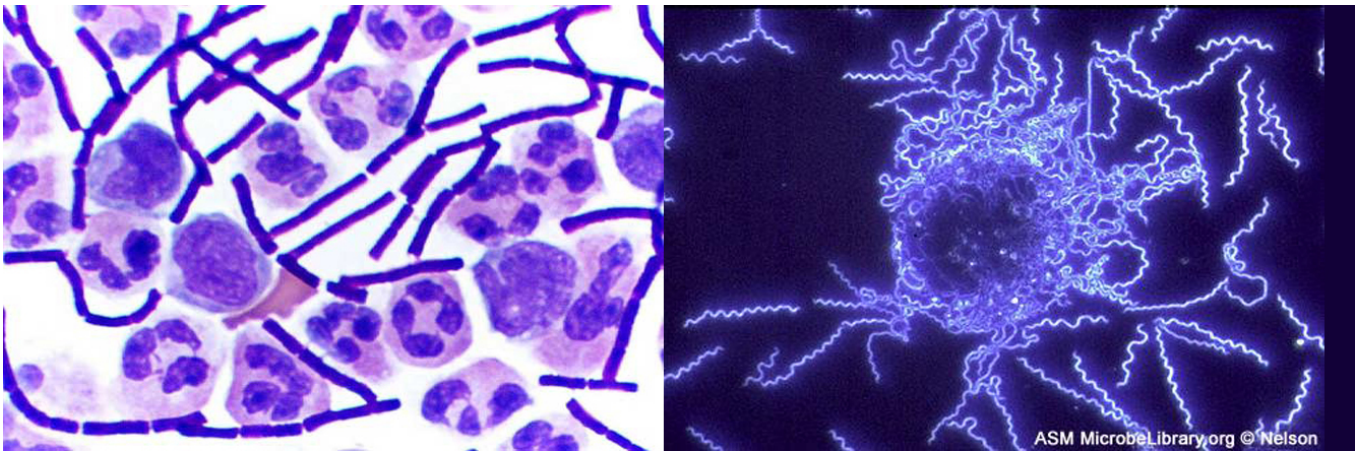


Figure **2.1**: Different types of microscopy are used to visualize different structures. Brightfield microscopy (left) renders a darker image on a lighter background, producing a clear image of these *Bacillus anthracis* cells in cerebrospinal fluid (the rod-shaped bacterial cells are surrounded by larger white blood cells). Darkfield microscopy (right) increases contrast, rendering a brighter image on a darker background, as demonstrated by this image of the bacterium *Borrelia burgdorferi*, which causes Lyme disease. (credit left: modification of work by Centers for Disease Control and Prevention; credit right: modification of work by American Society for Microbiology)

2.1: The Properties of Light
2.2: Peering into the Invisible World
2.3: Instruments of Microscopy
2.4: Staining Microscopic Specimens
2.E: How We See the Invisible World (Exercises)

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2.1: The Properties of Light

Learning Objectives

- Identify and define the characteristics of electromagnetic radiation (EMR) used in microscopy
- Explain how lenses are used in microscopy to manipulate visible and ultraviolet (UV) light

Visible light consists of electromagnetic waves that behave like other waves. Hence, many of the properties of light that are relevant to microscopy can be understood in terms of light's behavior as a wave. An important property of light waves is the **wavelength**, or the distance between one peak of a wave and the next peak. The height of each peak (or depth of each trough) is called the **amplitude**. In contrast, the **frequency** of the wave is the rate of vibration of the wave, or the number of wavelengths within a specified time period (Figure **2.1.1**).

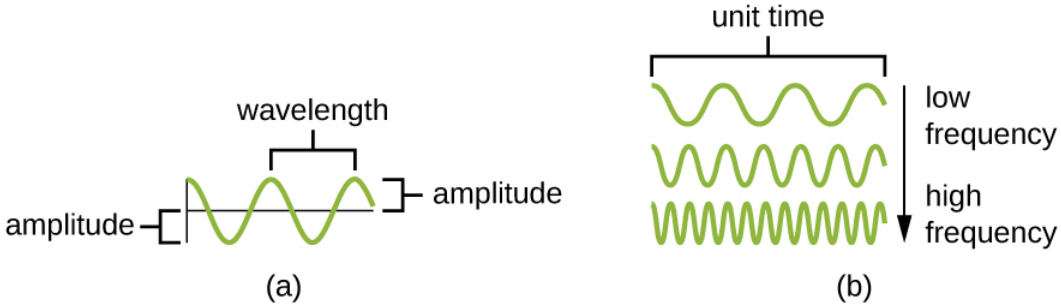
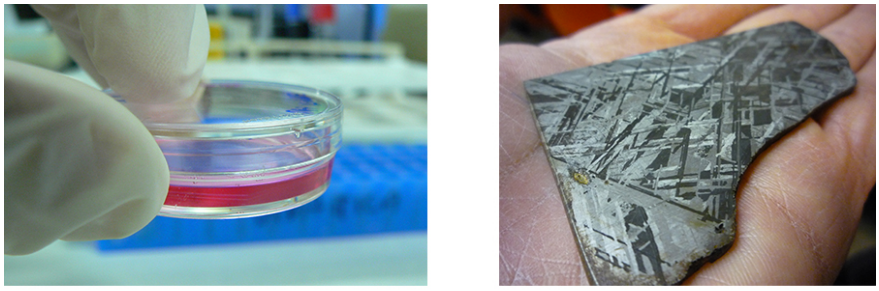


Figure **2.1.1**: (a) The amplitude is the height of a wave, whereas the wavelength is the distance between one peak and the next. (b) These waves have different frequencies, or rates of vibration. The wave at the top has the lowest frequency, since it has the fewest peaks per unit time. The wave at the bottom has the highest frequency.

Interactions of Light

Light waves interact with materials by being reflected, absorbed, or transmitted. **Reflection** occurs when a wave bounces off of a material. For example, a red piece of cloth may reflect red light to our eyes while absorbing other colors of light. **Absorbance** occurs when a material captures the energy of a light wave. In the case of glow-in-the-dark plastics, the energy from light can be absorbed and then later re-emitted as another form of phosphorescence. Transmission occurs when a wave travels through a material, like light through glass (the process of transmission is called **transmittance**). When a material allows a large proportion of light to be transmitted, it may do so because it is thinner, or more transparent (having more **transparency** and less **opacity**). Figure **2.1.2** illustrates the difference between transparency and opacity.



(a) (b)

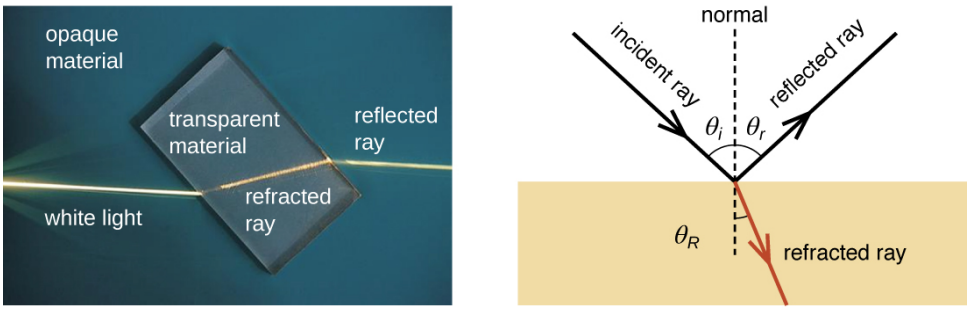
Figure **2.1.2**: (a) A Petri dish is made of transparent plastic or glass, which allows transmission of a high proportion of light. This transparency allows us to see through the sides of the dish to view the contents. (b) This slice of an iron meteorite is opaque (i.e., it has opacity). Light is not transmitted through the material, making it impossible to see the part of the hand covered by the object. (credit a: modification of work by Umberto Salvagnin; credit b: modification of work by "Waifer X"/Flickr)

Light waves can also interact with each other by **interference**, creating complex patterns of motion. Dropping two pebbles into a puddle causes the waves on the puddle's surface to interact, creating complex interference patterns. Light waves can interact in the same way.

In addition to interfering with each other, light waves can also interact with small objects or openings by bending or scattering. This is called **diffraction**. Diffraction is larger when the object is smaller relative to the wavelength of the light (the distance between two consecutive peaks of a light wave). Often, when waves diffract in different directions around an obstacle or opening, they will interfere with each other.

Lenses and Refraction

In the context of microscopy, refraction is perhaps the most important behavior exhibited by light waves. Refraction occurs when light waves change direction as they enter a new medium (Figure **2.1.3**). Different transparent materials transmit light at different speeds; thus, light can change speed when passing from one material to another. This change in speed usually also causes a change in direction (refraction), with the degree of change dependent on the angle of the incoming light.



(a) (b)

Figure **2.1.3**: (a) Refraction occurs when light passes from one medium, such as air, to another, such as glass, changing the direction of the light rays. (b) As shown in this diagram, light rays passing from one medium to another may be either refracted or reflected. (credit a: modification of work by "ajizai"/Wikimedia Commons).

The extent to which a material slows transmission speed relative to empty space is called the **refractive index** of that material. Large differences between the refractive indices of two materials will result in a large amount of refraction when light passes from one material to the other. For example, light moves much more slowly through water than through air, so light entering water from air can change direction greatly. We say that the water has a higher refractive index than air (Figure **2.1.4**).



Figure **2.1.4**: This straight pole appears to bend at an angle as it enters the water. This optical illusion is due to the large difference between the refractive indices of air and water.

When light crosses a boundary into a material with a higher refractive index, its direction turns to be closer to perpendicular to the boundary (i.e., more toward a normal to that boundary; Figure **2.1.5**). This is the principle behind lenses. We can think of a lens as an object with a curved boundary (or a collection of prisms) that collects all of the light that strikes it and refracts it so that it all meets at a single point called the **image point (focus)**. A convex lens can be used to magnify because it can focus at closer range than the human eye, producing a larger image. Concave lenses and mirrors can also be used in microscopes to redirect the light path. Figure **2.1.5** shows the **focal point** (the image point when light entering the lens is parallel) and the **focal length** (the distance to the focal point) for convex and concave lenses.

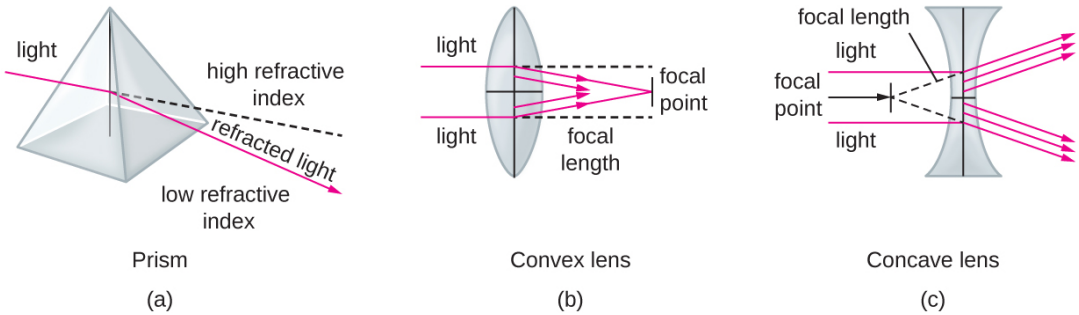


Figure **2.1.5**: (a) A lens is like a collection of prisms, such as the one shown here. (b) When light passes through a convex lens, it is refracted toward a focal point on the other side of the lens. The focal length is the distance to the focal point. (c) Light passing through a concave lens is refracted away from a focal point in front of the lens.

The human eye contains a lens that enables us to see images. This lens focuses the light reflecting off of objects in front of the eye onto the surface of the retina, which is like a screen in the back of the eye. Artificial lenses placed in front of the eye (contact lenses, glasses, or microscopic lenses) focus light before it is focused (again) by the lens of the eye, manipulating the image that ends up on the retina (e.g., by making it appear larger).

Images are commonly manipulated by controlling the distances between the object, the lens, and the screen, as well as the curvature of the lens. For example, for a given amount of curvature, when an object is closer to the lens, the focal points are farther from the lens. As a result, it is often necessary to manipulate these distances to create a focused image on a screen. Similarly, more curvature creates image points closer to the lens and a larger image when the image is in focus. This property is often described in terms of the focal distance, or distance to the focal point.

Electromagnetic Spectrum and Color

Visible light is just one form of **electromagnetic radiation (EMR)**, a type of energy that is all around us. Other forms of EMR include microwaves, X-rays, and radio waves, among others. The different types of EMR fall on the **electromagnetic spectrum**, which is defined in terms of wavelength and frequency. The spectrum of visible light occupies a relatively small range of frequencies between infrared and ultraviolet light (Figure **2.1.6**).

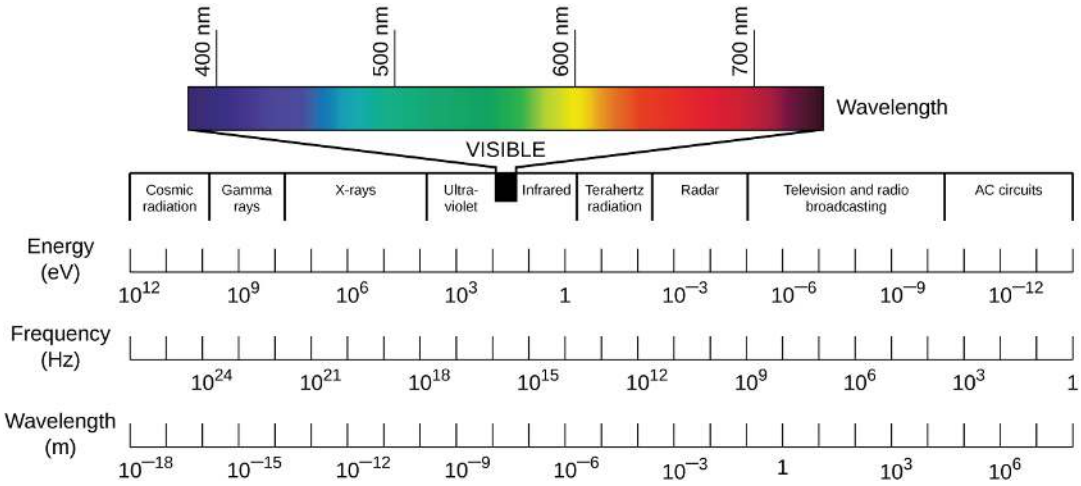


Figure **2.1.6**: The electromagnetic spectrum ranges from high-frequency gamma rays to low-frequency radio waves. Visible light is the relatively small range of electromagnetic frequencies that can be sensed by the human eye. On the electromagnetic spectrum, visible light falls between ultraviolet and infrared light. (credit: modification of work by Johannes Ahlmann).

Whereas wavelength represents the distance between adjacent peaks of a light wave, frequency, in a simplified definition, represents the rate of oscillation. Waves with higher frequencies have shorter wavelengths and, therefore, have more oscillations per unit time than lower-frequency waves. Higher-frequency waves also contain more energy than lower-frequency waves. This energy is delivered as elementary particles called photons. Higher-frequency waves deliver more energetic photons than lower-frequency waves.

Photons with different energies interact differently with the retina. In the spectrum of visible light, each color corresponds to a particular frequency and wavelength (Figure **2.1.6**).The lowest frequency of visible light appears as the color red, whereas the highest appears as the color violet. When the retina receives visible light of many different frequencies, we perceive this as white light. However, white light can be separated into its component colors using refraction. If we pass white light through a prism, different colors will be refracted in different directions, creating a rainbow-like spectrum on a screen behind the prism. This separation of colors is called dispersion, and it occurs because, for a given material, the refractive index is different for different frequencies of light.

Certain materials can refract nonvisible forms of EMR and, in effect, transform them into visible light. Certain **fluorescent dyes**, for instance, absorb ultraviolet or blue light and then use the energy to emit photons of a different color, giving off light rather than simply vibrating. This occurs because the energy absorption causes electrons to jump to higher energy states, after which they then almost immediately fall back down to their ground states, emitting specific amounts of energy as photons. Not all of the energy is emitted in a given photon, so the emitted photons will be of lower energy and, thus, of lower frequency than the absorbed ones. Thus, a dye such as Texas red may be excited by blue light, but emit red light; or a dye such as fluorescein isothiocyanate (FITC) may absorb (invisible) high-energy ultraviolet light and emit green light (Figure **2.1.7**). In some materials, the photons may be emitted following a delay after absorption; in this case, the process is called **phosphorescence**. Glow-in-the-dark plastic works by using phosphorescent material.

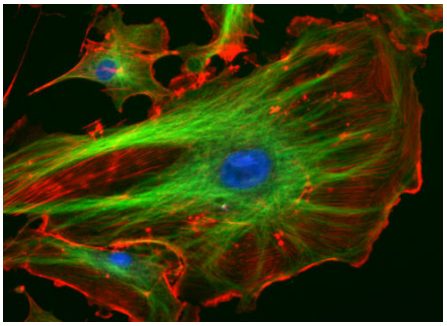


Figure **2.1.7**: The fluorescent dyes absorbed by these bovine pulmonary artery endothelial cells emit brilliant colors when excited by ultraviolet light under a fluorescence microscope. Various cell structures absorb different dyes. The nuclei are stained blue with 4’,6-diamidino-2-phenylindole (DAPI); microtubles are marked green by an antibody bound to FITC; and actin filaments are labeled red with phalloidin bound to tetramethylrhodamine (TRITC).

Magnification, Resolution, and Contrast

Microscopes magnify images and use the properties of light to create useful images of small objects. **Magnification** is defined as the ability of a lens to enlarge the image of an object when compared to the real object. For example, a magnification of 10× means that the image appears 10 times the size of the object as viewed with the naked eye.

Greater magnification typically improves our ability to see details of small objects, but magnification alone is not sufficient to make the most useful images. It is often useful to enhance the **resolution** of objects: the ability to tell that two separate points or objects are separate. A low-resolution image appears fuzzy, whereas a high-resolution image appears sharp. Two factors affect resolution. The first is wavelength. Shorter wavelengths are able to resolve smaller objects; thus, an electron microscope has a much higher resolution than a light microscope, since it uses an electron beam with a very short wavelength, as opposed to the long-wavelength visible light used by a light microscope. The second factor that affects resolution is numerical aperture, which is a measure of a lens’s ability to gather light. The higher the **numerical aperture**, the better the resolution.

Even when a microscope has high resolution, it can be difficult to distinguish small structures in many specimens because microorganisms are relatively transparent. It is often necessary to increase **contrast** to detect different structures in a specimen. Various types of microscopes use different features of light or electrons to increase contrast—visible differences between the parts of a specimen (see Instruments of Microscopy). Additionally, dyes that bind to some structures but not others can be used to improve the contrast between images of relatively transparent objects (see Staining Microscopic Specimens).

Key Concepts and Summary

- Light waves interacting with materials may be reflected, absorbed, or transmitted, depending on the properties of the material.
- Light waves can interact with each other (interference) or be distorted by interactions with small objects or openings (diffraction).
- Refraction occurs when light waves change speed and direction as they pass from one medium to another. Differences in the refraction indices of two materials determine the magnitude of directional changes when light passes from one to the other.
- A lens is a medium with a curved surface that refracts and focuses light to produce an image.
- Visible light is part of the electromagnetic spectrum; light waves of different frequencies and wavelengths are distinguished as colors by the human eye.
- A prism can separate the colors of white light (dispersion) because different frequencies of light have different refractive indices for a given material.
- Fluorescent dyes and phosphorescent materials can effectively transform nonvisible electromagnetic radiation into visible light.
- The power of a microscope can be described in terms of its magnification and resolution.
- Resolution can be increased by shortening wavelength, increasing the numerical aperture of the lens, or using stains that enhance contrast.

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2.2: Peering into the Invisible World

Learning Objectives

- Describe historical developments and individual contributions that led to the invention and development of the microscope
- Compare and contrast the features of simple and compound microscopes

Some of the fundamental characteristics and functions of microscopes can be understood in the context of the history of their use. Italian scholar Girolamo Fracastoro is regarded as the first person to formally postulate that disease was spread by tiny invisible*seminaria*, or "seeds of the contagion." In his book *De Contagione* (1546), he proposed that these seeds could attach themselves to certain objects (which he called *fomes* [cloth]) that supported their transfer from person to person. However, since the technology for seeing such tiny objects did not yet exist, the existence of the *seminaria* remained hypothetical for a little over a century—an invisible world waiting to be revealed.

Early Microscopes

Antonie van Leeuwenhoek, sometimes hailed as "the Father of Microbiology," is typically credited as the first person to have created microscopes powerful enough to view microbes (Figure **2.2.1**). Born in the city of Delft in the Dutch Republic, van Leeuwenhoek began his career selling fabrics. However, he later became interested in lens making (perhaps to look at threads) and his innovative techniques produced microscopes that allowed him to observe microorganisms as no one had before. In 1674, he described his observations of single-celled organisms, whose existence was previously unknown, in a series of letters to the Royal Society of London. His report was initially met with skepticism, but his claims were soon verified and he became something of a celebrity in the scientific community.

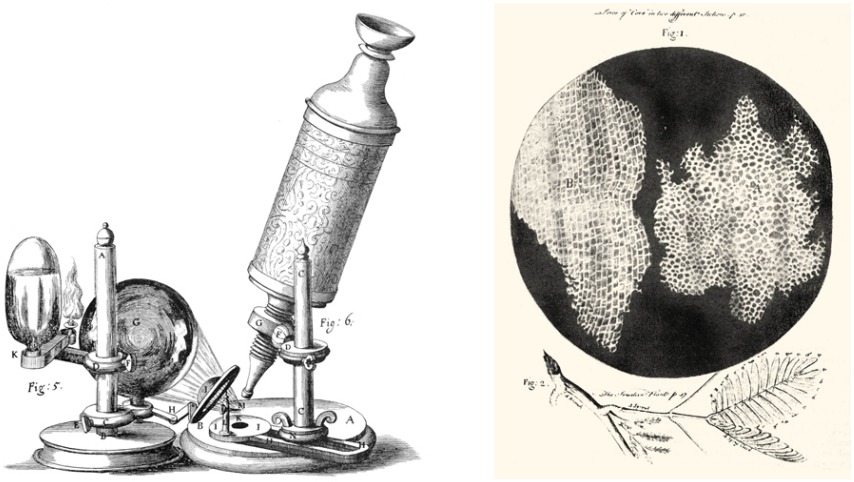


(a) (b) (c)

Figure **2.2.1**: (a) Antonie van Leeuwenhoek (1632–1723) is credited as being the first person to observe microbes, including bacteria, which he called "animalcules" and "wee little beasties." (b) Even though van Leeuwenhoek's microscopes were simple microscopes (as seen in this replica), they were more powerful and provided better resolution than the compound microscopes of his day. (credit b: modification of work by "Wellcome Images"/Wikimedia Commons) (c) Though more famous for developing the telescope, Galileo Galilei (1564–1642) was also one of the pioneers of microscopy.

While van Leeuwenhoek is credited with the discovery of microorganisms, others before him had contributed to the development of the microscope. These included eyeglass makers in the Netherlands in the late 1500s, as well as the Italian astronomer Galileo Galilei, who used a **compound microscope** to examine insect parts (Figure **2.2.1**). Whereas van Leeuwenhoek used a **simple microscope**, in which light is passed through just one lens, Galileo's compound microscope was more sophisticated, passing light through two sets of lenses.

Van Leeuwenhoek's contemporary, the Englishman **Robert Hooke** (1635–1703), also made important contributions to microscopy, publishing in his book *Micrographia* (1665) many observations using compound microscopes. Viewing a thin sample of cork through his microscope, he was the first to observe the structures that we now know as cells (Figure **2.2.2**). Hooke described these structures as resembling "Honey-comb," and as "small Boxes or Bladders of Air," noting that each "Cavern, Bubble, or Cell" is distinct from the others (in Latin, "cell" literally means "small room"). They likely appeared to Hooke to be filled with air because the cork cells were dead, with only the rigid cell walls providing the structure.



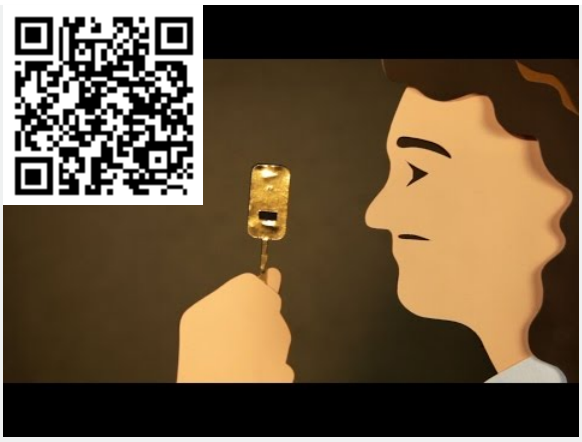
(a) (b)

Figure **2.2.2**: Robert Hooke used his (a) compound microscope to view (b) cork cells. Both of these engravings are from his seminal work *Micrographia*, published in 1665.

Video: Seeing the Invisible: van Leeuwenhoek's first glimpses of the microbial world

In 1674, Antonie van Leeuwenhoek looked at a drop of lake water through his homemade microscope and discovered an invisible world that no one knew existed. His work inspired countless microbiology researchers, including HHMI investigator Bonnie Bassler, one of the narrators of this animated feature. Leeuwenhoek was a haberdasher and city official in Delf, The Netherlands.He started making simple microscopes and using them to observe the world around him. He was the first to discover bacteria, protists, sperm cells, blood cells, rotifers, and much more.





Who Invented the Microscope?

While Antonie van Leeuwenhoek and Robert Hooke generally receive much of the credit for early advances in microscopy, neither can claim to be the inventor of the microscope. Some argue that this designation should belong to Hans and Zaccharias Janssen, Dutch spectacle-makers who may have invented the telescope, the simple microscope, and the compound microscope during the late 1500s or early 1600s (Figure **2.2.3**). Unfortunately, little is known for sure about the Janssens, not even the exact dates of their births and deaths. The Janssens were secretive about their work and never published. It is also possible that the Janssens did not invent anything at all; their neighbor, Hans Lippershey, also developed microscopes and telescopes during the same time frame, and he is often credited with inventing the telescope. The historical records from the time are as fuzzy and imprecise as the images viewed through those early lenses, and any archived records have been lost over the centuries.



Figure **2.2.3**: Zaccharias Janssen, along with his father Hans, may have invented the telescope, the simple microscope, and the compound microscope during the late 1500s or early 1600s. The historical evidence is inconclusive.

By contrast, van Leeuwenhoek and Hooke can thank ample documentation of their work for their respective legacies. Like Janssen, van Leeuwenhoek began his work in obscurity, leaving behind few records. However, his friend, the prominent physician Reinier de Graaf, wrote a letter to the editor of the *Philosophical Transactions of the Royal Society of London* calling attention to van Leeuwenhoek's powerful microscopes. From 1673 onward, van Leeuwenhoek began regularly submitting letters to the Royal Society detailing his observations. In 1674, his report describing single-celled organisms produced controversy in the scientific community, but his observations were soon confirmed when the society sent a delegation to investigate his findings. He subsequently enjoyed considerable celebrity, at one point even entertaining a visit by the czar of Russia.

Similarly, Robert Hooke had his observations using microscopes published by the Royal Society in a book called *Micrographia* in 1665. The book became a bestseller and greatly increased interest in microscopy throughout much of Europe.

Key Concepts and Summary

- Antonie van Leeuwenhoek is credited with the first observation of microbes, including protists and bacteria, with simple microscopes that he made.
- Robert Hooke was the first to describe what we now call cells.
- Simple microscopes have a single lens, while compound microscopes have multiple lenses.

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2.3: Instruments of Microscopy

Learning Objectives

- Identify and describe the parts of a brightfield microscope
- Calculate total magnification for a compound microscope
- Describe the distinguishing features and typical uses for various types of light microscopes, electron microscopes, and scanning probe microscopes

The early pioneers of microscopy opened a window into the invisible world of microorganisms. But microscopy continued to advance in the centuries that followed. In 1830, Joseph Jackson Lister created an essentially modern light microscope. The 20th century saw the development of microscopes that leveraged nonvisible light, such as fluorescence microscopy, which uses an ultraviolet light source, and electron microscopy, which uses short-wavelength electron beams. These advances led to major improvements in magnification, resolution, and contrast. By comparison, the relatively rudimentary microscopes of van Leeuwenhoek and his contemporaries were far less powerful than even the most basic microscopes in use today. In this section, we will survey the broad range of modern microscopic technology and common applications for each type of microscope.

Light Microscopy

Many types of microscopes fall under the category of light microscopes, which use light to visualize images. Examples of light microscopes include brightfield microscopes, darkfield microscopes, phase-contrast microscopes, differential interference contrast microscopes, fluorescence microscopes, confocal scanning laser microscopes, and two-photon microscopes. These various types of light microscopes can be used to complement each other in diagnostics and research.

Brightfield Microscopes

The **brightfield microscope**, perhaps the most commonly used type of microscope, is a compound microscope with two or more lenses that produce a dark image on a bright background. Some brightfield microscopes are **monocular** (having a single eyepiece), though most newer brightfield microscopes are **binocular** (having two eyepieces), like the one shown in Figure 2.3.1; in either case, each eyepiece contains a lens called an **ocular lens**. The ocular lenses typically magnify images 10 times (10×). At the other end of the body tube are a set of **objective lenses** on a rotating nosepiece. The magnification of these objective lenses typically ranges from 4× to 100×, with the magnification for each lens designated on the metal casing of the lens. The ocular and objective lenses work together to create a magnified image. The **total magnification** is the product of the ocular magnification times the objective magnification:

$$\text{ocular magnification} \times \text{objective magnification}$$

For example, if a 40× objective lens is selected and the ocular lens is 10×, the total magnification would be

$$(40\times)(10\times) = 400\times$$

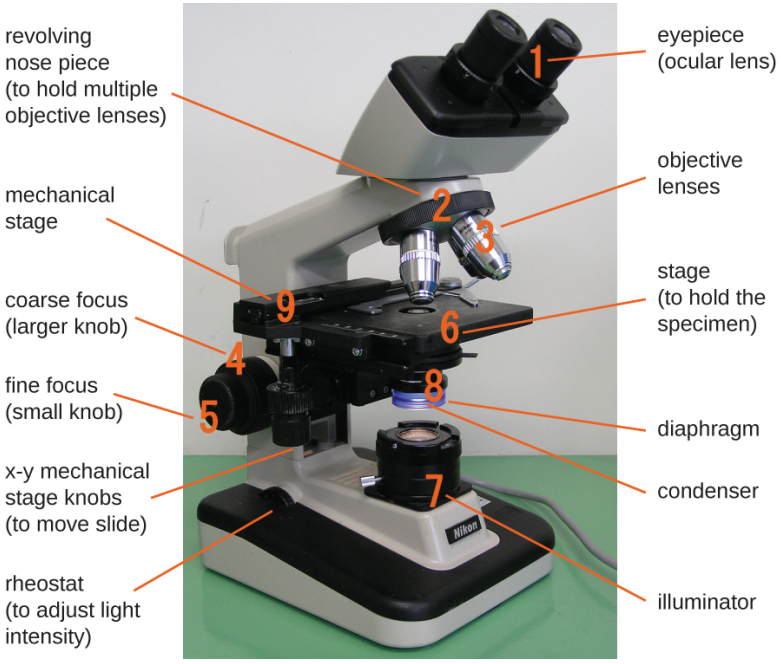


Figure **2.3.1**: Components of a typical brightfield microscope.

Components of a typical brightfield microscope.

The item being viewed is called a **specimen**. The specimen is placed on a glass slide, which is then clipped into place on the **stage**(a platform) of the microscope. Once the slide is secured, the specimen on the slide is positioned over the light using the **x-y mechanical stage knobs**. These knobs move the slide on the surface of the stage, but do not raise or lower the stage. Once the specimen is centered over the light, the stage position can be raised or lowered to focus the image. The **coarse focusing knob** is used for large-scale movements with 4× and 10× objective lenses; the **fine focusing knob** is used for small-scale movements, especially with 40× or 100× objective lenses.

When images are magnified, they become dimmer because there is less light per unit area of image. Highly magnified images produced by microscopes, therefore, require intense lighting. In a brightfield microscope, this light is provided by an **illuminator**, which is typically a high-intensity bulb below the stage. Light from the illuminator passes up through **condenser lens** (located below the stage), which focuses all of the light rays on the specimen to maximize illumination. The position of the condenser can be optimized using the attached condenser focus knob; once the optimal distance is established, the condenser should not be moved to adjust the brightness. If less-than-maximal light levels are needed, the amount of light striking the specimen can be easily adjusted by opening or closing a **diaphragm** between the condenser and the specimen. In some cases, brightness can also be adjusted using the **rheostat**, a dimmer switch that controls the intensity of the illuminator.

A brightfield microscope creates an image by directing light from the illuminator at the specimen; this light is differentially transmitted, absorbed, reflected, or refracted by different structures. Different colors can behave differently as they interact with**chromophores** (pigments that absorb and reflect particular wavelengths of light) in parts of the specimen. Often, chromophores are artificially added to the specimen using stains, which serve to increase contrast and resolution. In general, structures in the specimen will appear darker, to various extents, than the bright background, creating maximally sharp images at magnifications up to about 1000×. Further magnification would create a larger image, but without increased resolution. This allows us to see objects as small as bacteria, which are visible at about 400× or so, but not smaller objects such as viruses.

At very high magnifications, resolution may be compromised when light passes through the small amount of air between the specimen and the lens. This is due to the large difference between the refractive indices of air and glass; the air scatters the light rays before they can be focused by the lens. To solve this problem, a drop of oil can be used to fill the space between the specimen and an **oil immersion lens**, a special lens designed to be used with immersion oils. Since the oil has a refractive index very similar

to that of glass, it increases the maximum angle at which light leaving the specimen can strike the lens. This increases the light collected and, thus, the resolution of the image (Figure **2.3.2**). A variety of oils can be used for different types of light.

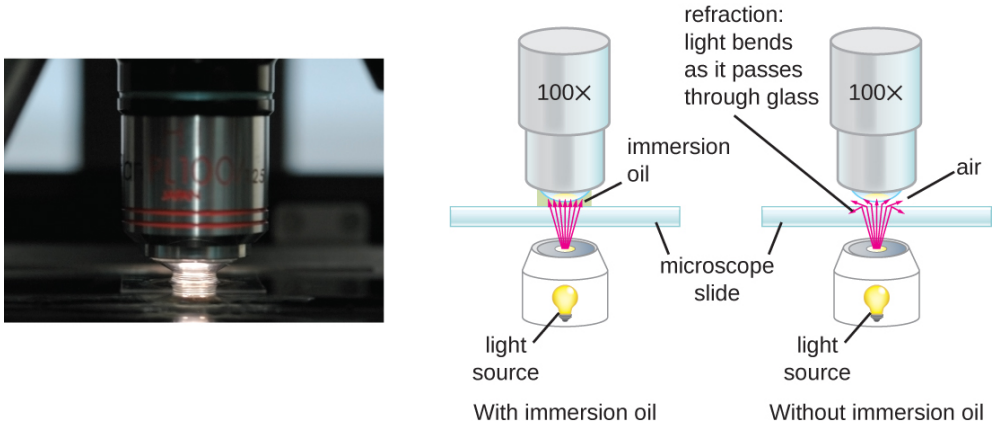


Figure **2.3.2**: (a) Oil immersion lenses like this one are used to improve resolution. (b) Because immersion oil and glass have very similar refractive indices, there is a minimal amount of refraction before the light reaches the lens. Without immersion oil, light scatters as it passes through the air above the slide, degrading the resolution of the image.

Microscope Maintenance: Best Practices

Even a very powerful microscope cannot deliver high-resolution images if it is not properly cleaned and maintained. Since lenses are carefully designed and manufactured to refract light with a high degree of precision, even a slightly dirty or scratched lens will refract light in unintended ways, degrading the image of the specimen. In addition, microscopes are rather delicate instruments, and great care must be taken to avoid damaging parts and surfaces. Among other things, proper care of a microscope includes the following:

- cleaning the lenses with lens paper
- not allowing lenses to contact the slide (e.g., by rapidly changing the focus)
- protecting the bulb (if there is one) from breakage
- not pushing an objective into a slide
- not using the coarse focusing knob when using the 40× or greater objective lenses
- only using immersion oil with a specialized oil objective, usually the 100× objective
- cleaning oil from immersion lenses after using the microscope
- cleaning any oil accidentally transferred from other lenses
- covering the microscope or placing it in a cabinet when not in use

Key Concepts and Summary

- Numerous types of microscopes use various technologies to generate micrographs. Most are useful for a particular type of specimen or application.
- Light microscopy uses lenses to focus light on a specimen to produce an image. Commonly used light microscopes include brightfield, darkfield, phase-contrast, differential interference contrast, fluorescence, confocal, and two-photon microscopes.

Footnotes

1. 1 “JEM-ARM200F Transmission Electron Microscope,” *JEOL USA Inc*, www.jeolusa.com/PRODUCTS/Tran...specifications. Accessed 8/28/2015.

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2.4: Staining Microscopic Specimens

Learning Objectives

- Differentiate between wet mount and fixation in terms of microscopic sample preparation
- Describe the unique features of commonly used stains
- Identify appropriate dyes for positive vs negative stains
- Differentiate between simple and differential stains

In their natural state, most of the cells and microorganisms that we observe under the microscope lack color and contrast. This makes it difficult, if not impossible, to detect important cellular structures and their distinguishing characteristics without artificially treating specimens. We have already alluded to certain techniques involving stains and fluorescent dyes, and in this section we will discuss specific techniques for sample preparation in greater detail. Indeed, numerous methods have been developed to identify specific microbes, cellular structures, DNA sequences, or indicators of infection in tissue samples, under the microscope. Here, we will focus on the most clinically relevant techniques.

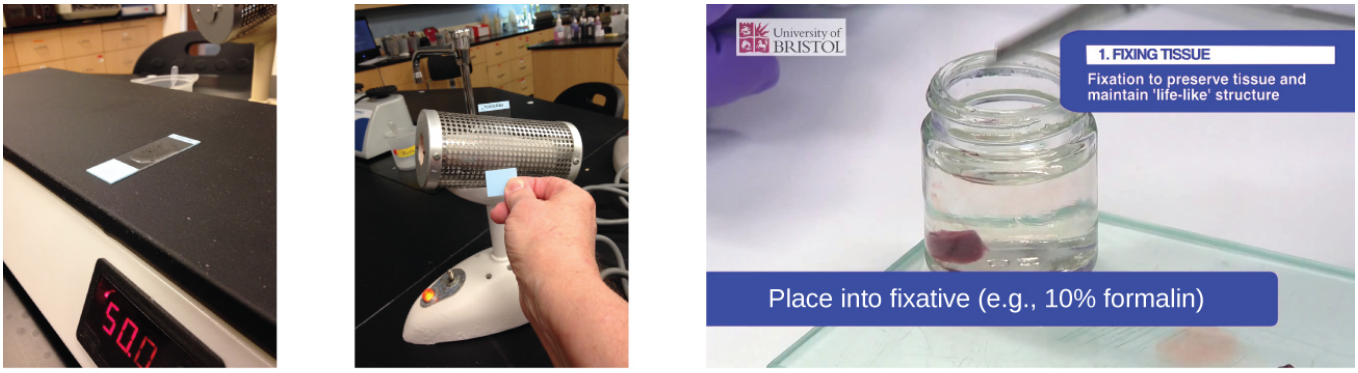
Preparing Specimens for Light Microscopy

In clinical settings, light microscopes are the most commonly used microscopes. There are two basic types of preparation used to view specimens with a light microscope: wet mounts and fixed specimens.

The simplest type of preparation is the **wet mount**, in which the specimen is placed on the slide in a drop of liquid. Some specimens, such as a drop of urine, are already in a liquid form and can be deposited on the slide using a dropper. Solid specimens, such as a skin scraping, can be placed on the slide before adding a drop of liquid to prepare the wet mount. Sometimes the liquid used is simply water, but often stains are added to enhance contrast. Once the liquid has been added to the slide, a coverslip is placed on top and the specimen is ready for examination under the microscope.

The second method of preparing specimens for light microscopy is **fixation**. The “fixing” of a sample refers to the process of attaching cells to a slide. Fixation is often achieved either by heating (heat fixing) or chemically treating the specimen. In addition to attaching the specimen to the slide, fixation also kills microorganisms in the specimen, stopping their movement and metabolism while preserving the integrity of their cellular components for observation.

To heat-fix a sample, a thin layer of the specimen is spread on the slide (called a smear), and the slide is then briefly heated over a heat source (Figure **2.4.1**). Chemical fixatives are often preferable to heat for tissue specimens. Chemical agents such as acetic acid, ethanol, methanol, formaldehyde (formalin), and glutaraldehyde can denature proteins, stop biochemical reactions, and stabilize cell structures in tissue samples (Figure **2.4.1**).

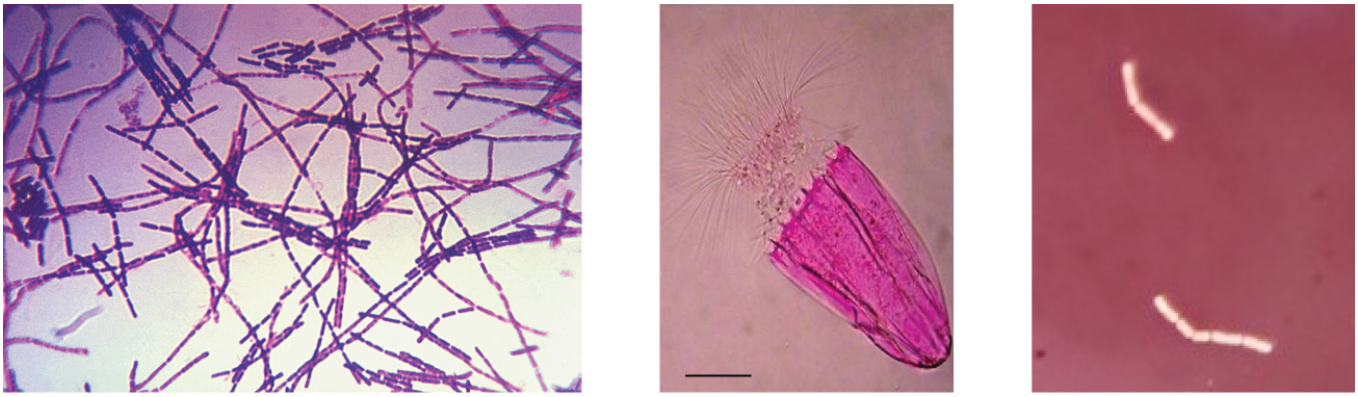


(a) (b) (c)

Figure **2.4.1**: (a) A specimen can be heat-fixed by using a slide warmer like this one. (b) Another method for heat-fixing a specimen is to hold a slide with a smear over a microincinerator. (c) This tissue sample is being fixed in a solution of formalin (also known as formaldehyde). Chemical fixation kills microorganisms in the specimen, stopping degradation of the tissues and preserving their structure so that they can be examined later under the microscope. (credit a: modification of work by Nina Parker; credit b: modification of work by Nina Parker; credit c: modification of work by “University of Bristol”/YouTube)

In addition to fixation, **staining** is almost always applied to color certain features of a specimen before examining it under a light microscope. Stains, or dyes, contain salts made up of a positive ion and a negative ion. Depending on the type of dye, the positive or the negative ion may be the chromophore (the colored ion); the other, uncolored ion is called the counterion. If the chromophore is the positively charged ion, the stain is classified as a **basic dye**; if the negative ion is the chromophore, the stain is considered an **acidic dye**.

Dyes are selected for staining based on the chemical properties of the dye and the specimen being observed, which determine how the dye will interact with the specimen. In most cases, it is preferable to use a **positive stain**, a dye that will be absorbed by the cells or organisms being observed, adding color to objects of interest to make them stand out against the background. However, there are scenarios in which it is advantageous to use a **negative stain**, which is absorbed by the background but not by the cells or organisms in the specimen. Negative staining produces an outline or silhouette of the organisms against a colorful background (Figure **2.4.2**).



(a) (b) (c)

Figure **2.4.2**: (a) These *Bacillus anthracis* cells have absorbed crystal violet, a basic positive stain. (b) This specimen of *Spinoloricus*, a microscopic marine organism, has been stained with rose bengal, a positive acidic stain. (c) These *B. megaterium* appear to be white because they have not absorbed the negative red stain applied to the slide. (credit a: modification of work by Centers for Disease Control and Prevention; credit b: modification of work by Roberto Danovaro, Antonio Pusceddu, Cristina Gambi, Iben Heiner, Reinhardt Mobjerg Kristensen; credit c: modification of work by Anh-Hue Tu)

Because cells typically have negatively charged cell walls, the positive chromophores in basic dyes tend to stick to the cell walls, making them positive stains. Thus, commonly used basic dyes such as basic fuchsin, crystal violet, malachite green, methylene blue, and safranin typically serve as positive stains. On the other hand, the negatively charged chromophores in acidic dyes are repelled by negatively charged cell walls, making them negative stains. Commonly used acidic dyes include acid fuchsin, eosin, and rose bengal. Figure **2.4.10** provides more detail.

Some staining techniques involve the application of only one dye to the sample; others require more than one dye. In **simple staining**, a single dye is used to emphasize particular structures in the specimen. A simple stain will generally make all of the organisms in a sample appear to be the same color, even if the sample contains more than one type of organism. In contrast, **differential staining** distinguishes organisms based on their interactions with multiple stains. In other words, two organisms in a differentially stained sample may appear to be different colors. Differential staining techniques commonly used in clinical settings include Gram staining, acid-fast staining, endospore staining, flagella staining, and capsule staining. Figure **2.4.11** provides more detail on these differential staining techniques.

Key Concepts and Summary

- Samples must be properly prepared for microscopy. This may involve **staining**, **fixation**, and/or cutting **thin sections**.

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2.E: How We See the Invisible World (Exercises)

2.1: The Properties of Light

Visible light consists of electromagnetic waves that behave like other waves. Hence, many of the properties of light that are relevant to microscopy can be understood in terms of light's behavior as a wave. An important property of light waves is the wavelength, or the distance between one peak of a wave and the next peak. The height of each peak (or depth of each trough) is called the amplitude.

Multiple Choice

Which of the following has the highest energy?

A. light with a long wavelength
B. light with an intermediate wavelength
C. light with a short wavelength
D. It is impossible to tell from the information given.

Answer

C

You place a specimen under the microscope and notice that parts of the specimen begin to emit light immediately. These materials can be described as _____.

A. fluorescent
B. phosphorescent
C. transparent
D. opaque

Answer

A

Fill in the Blank

When you see light bend as it moves from air into water, you are observing _____.

Answer

refraction

Short Answer

Explain how a prism separates white light into different colors.

Critical Thinking

In Figure 2.1.6, which of the following has the lowest energy?

A. visible light
B. X-rays
C. ultraviolet rays
D. infrared rays

2.2: Peering into the Invisible World

Italian scholar Girolamo Fracastoro is regarded as the first person to formally postulate that disease was spread by tiny invisible seminaria. He proposed that these seeds could attach themselves to certain objects that supported their transfer from person to person. However, since the technology for seeing such tiny objects did not yet exist, the existence of the seminaria remained hypothetical for a little over a century—an invisible world waiting to be revealed.

Short Answer

Why is Antonie van Leeuwenhoek’s work much better known than that of Zaccharias Janssen?

Why did the cork cells observed by Robert Hooke appear to be empty, as opposed to being full of other structures?

Multiple Choice

Who was the first to describe “cells” in dead cork tissue?

A. Hans Janssen
B. Zaccharias Janssen
C. Antonie van Leeuwenhoek
D. Robert Hooke

Answer

D

Who is the probable inventor of the compound microscope?

A. Girolamo Fracastoro
B. Zaccharias Janssen
C. Antonie van Leeuwenhoek
D. Robert Hooke

Answer

B

Fill in the Blank

A microscope that uses multiple lenses is called a _____ microscope.

Answer

compound

2.3: Instruments of Microscopy

The 20th century saw the development of microscopes that leveraged nonvisible light, such as fluorescence microscopy, which uses an ultraviolet light source, and electron microscopy, which uses short-wavelength electron beams. These advances led to major improvements in magnification, resolution, and contrast. In this section, we survey the broad range of modern microscopic technology and common applications for each type of microscope.

Multiple Choice

Which would be the best choice for viewing internal structures of a living protist such as a *Paramecium*?

A. a brightfield microscope with a stain
B. a brightfield microscope without a stain
C. a darkfield microscope
D. a transmission electron microscope

Answer

C

Which type of microscope is especially useful for viewing thick structures such as biofilms?

A. a transmission electron microscope
B. a scanning electron microscopes
C. a phase-contrast microscope

D. a confocal scanning laser microscope
E. an atomic force microscope

Answer

D

Which type of microscope would be the best choice for viewing very small surface structures of a cell?

A. a transmission electron microscope
B. a scanning electron microscope
C. a brightfield microscope
D. a darkfield microscope
E. a phase-contrast microscope

Answer

B

What type of microscope uses an annular stop?

A. a transmission electron microscope
B. a scanning electron microscope
C. a brightfield microscope
D. a darkfield microscope
E. a phase-contrast microscope

Answer

E

What type of microscope uses a cone of light so that light only hits the specimen indirectly, producing a darker image on a brighter background?

A. a transmission electron microscope
B. a scanning electron microscope
C. a brightfield microscope
D. a darkfield microscope
E. a phase-contrast microscope

Answer

D

Fill in the Blank

Chromophores that absorb and then emit light are called _____.

Answer

fluorochromes

In a(n) _____ microscope, a probe located just above the specimen moves up and down in response to forces between the atoms and the tip of the probe.

Answer

atomic force microscope

What is the total magnification of a specimen that is being viewed with a standard ocular lens and a 40× objective lens?

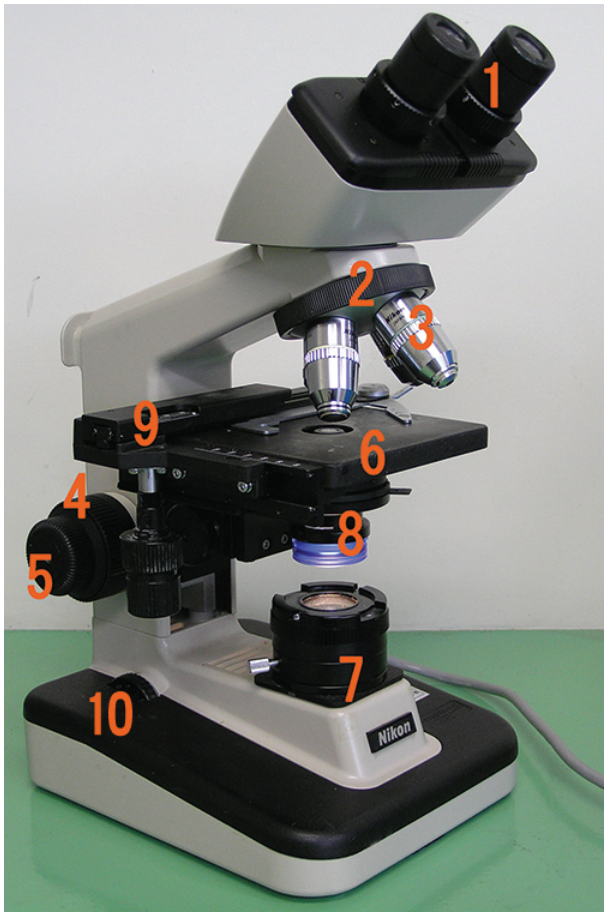
Answer

400×

Short Answer

What is the function of the condenser in a brightfield microscope?

Label each component of the brightfield microscope.



Critical Thinking

When focusing a light microscope, why is it best to adjust the focus using the coarse focusing knob before using the fine focusing knob?

You need to identify structures within a cell using a microscope. However, the image appears very blurry even though you have a high magnification. What are some things that you could try to improve the resolution of the image? Describe the most basic factors that affect resolution when you first put the slide onto the stage; then consider more specific factors that could affect resolution for 40× and 100× lenses.

2.4: Staining Microscopic Specimens

In their natural state, most of the cells and microorganisms that we observe under the microscope lack color and contrast. This makes it difficult, if not impossible, to detect important cellular structures and their distinguishing characteristics without artificially treating specimens. We focus on the most clinically relevant techniques developed to identify specific microbes, cellular structures, DNA sequences, or indicators of infection in tissue samples, under the microscope.

Multiple Choice

What mordant is used in Gram staining?

A. crystal violet
B. safranin
C. acid-alcohol
D. iodine

Answer

D

What is one difference between specimen preparation for a transmission electron microscope (TEM) and preparation for a scanning electron microscope (SEM)?

A. Only the TEM specimen requires sputter coating.
B. Only the SEM specimen requires sputter-coating.
C. Only the TEM specimen must be dehydrated.
D. Only the SEM specimen must be dehydrated.

Answer

B

Fill in the Blank

Ziehl-Neelsen staining, a type of _____ staining, is diagnostic for *Mycobacterium tuberculosis.*

Answer

acid-fast

The _____ is used to differentiate bacterial cells based on the components of their cell walls.

Answer

Gram stain

Short Answer

How could you identify whether a particular bacterial sample contained specimens with mycolic acid-rich cell walls?

Critical Thinking

You use the Gram staining procedure to stain an L-form bacterium (a bacterium that lacks a cell wall). What color will the bacterium be after the staining procedure is finished?

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CHAPTER OVERVIEW

3: Macromolecules

The earth is estimated to be 4.6 billion years old, but for the first 2 billion years, the atmosphere lacked oxygen, without which the earth could not support life as we know it. One hypothesis about how life emerged on earth involves the concept of a “primordial soup.” This idea proposes that life began in a body of water when metals and gases from the atmosphere combined with a source of energy, such as lightning or ultraviolet light, to form the carbon compounds that are the chemical building blocks of life. In 1952, Stanley Miller (1930–2007), a graduate student at the University of Chicago, and his professor Harold Urey (1893–1981), set out to confirm this hypothesis in a now-famous experiment. Miller and Urey combined what they believed to be the major components of the earth’s early atmosphere—water (H_2O), methane (CH_4), hydrogen (H_2), and ammonia (NH_3)—and sealed them in a sterile flask. Next, they heated the flask to produce water vapor and passed electric sparks through the mixture to mimic lightning in the atmosphere (Figure **3.1**). When they analyzed the contents of the flask a week later, they found amino acids, the structural units of proteins—molecules essential to the function of all organisms.

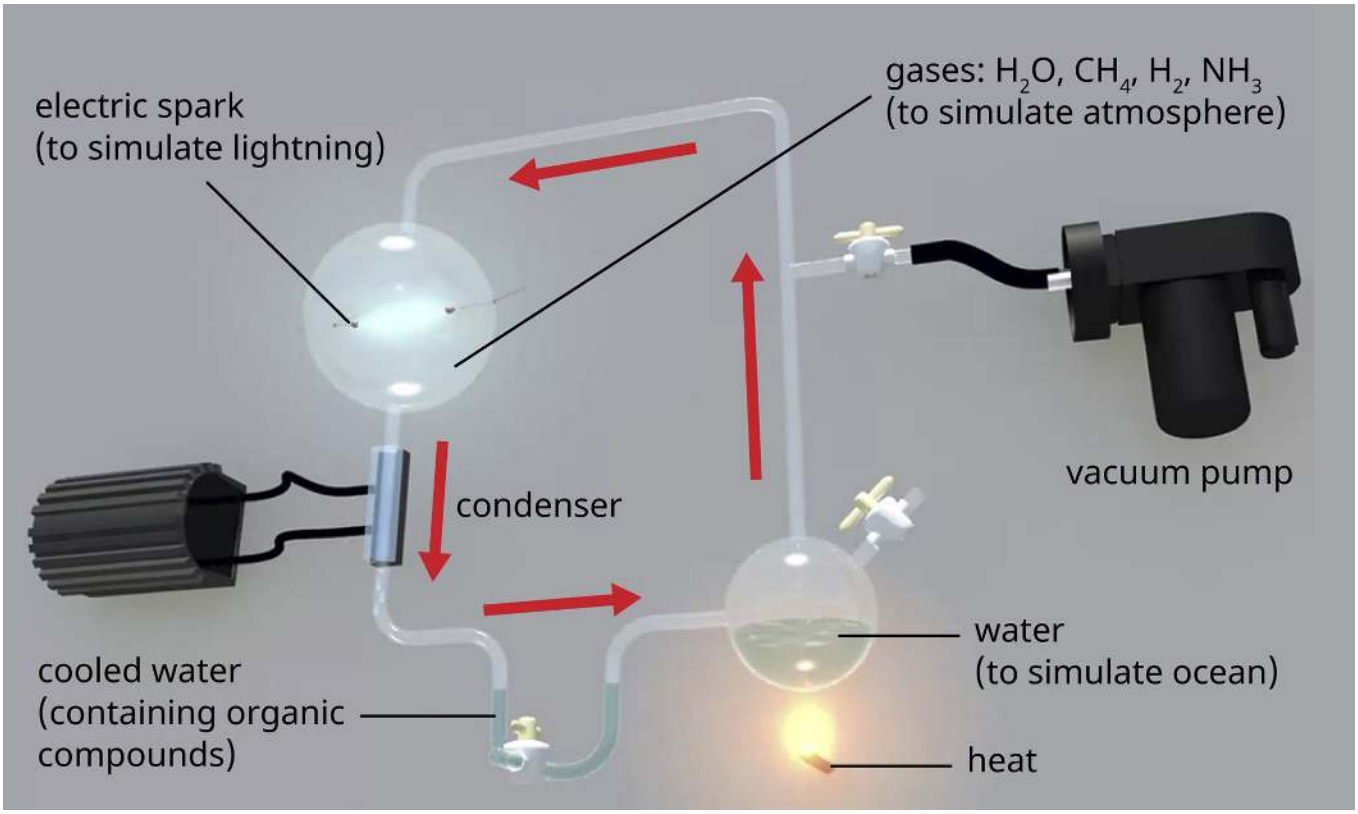


Figure **3.1**: Scientist Stanley Miller and Harold Urey demonstrated that organic compounds may have originated naturally from inorganic matter. The Miller-Urey experiment illustrated here simulated the effects of lightning on chemical compounds found in the earth’s early atmosphere. The resulting reactions yielded amino acids, the chemical building blocks of proteins. (credit: modification of work by Courtney Harrington)

3.1: Organic Molecules

3.2: Isomerism

3.3: Functional Groups and Dehydration Reaction

3.4: Carbohydrates - Monosaccharides and Disaccharides

3.5: Carbohydrates - Polysaccharides

3.6: Lipids - Triacylglycerides

3.7: Lipids - Phospholipids and Membrane

3.8: Proteins - Building Blocks
3.9: Protein Structures
3.10: Using Biochemistry to Identify Microorganisms
3.E: Microbial Biochemistry (Exercises)

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3.1: Organic Molecules

Learning Objectives

- Identify common elements and structures found in organic molecules
- Distinguish between macronutrients and micronutrients (trace elements)
- Differentiate inorganic from organic molecules

Biochemistry is the discipline that studies the chemistry of life, and its objective is to explain form and function based on chemical principles. Organic chemistry is the discipline devoted to the study of carbon-based chemistry, which is the foundation for the study of biomolecules and the discipline of biochemistry. Both biochemistry and organic chemistry are based on the concepts of general chemistry, some of which are presented in Appendix A.

Elements in Living Cells

The most abundant element in cells is hydrogen (H), followed by carbon (C), oxygen (O), nitrogen (N), phosphorous (P), and sulfur (S). We call these elements **macronutrients**, and they account for about 99% of the dry weight of cells. Some elements, such as sodium (Na), potassium (K), magnesium (Mg), zinc (Zn), iron (Fe), calcium (Ca), molybdenum (Mo), copper (Cu), cobalt (Co), manganese (Mn), or vanadium (Va), are required by some cells in very small amounts and are called **micronutrients** or **trace elements**. All of these elements are essential to the function of many biochemical reactions, and, therefore, are essential to life.

The four most abundant elements in living matter (C, N, O, and H) have low atomic numbers and are thus light elements capable of forming strong bonds with other atoms to produce molecules (Figure **3.1.1**). Carbon forms four chemical bonds, whereas nitrogen forms three, oxygen forms two, and hydrogen forms one. When bonded together within molecules, oxygen, sulfur, and nitrogen often have one or more “lone pairs” of electrons that play important roles in determining many of the molecules’ physical and chemical properties (see Appendix A). These traits in combination permit the formation of a vast number of diverse molecular species necessary to form the structures and enable the functions of living organisms.

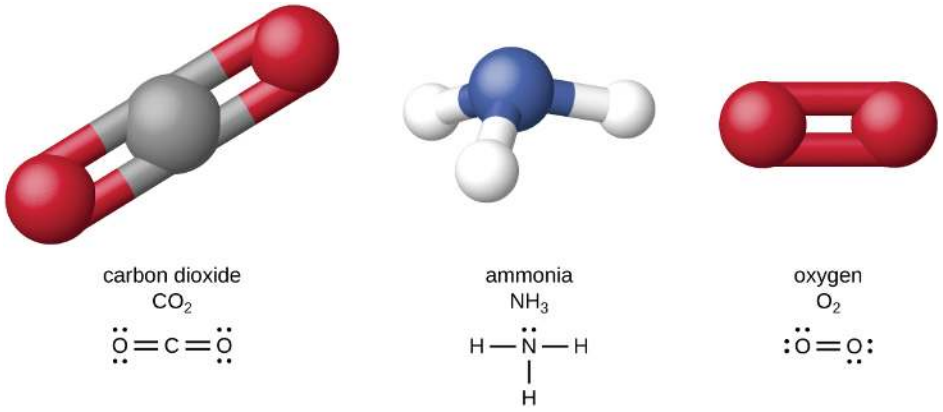


Figure **3.1.1**: Some common molecules include carbon dioxide, ammonia, and oxygen, which consist of combinations of oxygen atoms (red spheres), carbon atoms (gray spheres), hydrogen atoms (white spheres), or nitrogen atoms (blue spheres).

Living organisms contain **inorganic compounds** (mainly water and salts; see Appendix A) and organic molecules. **Organic molecules** contain carbon; inorganic compounds do not. Carbon oxides and carbonates are exceptions; they contain carbon but are considered inorganic because they do not contain hydrogen. The atoms of an organic molecule are typically organized around chains of carbon atoms.

Inorganic compounds make up 1%–1.5% of a living cell’s mass. They are small, simple compounds that play important roles in the cell, although they do not form cell structures. Most of the carbon found in organic molecules originates from inorganic carbon sources such as carbon dioxide captured via carbon fixation by microorganisms.

Key Concepts and Summary

- The most abundant elements in cells are hydrogen, carbon, oxygen, nitrogen, phosphorus, and sulfur.

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3.2: Isomerism

Learning Objectives

- Explain the concept of isomerism
- Distinguish between structural isomers and enantiomers

Organic Molecules

Organic molecules in organisms are generally larger and more complex than inorganic molecules. Their carbon skeletons are held together by covalent bonds. They form the cells of an organism and perform the chemical reactions that facilitate life. All of these molecules, called **biomolecules** because they are part of living matter, contain carbon, which is the building block of life. Carbon is a very unique element in that it has four valence electrons in its outer orbitals and can form four single covalent bonds with up to four other atoms at the same time (see Appendix A). These atoms are usually oxygen, hydrogen, nitrogen, sulfur, phosphorous, and carbon itself; the simplest organic compound is methane, in which carbon binds only to hydrogen (Figure **3.2.1**).

As a result of carbon's unique combination of size and bonding properties, carbon atoms can bind together in large numbers, thus producing a chain or carbon skeleton. The **carbon skeleton** of organic molecules can be straight, branched, or ring shaped (cyclic). Organic molecules are built on chains of carbon atoms of varying lengths; most are typically very long, which allows for a huge number and variety of compounds. No other element has the ability to form so many different molecules of so many different sizes and shapes.

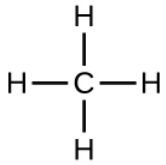


Figure **3.2.1**: A carbon atom can bond with up to four other atoms. The simplest organic molecule is methane (CH_4), depicted here.

Isomerism

Molecules with the same atomic makeup but different structural arrangement of atoms are called **isomers**. The concept of isomerism is very important in chemistry because the structure of a molecule is always directly related to its function. Slight changes in the structural arrangements of atoms in a molecule may lead to very different properties. Chemists represent molecules by their structural formula, which is a graphic representation of the molecular structure, showing how the atoms are arranged. Compounds that have identical molecular formulas but differ in the bonding sequence of the atoms are called **structural isomers**. The monosaccharides glucose, galactose, and fructose all have the same molecular formula, $C_6H_{12}O_6$, but we can see from Figure **3.2.2** that the atoms are bonded together differently.

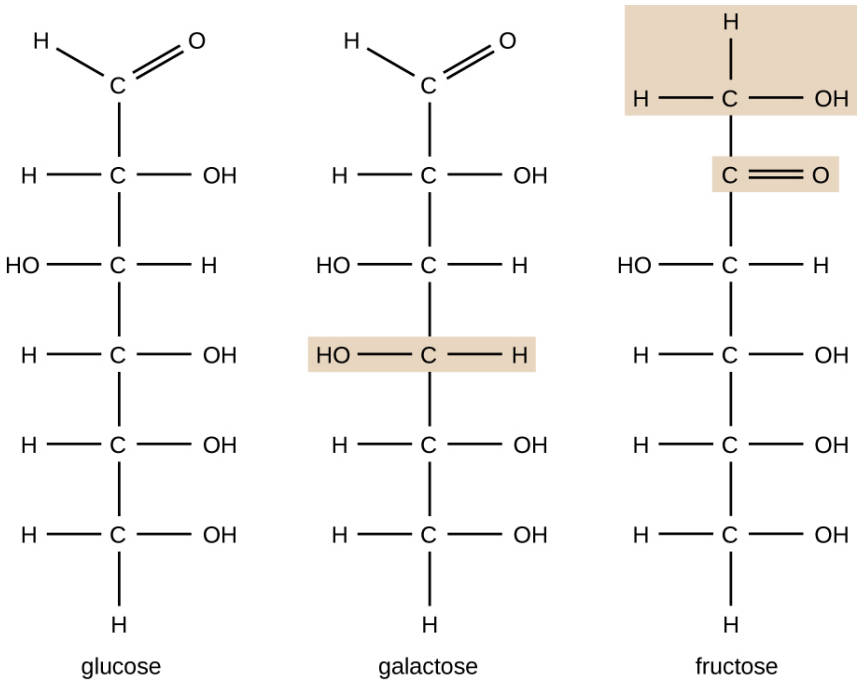


Figure **3.2.2**: Glucose, galactose, and fructose have the same chemical formula ($C_6H_{12}O_6$), but these structural isomers differ in their physical and chemical properties.

Isomers that differ in the spatial arrangements of atoms are called stereoisomers; one unique type is **enantiomers**. The properties of enantiomers were originally discovered by Louis Pasteur in 1848 while using a microscope to analyze crystallized fermentation products of wine. Enantiomers are molecules that have the characteristic of chirality, in which their structures are nonsuperimposable mirror images of each other. Chirality is an important characteristic in many biologically important molecules, as illustrated by the examples of structural differences in the enantiomeric forms of the monosaccharide glucose or the amino acid alanine (Figure **3.2.3**).

Many organisms are only able to use one enantiomeric form of certain types of molecules as nutrients and as building blocks to make structures within a cell. Some enantiomeric forms of amino acids have distinctly different tastes and smells when consumed as food. For example, L-aspartame, commonly called aspartame, tastes sweet, whereas D-aspartame is tasteless. Drug enantiomers can have very different pharmacologic affects. For example, the compound methorphan exists as two enantiomers, one of which acts as an antitussive (*dextro*methorphan, a cough suppressant), whereas the other acts as an analgesic (*levo*methorphan, a drug similar in effect to codeine).

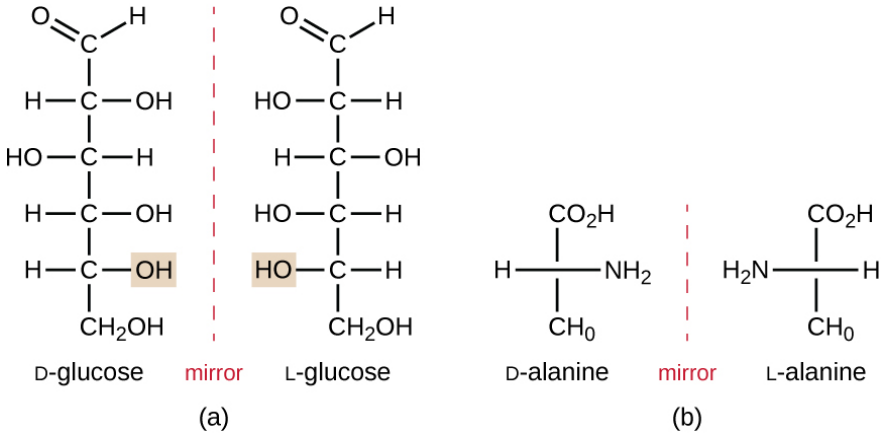


Figure **3.2.3**: Enantiomers are stereoisomers that exhibit chirality. Their chemical structures are nonsuperimposable mirror images of each other. (a) D-glucose and L-glucose are monosaccharides that are enantiomers. (b) The enantiomers D-alanine and L-alanine are enantiomers found in bacterial cell walls and human cells, respectively.

Enantiomers are also called **optical isomers** because they can rotate the plane of polarized light. Some of the crystals Pasteur observed from wine fermentation rotated light clockwise whereas others rotated the light counterclockwise. Today, we denote enantiomers that rotate polarized light clockwise (+) as *d* forms, and the mirror image of the same molecule that rotates polarized light counterclockwise (−) as the *l* form. The *d* and *l* labels are derived from the Latin words *dexter* (on the right) and *laevus* (on the left), respectively. These two different optical isomers often have very different biological properties and activities. Certain species of molds, yeast, and bacteria, such as *Rhizopus*, *Yarrowia*, and *Lactobacillus* spp., respectively, can only metabolize one type of optical isomer; the opposite isomer is not suitable as a source of nutrients. Another important reason to be aware of optical isomers is the therapeutic use of these types of chemicals for drug treatment, because some microorganisms can only be affected by one specific optical isomer.

Key Concepts and Summary

- Life is carbon based. Each carbon atom can bind to another one producing a **carbon skeleton** that can be straight, branched, or ring shaped.
- The same numbers and types of atoms may bond together in different ways to yield different molecules called **isomers**. Isomers may differ in the bonding sequence of their atoms (**structural isomers**) or in the spatial arrangement of atoms whose bonding sequences are the same (**stereoisomers**), and their physical and chemical properties may vary slightly or drastically.

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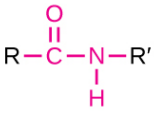
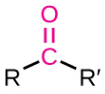
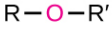
3.3: Functional Groups and Dehydration Reaction

Learning Objectives

- Identifying examples of functional groups
- Describe the roles of functional groups in synthesizing polymers

Biologically Significant Functional Groups

In addition to containing carbon atoms, biomolecules also contain **functional groups** — groups of atoms within molecules that are categorized by their specific chemical composition and the chemical reactions they perform, regardless of the molecule in which the group is found. Some of the most common functional groups are listed in Figure **3.3.1**. In the formulas, the symbol R stands for "residue" and represents the remainder of the molecule. R might symbolize just a single hydrogen atom or it may represent a group of many atoms. Notice that some functional groups are relatively simple, consisting of just one or two atoms, while some comprise two of these simpler functional groups. For example, a carbonyl group is a functional group composed of a carbon atom double bonded to an oxygen atom: C=O. It is present in several classes of organic compounds as part of larger functional groups such as ketones, aldehydes, carboxylic acids, and amides. In ketones, the carbonyl is present as an internal group, whereas in aldehydes it is a terminal group.

Common Functional Groups Found in Biomolecules		
Name	**Functional Group**	**Compounds**
Aldehyde		Carbohydrates
Amide		Proteins
Amino		Amino acids, proteins
Carbonyl		Ketones, aldehydes, carboxylic acids, amides
Carboxylic acid		Amino acids, proteins, fatty acids
Ester		Lipids, nucleic acids
Ether		Disaccharides, polysaccharides, lipids
Hydroxyl		Alcohols, monosaccharides, amino acids, nucleic acids
Ketone		Carbohydrates
Methyl		Methylated compounds such as methyl alcohols and methyl esters
Phosphate		Nucleic acids, phospholipids, ATP
Sulfhydryl		Amino acids, proteins

*Functional groups are represented in pink. Ketone and aldehyde both contain a carbonyl group, highlighted in blue.

Figure **3.3.1**: Functional groups.

Macromolecule Synthesis: Dehydration Reaction

Carbon chains form the skeletons of most organic molecules. Functional groups combine with the chain to form biomolecules. Because these biomolecules are typically large, we call them **macromolecules**. Many biologically relevant macromolecules are formed by linking together a great number of identical, or very similar, smaller organic molecules. The smaller molecules act as

building blocks and are called **monomers**, and the macromolecules that result from their linkage are called **polymers**. Cells and cell structures include four main groups of carbon-containing macromolecules: polysaccharides, proteins, lipids, and nucleic acids. The first three groups of molecules will be studied throughout this chapter. The biochemistry of nucleic acids will be discussed in Biochemistry of the Genome.

Of the many possible ways that monomers may be combined to yield polymers, one common approach encountered in the formation of biological macromolecules is **dehydration synthesis**. In this chemical reaction, monomer molecules bind end to end in a process that results in the formation of water molecules as a byproduct:

$$\text{H—monomer—OH} + \text{H—monomer—OH} \longrightarrow \text{H—monomer—monomer—OH} + \text{H}_2\text{O} \tag{3.3.1}$$

Figure **3.3.2** shows dehydration synthesis of glucose binding together to form maltose and a water molecule. Table **3.3.1** summarizes macromolecules and some of their functions.

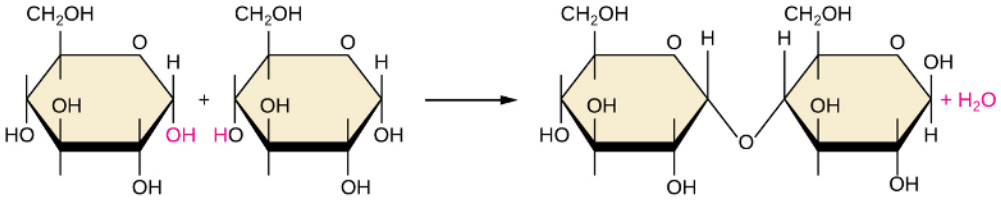


Figure **3.3.2**: In this dehydration synthesis reaction, two molecules of glucose are linked together to form maltose. In the process, a water molecule is formed.

Table **3.3.1**: Functions of Macromolecules

Macromolecule	Functions
Carbohydrates	Energy storage, receptors, food, structural role in plants, fungal cell walls, exoskeletons of insects
Lipids	Energy storage, membrane structure, insulation, hormones, pigments
Nucleic acids	Storage and transfer of genetic information
Proteins	Enzymes, structure, receptors, transport, structural role in the cytoskeleton of a cell and the extracellular matrix

Key Concepts and Summary

- **Functional groups** confer specific chemical properties to molecules bearing them. Common functional groups in biomolecules are hydroxyl, methyl, carbonyl, carboxyl, amino, phosphate, and sulfhydryl.
- **Macromolecules** are **polymers** assembled from individual units, the **monomers**, which bind together like building blocks. Many biologically significant macromolecules are formed by **dehydration synthesis**, a process in which monomers bind together by combining their functional groups and generating water molecules as byproducts.

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3.4: Carbohydrates - Monosaccharides and Disaccharides

Learning Objectives

- Give examples of monosaccharides and polysaccharides
- Describe the function of the simple sugars within a cell

The most abundant biomolecules on earth are **carbohydrates**. From a chemical viewpoint, carbohydrates are primarily a combination of carbon and water, and many of them have the empirical formula $(CH_2O)_n$, where *n* is the number of repeated units. This view represents these molecules simply as "hydrated" carbon atom chains in which water molecules attach to each carbon atom, leading to the term "carbohydrates." Although all carbohydrates contain carbon, hydrogen, and oxygen, there are some that also contain nitrogen, phosphorus, and/or sulfur. Carbohydrates have myriad different functions. They are abundant in terrestrial ecosystems, many forms of which we use as food sources. These molecules are also vital parts of macromolecular structures that store and transmit genetic information (i.e., DNA and RNA). They are the basis of biological polymers that impart strength to various structural components of organisms (e.g., cellulose and chitin), and they are the primary source of energy storage in the form of starch and glycogen.

Monosaccharides: The Sweet Ones

In biochemistry, carbohydrates are often called **saccharides**, from the Greek *sakcharon*, meaning sugar, although not all the saccharides are sweet. The simplest carbohydrates are called **monosaccharides**, or simple sugars. They are the building blocks (monomers) for the synthesis of polymers or complex carbohydrates, as will be discussed further in this section. Monosaccharides are classified based on the number of carbons in the molecule. General categories are identified using a prefix that indicates the number of carbons and the suffix *–ose*, which indicates a saccharide; for example, triose (three carbons), tetrose (four carbons), pentose (five carbons), and hexose (six carbons) (Figure **3.4.1**). The hexose D-glucose is the most abundant monosaccharide in nature. Other very common and abundant hexose monosaccharides are galactose, used to make the disaccharide milk sugar lactose, and the fruit sugar fructose.

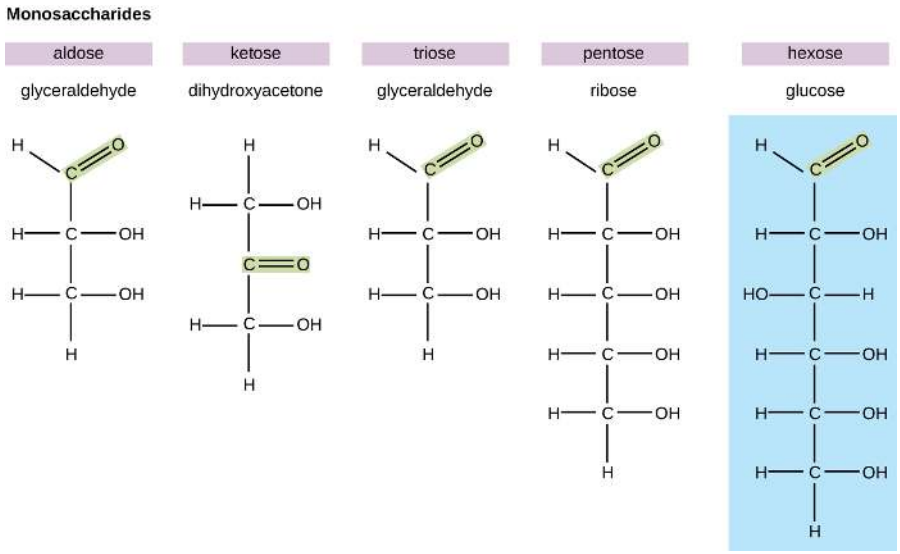


Figure **3.4.1**: Monosaccharides are classified based on the position of the carbonyl group and the number of carbons in the backbone.

Monosaccharides of four or more carbon atoms are typically more stable when they adopt cyclic, or ring, structures. These ring structures result from a chemical reaction between functional groups on opposite ends of the sugar's flexible carbon chain, namely the carbonyl group and a relatively distant hydroxyl group. Glucose, for example, forms a six-membered ring (Figure **3.4.2**).

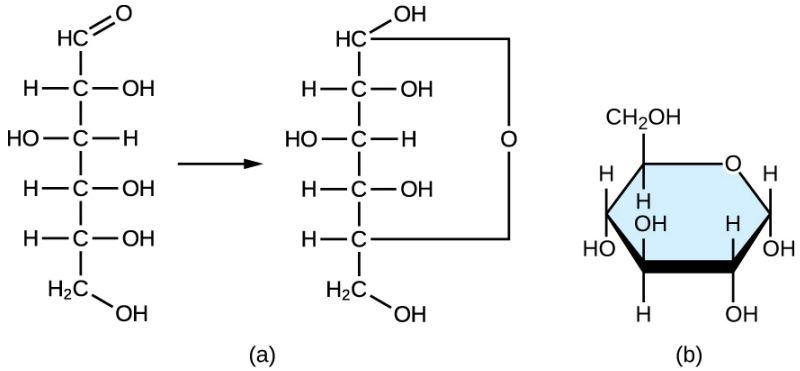


Figure **3.4.2**: (a) A linear monosaccharide (glucose in this case) forms a cyclic structure. (b) This illustration shows a more realistic depiction of the cyclic monosaccharide structure. Note in these cyclic structural diagrams, the carbon atoms composing the ring are not explicitly shown.

Disaccharides

Two monosaccharide molecules may chemically bond to form a **disaccharide**. The name given to the covalent bond between the two monosaccharides is a glycosidic bond. **Glycosidic bonds** form between hydroxyl groups of the two saccharide molecules, an example of the dehydration synthesis described in the previous section of this chapter:

$$\text{monosaccharide—OH} + \text{HO—monosaccharide} \longrightarrow \underbrace{\text{monosaccharide—O—monosaccharide}}_{\text{disaccharide}} \quad (3.4.1)$$

Common disaccharides are the grain sugar maltose, made of two glucose molecules; the milk sugar lactose, made of a galactose and a glucose molecule; and the table sugar sucrose, made of a glucose and a fructose molecule (Figure **3.4.3**).

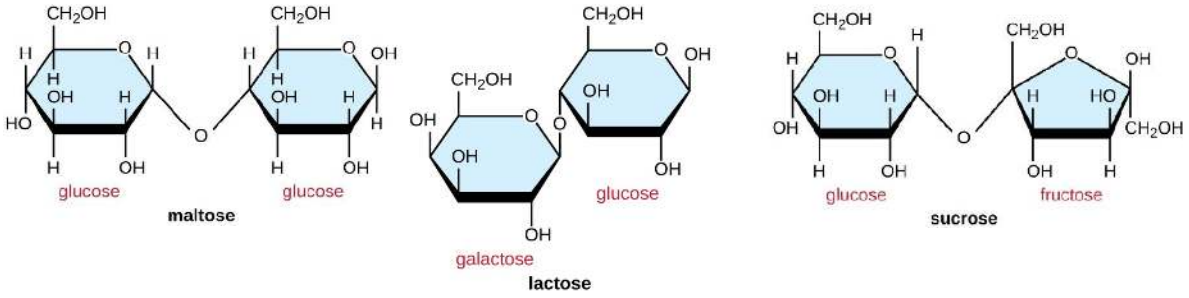


Figure **3.4.3**: Common disaccharides include maltose, lactose, and sucrose.

Key Concepts and Summary

- **Carbohydrates**, the most abundant biomolecules on earth, are widely used by organisms for structural and energy-storage purposes.
- Carbohydrates include individual sugar molecules (**monosaccharides**) as well as two or more molecules chemically linked by **glycosidic bonds**. **Monosaccharides** are classified based on the number of carbons in the molecule as trioses (3 C), tetroses (4 C), pentoses (5 C), and hexoses (6 C). They are the building blocks for the synthesis of polymers or complex carbohydrates.
- **Disaccharides** such as sucrose, lactose, and maltose are molecules composed of two monosaccharides linked together by a glycosidic bond.

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3.5: Carbohydrates - Polysaccharides

Learning Objectives

- Describe the structures of starch, glycogen and cellulose
- Describe the functions of these polysaccharides

Polysaccharides

Polysaccharides, also called glycans, are large polymers composed of hundreds of monosaccharide monomers. Unlike mono- and disaccharides, polysaccharides are not sweet and, in general, they are not soluble in water. Like disaccharides, the monomeric units of polysaccharides are linked together by glycosidic bonds.

Polysaccharides are very diverse in their structure. Three of the most biologically important polysaccharides — **starch**, **glycogen**, and **cellulose** — are all composed of repetitive glucose units, although they differ in their structure (Figure **3.5.1**). Cellulose consists of a linear chain of glucose molecules and is a common structural component of cell walls in plants and other organisms. Glycogen and starch are branched polymers; glycogen is the primary energy-storage molecule in animals and bacteria, whereas plants primarily store energy in starch. The orientation of the glycosidic linkages in these three polymers is different as well and, as a consequence, linear and branched macromolecules have different properties.

Modified glucose molecules can be fundamental components of other structural polysaccharides. Examples of these types of structural polysaccharides are N-acetyl glucosamine (NAG) and N-acetyl muramic acid (NAM) found in bacterial cell wall peptidoglycan. Polymers of NAG form chitin, which is found in fungal cell walls and in the exoskeleton of insects.

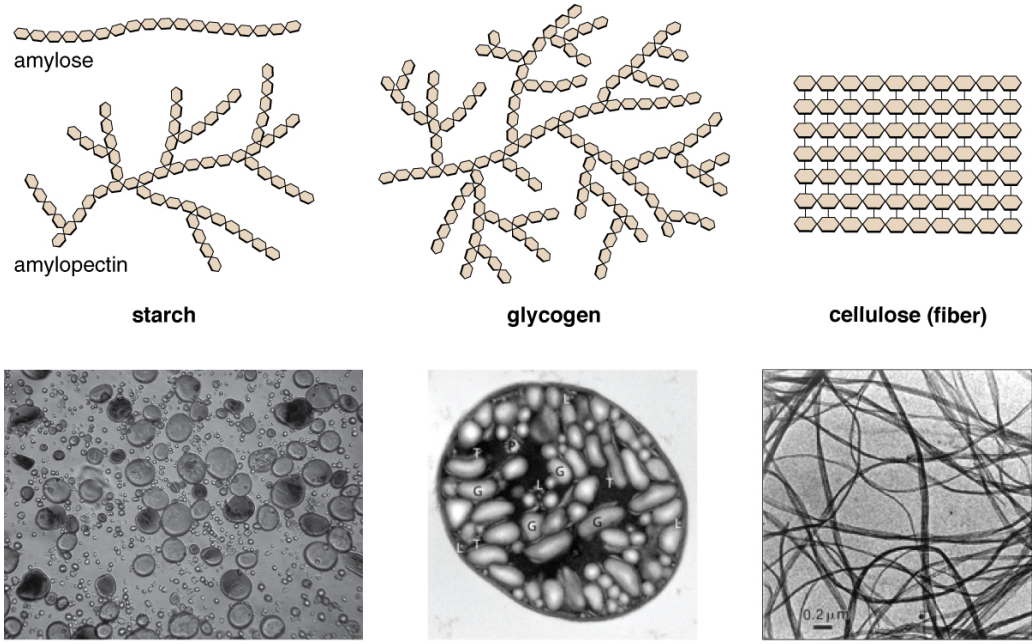


Figure **3.5.1**: Starch, glycogen, and cellulose are three of the most important polysaccharides. In the top row, hexagons represent individual glucose molecules. Micrographs (bottom row) show wheat starch granules stained with iodine (left), glycogen granules (G) inside the cell of a cyanobacterium (middle), and bacterial cellulose fibers (right). (credit “iodine granules”: modification of work by Kiselov Yuri; credit “glycogen granules”: modification of work by Stöckel J, Elvitigala TR, Liberton M, Pakrasi HB; credit “cellulose”: modification of work by American Society for Microbiology)

Key Concepts and Summary

- **Polysaccharides**, or **glycans**, are polymers composed of hundreds of monosaccharide monomers linked together by glycosidic bonds. The energy-storage polymers **starch** and **glycogen** are examples of polysaccharides and are all composed of branched chains of glucose molecules.

- The polysaccharide **cellulose** is a common structural component of the cell walls of organisms. Other structural polysaccharides, such as N-acetyl glucosamine (NAG) and N-acetyl muramic acid (NAM), incorporate modified glucose molecules and are used in the construction of peptidoglycan or chitin.

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3.6: Lipids - Triacylglycerides

3.06: Lipids - Triacylglycerides

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3.7: Lipids - Phospholipids and Membrane

Learning Objectives

- Describe the chemical composition of lipids
- Describe the unique characteristics and diverse structures of lipids
- Compare and contrast triacylglycerides (triglycerides) and phospholipids
- Describe how phospholipids are used to construct biological membranes

Although they are composed primarily of carbon and hydrogen, lipid molecules may also contain oxygen, nitrogen, sulfur, and phosphorous. Lipids serve numerous and diverse purposes in the structure and functions of organisms. They can be a source of nutrients, a storage form for carbon, energy-storage molecules, or structural components of membranes and hormones. Lipids comprise a broad class of many chemically distinct compounds, the most common of which are discussed in this section.

Fatty Acids and Triacylglycerides

The fatty acids are lipids that contain long-chain hydrocarbons terminated with a carboxylic acid functional group. Because of the long hydrocarbon chain, fatty acids are hydrophobic ("water fearing") or nonpolar. Fatty acids with hydrocarbon chains that contain only single bonds are called saturated fatty acids because they have the greatest number of hydrogen atoms possible and are, therefore, "saturated" with hydrogen. Fatty acids with hydrocarbon chains containing at least one double bond are called unsaturated fatty acids because they have fewer hydrogen atoms. Saturated fatty acids have a straight, flexible carbon backbone, whereas unsaturated fatty acids have "kinks" in their carbon skeleton because each double bond causes a rigid bend of the carbon skeleton. These differences in saturated versus unsaturated fatty acid structure result in different properties for the corresponding lipids in which the fatty acids are incorporated. For example, lipids containing saturated fatty acids are solids at room temperature, whereas lipids containing unsaturated fatty acids are liquids.

A triacylglycerol, or triglyceride, is formed when three fatty acids are chemically linked to a glycerol molecule (Figure **3.7.1**). Triglycerides are the primary components of adipose tissue (body fat), and are major constituents of sebum (skin oils). They play an important metabolic role, serving as efficient energy-storage molecules that can provide more than double the caloric content of both carbohydrates and proteins.

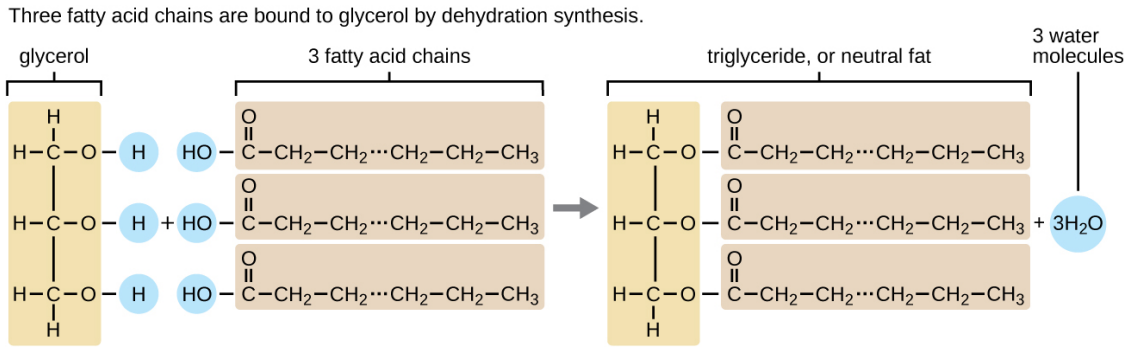


Figure **3.7.1**: Triglycerides are composed of a glycerol molecule attached to three fatty acids by a dehydration synthesis reaction.

Phospholipids and Biological Membranes

Triglycerides are classified as simple lipids because they are formed from just two types of compounds: glycerol and fatty acids. In contrast, complex lipids contain at least one additional component, for example, a phosphate group (phospholipids) or a carbohydrate moiety (glycolipids). Figure **3.7.2** depicts a typical phospholipid composed of two fatty acids linked to glycerol (a diglyceride). The two fatty acid carbon chains may be both saturated, both unsaturated, or one of each. Instead of another fatty acid molecule (as for triglycerides), the third binding position on the glycerol molecule is occupied by a modified phosphate group.

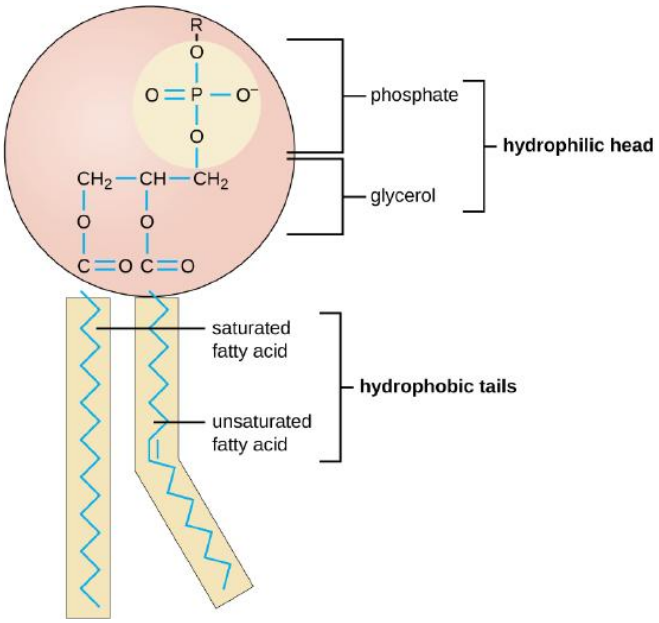


Figure **3.7.2**: This illustration shows a phospholipid with two different fatty acids, one saturated and one unsaturated, bonded to the glycerol molecule. The unsaturated fatty acid has a slight kink in its structure due to the double bond.

The molecular structure of lipids results in unique behavior in aqueous environments. Figure **3.7.1** depicts the structure of a triglyceride. Because all three substituents on the glycerol backbone are long hydrocarbon chains, these compounds are nonpolar and not significantly attracted to polar water molecules—they are hydrophobic. Conversely, phospholipids such as the one shown in Figure **3.7.2** have a negatively charged phosphate group. Because the phosphate is charged, it is capable of strong attraction to water molecules and thus is hydrophilic, or "water loving." The hydrophilic portion of the phospholipid is often referred to as a polar "head," and the long hydrocarbon chains as nonpolar "tails." A molecule presenting a hydrophobic portion and a hydrophilic moiety is said to be amphipathic. Notice the "R" designation within the hydrophilic head depicted in Figure **3.7.2**, indicating that a polar head group can be more complex than a simple phosphate moiety. Glycolipids are examples in which carbohydrates are bonded to the lipids' head groups.

The amphipathic nature of phospholipids enables them to form uniquely functional structures in aqueous environments. As mentioned, the polar heads of these molecules are strongly attracted to water molecules, and the nonpolar tails are not. Because of their considerable lengths, these tails are, in fact, strongly attracted to one another. As a result, energetically stable, large-scale assemblies of phospholipid molecules are formed in which the hydrophobic tails congregate within enclosed regions, shielded from contact with water by the polar heads (Figure **3.7.3**). The simplest of these structures are micelles, spherical assemblies containing a hydrophobic interior of phospholipid tails and an outer surface of polar head groups. Larger and more complex structures are created from lipid-bilayer sheets, or unit membranes, which are large, two-dimensional assemblies of phospholipids congregated tail to tail. The cell membranes of nearly all organisms are made from lipid-bilayer sheets, as are the membranes of many intracellular components. These sheets may also form lipid-bilayer spheres that are the structural basis of vesicles and liposomes, subcellular components that play a role in numerous physiological functions.

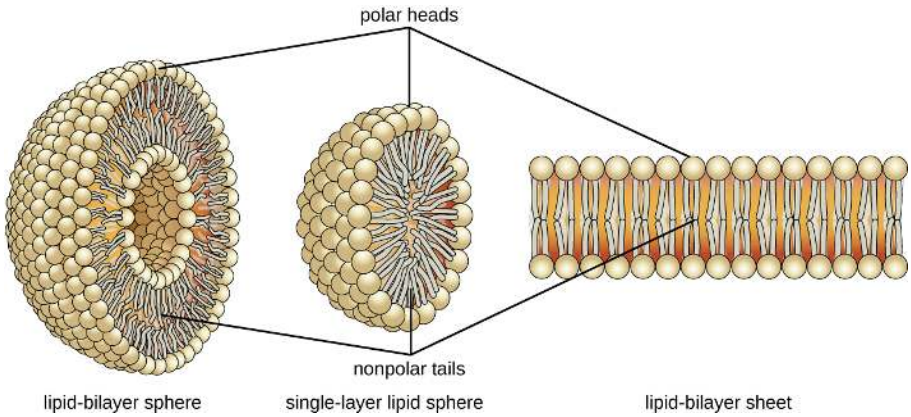


Figure **3.7.3**: Phospholipids tend to arrange themselves in aqueous solution forming liposomes, micelles, or lipid bilayer sheets. (credit: modification of work by Mariana Ruiz Villarreal)

Isoprenoids and Sterols

The isoprenoids are branched lipids, also referred to as terpenoids, that are formed by chemical modifications of the isoprene molecule (Figure **3.7.4**). These lipids play a wide variety of physiological roles in plants and animals, with many technological uses as pharmaceuticals (capsaicin), pigments (e.g., orange beta carotene, xanthophylls), and fragrances (e.g., menthol, camphor, limonene [lemon fragrance], and pinene [pine fragrance]). Long-chain isoprenoids are also found in hydrophobic oils and waxes. Waxes are typically water resistant and hard at room temperature, but they soften when heated and liquefy if warmed adequately. In humans, the main wax production occurs within the sebaceous glands of hair follicles in the skin, resulting in a secreted material called sebum, which consists mainly of triacylglycerol, wax esters, and the hydrocarbon squalene. There are many bacteria in the microbiota on the skin that feed on these lipids. One of the most prominent bacteria that feed on lipids is *Propionibacterium acnes*, which uses the skin's lipids to generate short-chain fatty acids and is involved in the production of acne.

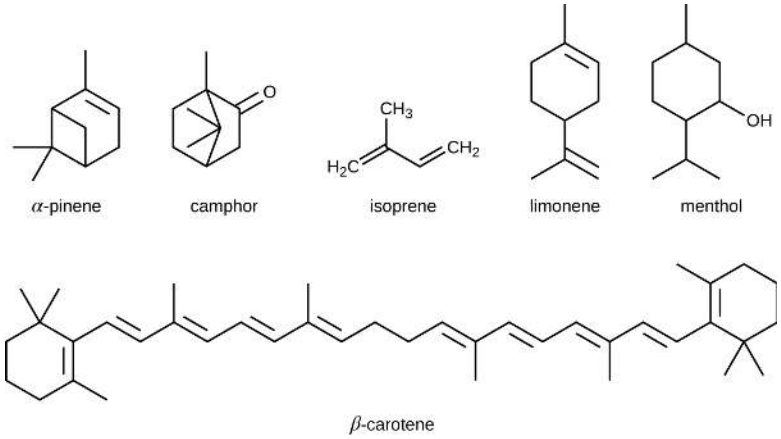
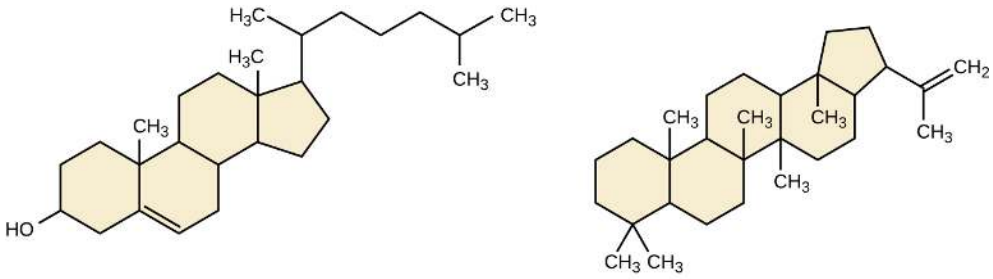


Figure **3.7.4**: Five-carbon isoprene molecules are chemically modified in various ways to yield isoprenoids.

Another type of lipids are steroids, complex, ringed structures that are found in cell membranes; some function as hormones. The most common types of steroids are sterols, which are steroids containing an OH group. These are mainly hydrophobic molecules, but also have hydrophilic hydroxyl groups. The most common sterol found in animal tissues is cholesterol. Its structure consists of four rings with a double bond in one of the rings, and a hydroxyl group at the sterol-defining position. The function of cholesterol is to strengthen cell membranes in eukaryotes and in bacteria without cell walls, such as *Mycoplasma*. Prokaryotes generally do not produce cholesterol, although bacteria produce similar compounds called hopanoids, which are also multiringed structures that strengthen bacterial membranes (Figure **3.7.5**). Fungi and some protozoa produce a similar compound called ergosterol, which strengthens the cell membranes of these organisms.



cholesterol hopene

Figure 3.7.5: Cholesterol and hopene (a hopanoid compound) are molecules that reinforce the structure of the cell membranes in eukaryotes and prokaryotes, respectively.

Link to Learning: Liposomes

This video provides additional information about phospholipids and liposomes.

Key Concepts and Summary

- **Lipids** are composed mainly of carbon and hydrogen, but they can also contain oxygen, nitrogen, sulfur, and phosphorous. They provide nutrients for organisms, store carbon and energy, play structural roles in membranes, and function as hormones, pharmaceuticals, fragrances, and pigments.
- Fatty acids are long-chain hydrocarbons with a carboxylic acid functional group. Their relatively long nonpolar hydrocarbon chains make them **hydrophobic**. Fatty acids with no double bonds are **saturated**; those with double bonds are **unsaturated**.
- Fatty acids chemically bond to glycerol to form structurally essential lipids such as **triglycerides** and **phospholipids.** Triglycerides comprise three fatty acids bonded to glycerol, yielding a hydrophobic molecule. Phospholipids contain both hydrophobic hydrocarbon chains and polar head groups, making them **amphipathic** and capable of forming uniquely functional large scale structures.
- Biological membranes are large-scale structures based on phospholipid bilayers that provide hydrophilic exterior and interior surfaces suitable for aqueous environments, separated by an intervening hydrophobic layer. These bilayers are the structural basis for cell membranes in most organisms, as well as subcellular components such as vesicles.
- **Isoprenoids** are lipids derived from isoprene molecules that have many physiological roles and a variety of commercial applications.
- A wax is a long-chain isoprenoid that is typically water resistant; an example of a wax-containing substance is sebum, produced by sebaceous glands in the skin. **Steroids** are lipids with complex, ringed structures that function as structural components of cell membranes and as hormones. **Sterols** are a subclass of steroids containing a hydroxyl group at a specific location on one of the molecule's rings; one example is cholesterol.
- Bacteria produce hopanoids, structurally similar to cholesterol, to strengthen bacterial membranes. Fungi and protozoa produce a strengthening agent called ergosterol.

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3.8: Proteins - Building Blocks

3.09: Proteins - Building Blocks

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3.9: Protein Structures

3.10: Protein Structures

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3.10: Using Biochemistry to Identify Microorganisms

Learning Objectives

- Describe examples of biosynthesis products within a cell that can be detected to identify bacteria
- Explain the principles and workflow of MALDI-TOF mass spectrometry
- Interpret how lipid profile can be used to identify bacteria by FAME analysis

Using Biochemistry to Identify Microorganisms

Accurate identification of bacterial isolates is essential in a clinical microbiology laboratory because the results often inform decisions about treatment that directly affect patient outcomes. For example, cases of food poisoning require accurate identification of the causative agent so that physicians can prescribe appropriate treatment. Likewise, it is important to accurately identify the causative pathogen during an outbreak of disease so that appropriate strategies can be employed to contain the epidemic.

There are many ways to detect, characterize, and identify microorganisms. Some methods rely on phenotypic biochemical characteristics, while others use genotypic identification. The biochemical characteristics of a bacterium provide many traits that are useful for classification and identification. Analyzing the nutritional and metabolic capabilities of the bacterial isolate is a common approach for determining the genus and the species of the bacterium. Some of the most important metabolic pathways that bacteria use to survive will be discussed in Microbial Metabolism. In this section, we will discuss a few methods that use biochemical characteristics to identify microorganisms.

Some microorganisms store certain compounds as granules within their cytoplasm, and the contents of these granules can be used for identification purposes. For example, poly-β-hydroxybutyrate (PHB) is a carbon- and energy-storage compound found in some nonfluorescent bacteria of the genus *Pseudomonas*. Different species within this genus can be classified by the presence or the absence of PHB and fluorescent pigments. The human pathogen *P. aeruginosa* and the plant pathogen *P. syringae* are two examples of fluorescent *Pseudomonas* species that do not accumulate PHB granules.

Other systems rely on biochemical characteristics to identify microorganisms by their biochemical reactions, such as carbon utilization and other metabolic tests. In small laboratory settings or in teaching laboratories, those assays are carried out using a limited number of test tubes. However, more modern systems, such as the one developed by Biolog, Inc., are based on panels of biochemical reactions performed simultaneously and analyzed by software. Biolog's system identifies cells based on their ability to metabolize certain biochemicals and on their physiological properties, including pH and chemical sensitivity. It uses all major classes of biochemicals in its analysis. Identifications can be performed manually or with the semi- or fully automated instruments.

Another automated system identifies microorganisms by determining the specimen's mass spectrum and then comparing it to a database that contains known mass spectra for thousands of microorganisms. This method is based on matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF) and uses disposable MALDI plates on which the microorganism is mixed with a specialized matrix reagent (Figure 3.10.1). The sample/reagent mixture is irradiated with a high-intensity pulsed ultraviolet laser, resulting in the ejection of gaseous ions generated from the various chemical constituents of the microorganism. These gaseous ions are collected and accelerated through the mass spectrometer, with ions traveling at a velocity determined by their mass-to-charge ratio (m/z), thus, reaching the detector at different times. A plot of detector signal versus m/z yields a mass spectrum for the organism that is uniquely related to its biochemical composition. Comparison of the mass spectrum to a library of reference spectra obtained from identical analyses of known microorganisms permits identification of the unknown microbe.

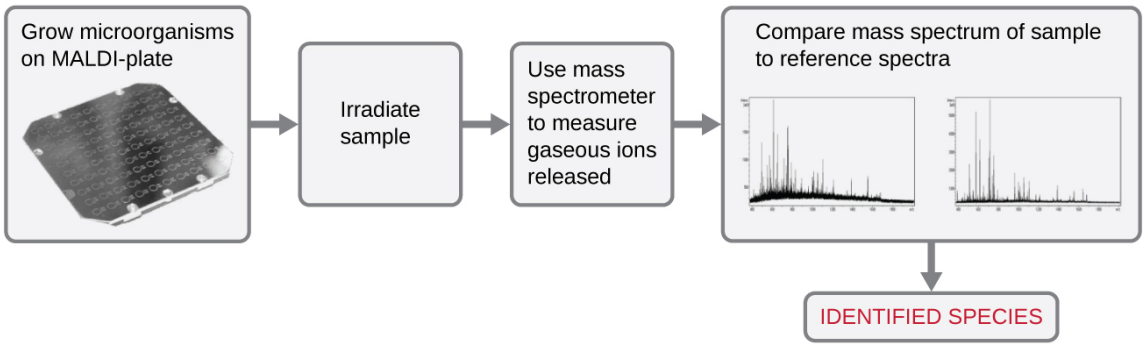


Figure 3.10.1: MALDI-TOF methods are now routinely used for diagnostic procedures in clinical microbiology laboratories. This technology is able to rapidly identify some microorganisms that cannot be readily identified by more traditional methods. (credit "MALDI plate photo": modification of work by Chen Q, Liu T, Chen G; credit "graphs": modification of work by Bailes J, Vidal L, Ivanov DA, Soloviev M)

Microbes can also be identified by measuring their unique lipid profiles. As we have learned, fatty acids of lipids can vary in chain length, presence or absence of double bonds, and number of double bonds, hydroxyl groups, branches, and rings. To identify a microbe by its lipid composition, the fatty acids present in their membranes are analyzed. A common biochemical analysis used for this purpose is a technique used in clinical, public health, and food laboratories. It relies on detecting unique differences in fatty acids and is called fatty acid methyl ester (FAME) analysis. In a FAME analysis, fatty acids are extracted from the membranes of microorganisms, chemically altered to form volatile methyl esters, and analyzed by gas chromatography (GC). The resulting GC chromatogram is compared with reference chromatograms in a database containing data for thousands of bacterial isolates to identify the unknown microorganism (Figure 3.10.2).

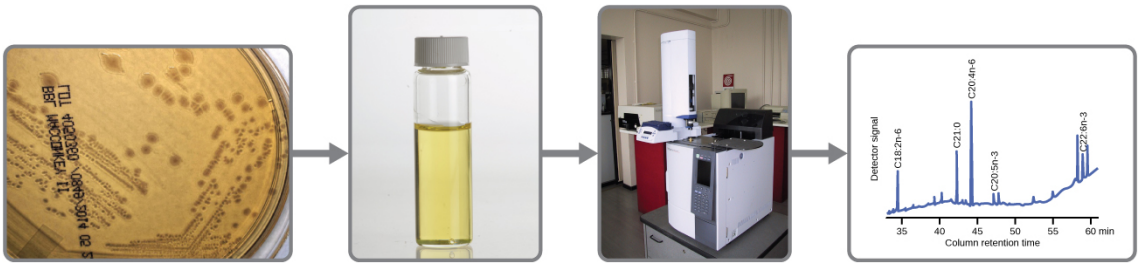


Figure 3.10.2: Fatty acid methyl ester (FAME) analysis in bacterial identification results in a chromatogram unique to each bacterium. Each peak in the gas chromatogram corresponds to a particular fatty acid methyl ester and its height is proportional to the amount present in the cell. (credit "culture": modification of work by the Centers for Disease Control and Prevention; credit "graph": modification of work by Zhang P. and Liu P.)

A related method for microorganism identification is called phospholipid-derived fatty acids (PLFA) analysis. Membranes are mostly composed of phospholipids, which can be saponified (hydrolyzed with alkali) to release the fatty acids. The resulting fatty acid mixture is then subjected to FAME analysis, and the measured lipid profiles can be compared with those of known microorganisms to identify the unknown microorganism.

Bacterial identification can also be based on the proteins produced under specific growth conditions within the human body. These types of identification procedures are called proteomic analysis. To perform proteomic analysis, proteins from the pathogen are first separated by high-pressure liquid chromatography (HPLC), and the collected fractions are then digested to yield smaller peptide fragments. These peptides are identified by mass spectrometry and compared with those of known microorganisms to identify the unknown microorganism in the original specimen.

Microorganisms can also be identified by the carbohydrates attached to proteins (glycoproteins) in the plasma membrane or cell wall. Antibodies and other carbohydrate-binding proteins can attach to specific carbohydrates on cell surfaces, causing the cells to clump together. Serological tests (e.g., the Lancefield groups tests, which are used for identification of *Streptococcus* species) are performed to detect the unique carbohydrates located on the surface of the cell.

Key Concepts and Summary

- Accurate identification of bacteria is essential in a clinical laboratory for diagnostic and management of epidemics, pandemics, and food poisoning caused by bacterial outbreaks.
- The phenotypic identification of microorganisms involves using observable traits, including profiles of structural components such as lipids, biosynthetic products such as sugars or amino acids, or storage compounds such as poly-β-hydroxybutyrate.
- An unknown microbe may be identified from the unique mass spectrum produced when it is analyzed by **matrix assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF)**.
- Microbes can be identified by determining their lipid compositions, using **fatty acid methyl esters** (**FAME**) or **phospholipid-derived fatty acids** (**PLFA**) **analysis**.
- **Proteomic analysis**, the study of all accumulated proteins of an organism, can also be used for bacterial identification.
- Glycoproteins in the plasma membrane or cell wall structures can bind to lectins or antibodies and can be used for identification.

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3.E: Microbial Biochemistry (Exercises)

3.1: Organic Molecules

Biochemistry is the discipline that studies the chemistry of life, and its objective is to explain form and function based on chemical principles. Organic chemistry is the discipline devoted to the study of carbon-based chemistry, which is the foundation for the study of biomolecules and the discipline of biochemistry. Both biochemistry and organic chemistry are based on the concepts of general chemistry.

Multiple Choice

Which of these elements is *not* a micronutrient?

A. C
B. Ca
C. Co
D. Cu

Answer

A

Which of the following is the name for molecules whose structures are nonsuperimposable mirror images?

A. structural isomers
B. monomers
C. polymers
D. enantiomers

Answer

D

True/False

Aldehydes, amides, carboxylic acids, esters, and ketones all contain carbonyl groups.

Answer

True

Two molecules containing the same types and numbers of atoms but different bonding sequences are called enantiomers.

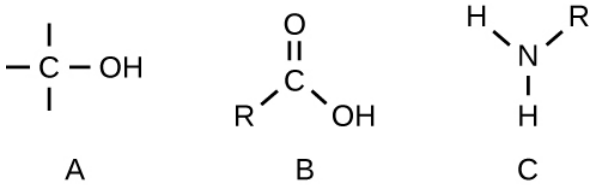
Answer

False

Short Answer

Why are carbon, nitrogen, oxygen, and hydrogen the most abundant elements in living matter and, therefore, considered macronutrients?

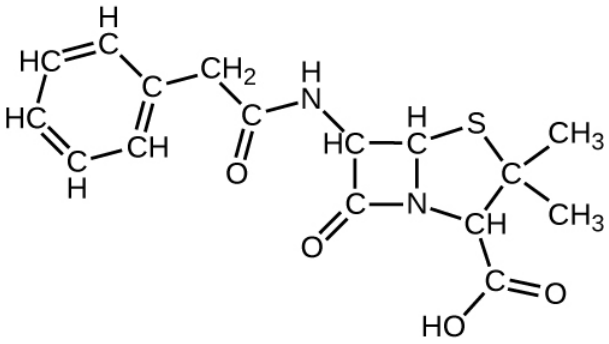
Identify the functional group in each of the depicted structural formulas.



A B C

Critical Thinking

The structural formula shown corresponds to penicillin G, a narrow-spectrum antibiotic that is given intravenously or intramuscularly as a treatment for several bacterial diseases. The antibiotic is produced by fungi of the genus *Penicillium*. (a) Identify three major functional groups in this molecule that each comprise two simpler functional groups. (b) Name the two simpler functional groups composing each of the major functional groups identified in (a).



3.2: Carbohydrates

The most abundant biomolecules on earth are carbohydrates. From a chemical viewpoint, carbohydrates are primarily a combination of carbon and water, and many of them have the empirical formula $(CH_2O)_n$, where *n* is the number of repeated units. This view represents these molecules simply as “hydrated” carbon atom chains in which water molecules attach to each carbon atom, leading to the term “carbohydrates.”

Multiple Choice

By definition, carbohydrates contain which elements?

A. carbon and hydrogen
B. carbon, hydrogen, and nitrogen
C. carbon, hydrogen, and oxygen
D. carbon and oxygen

Answer

C

Monosaccharides may link together to form polysaccharides by forming which type of bond?

A. hydrogen
B. peptide
C. ionic
D. glycosidic

Answer

D

Matching

Match each polysaccharide with its description.

___chitin	A. energy storage polymer in plants
___glycogen	B. structural polymer found in plants
___starch	C. structural polymer found in cell walls of fungi and exoskeletons of some animals
___cellulose	D. energy storage polymer found in animal cells and bacteria

Answer

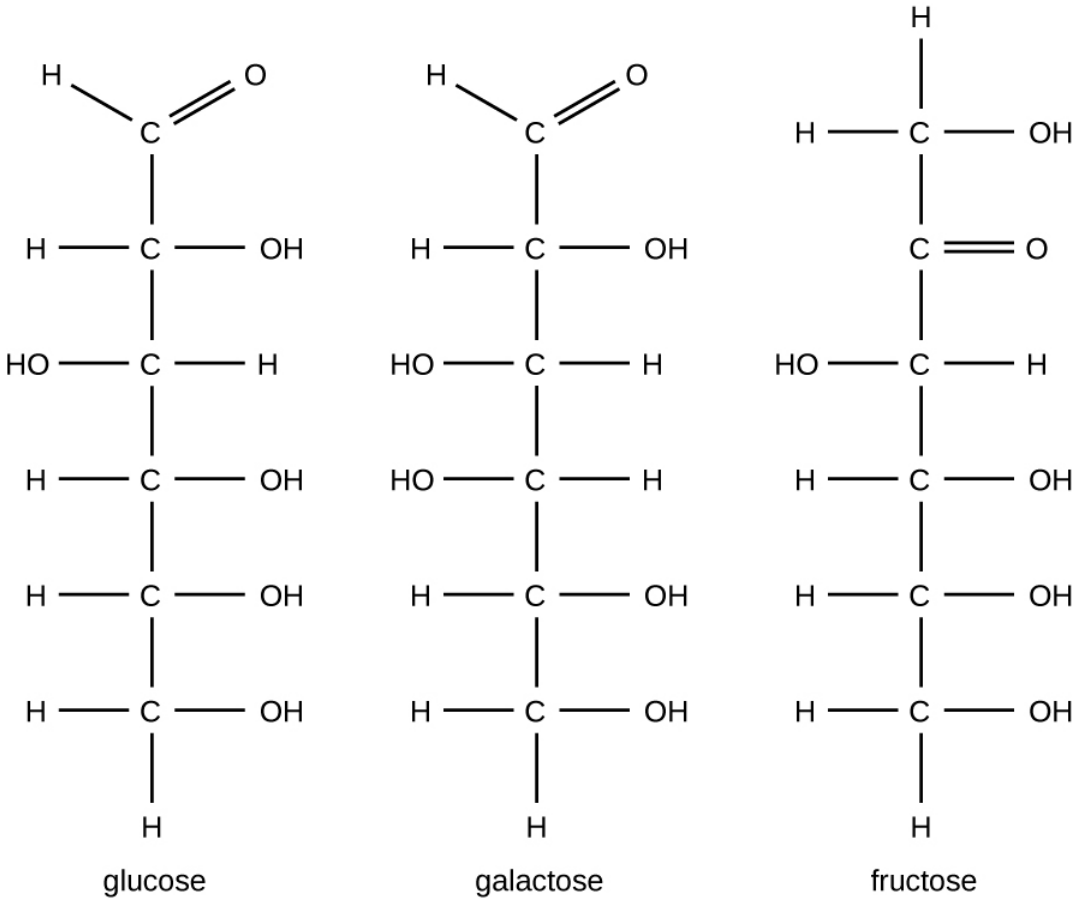
C, D, A, B

Short Answer

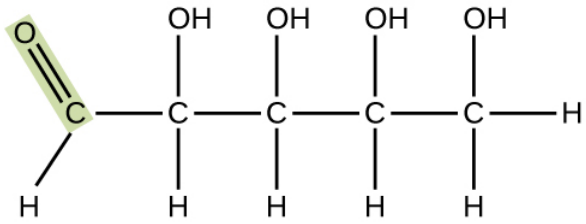
What are monosaccharides, disaccharides, and polysaccharides?

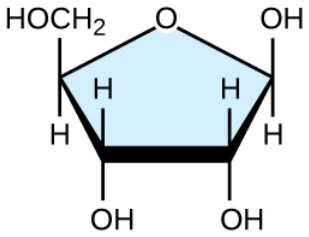
Critical Thinking

The figure depicts the structural formulas of glucose, galactose, and fructose. (a) Circle the functional groups that classify the sugars either an aldose or a ketose, and identify each sugar as one or the other. (b) The chemical formula of these compounds is the same, although the structural formula is different. What are such compounds called?



Structural diagrams for the linear and cyclic forms of a monosaccharide are shown. (a) What is the molecular formula for this monosaccharide? (Count the C, H and O atoms in each to confirm that these two molecules have the same formula, and report this formula.) (b) Identify which hydroxyl group in the linear structure undergoes the ring-forming reaction with the carbonyl group.





The term “dextrose” is commonly used in medical settings when referring to the biologically relevant isomer of the monosaccharide glucose. Explain the logic of this alternative name.

3.3: Lipids

Although they are composed primarily of carbon and hydrogen, lipid molecules may also contain oxygen, nitrogen, sulfur, and phosphorous. Lipids serve numerous and diverse purposes in the structure and functions of organisms. They can be a source of nutrients, a storage form for carbon, energy-storage molecules, or structural components of membranes and hormones. Lipids comprise a broad class of many chemically distinct compounds, the most common of which are discussed in this section.

Multiple Choice

Which of the following describes lipids?

A. a source of nutrients for organisms
B. energy-storage molecules
C. molecules having structural role in membranes
D. molecules that are part of hormones and pigments
E. all of the above

Answer

E

Molecules bearing both polar and nonpolar groups are said to be which of the following?

A. hydrophilic
B. amphipathic
C. hydrophobic
D. polyfunctional

Answer

B

True/False

Lipids are a naturally occurring group of substances that are not soluble in water but are freely soluble in organic solvents.

Answer

False

Fatty acids having no double bonds are called “unsaturated.”

Answer

False

A triglyceride is formed by joining three glycerol molecules to a fatty acid backbone in a dehydration reaction.

Answer

False

Fill in the Blank

Waxes contain esters formed from long-chain _____ and saturated _____, and they may also contain substituted hydrocarbons.

Answer

alcohols; fatty acids

Cholesterol is the most common member of the _____ group, found in animal tissues; it has a tetracyclic carbon ring system with a _____ bond in one of the rings and one free _____group.

Answer

steroid; double; hydroxyl

Critical Thinking

Microorganisms can thrive under many different conditions, including high-temperature environments such as hot springs. To function properly, cell membranes have to be in a fluid state. How do you expect the fatty acid content (saturated versus unsaturated) of bacteria living in high-temperature environments might compare with that of bacteria living in more moderate temperatures?

Short Answer

Describe the structure of a typical phospholipid. Are these molecules polar or nonpolar?

3.4: Proteins

Amino acids are capable of bonding together in essentially any number, yielding molecules of essentially any size that possess a wide array of physical and chemical properties and perform numerous functions vital to all organisms. The molecules derived from amino acids can function as structural components of cells and subcellular entities, as sources of nutrients, as atom- and energy-storage reservoirs, and as functional species such as hormones, enzymes, receptors, and transport molecules.

Multiple Choice

Which of the following groups varies among different amino acids?

A. hydrogen atom
B. carboxyl group
C. *R* group
D. amino group

Answer

C

The amino acids present in proteins differ in which of the following?

A. size
B. shape
C. side groups
D. all of the above

Answer

D

Which of the following bonds are not involved in tertiary structure?

A. peptide bonds
B. ionic bonds
C. hydrophobic interactions
D. hydrogen bonds

Answer

A

Fill in the Blank

The sequence of amino acids in a protein is called its _____.

Answer

Primary structure

Denaturation implies the loss of the _____ and _____ structures without the loss of the _____ structure.

Answer

secondary, tertiary, primary

True/False

A change in one amino acid in a protein sequence always results in a loss of function.

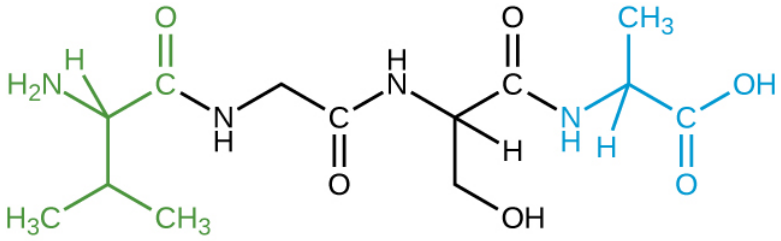
Answer

False

Critical Thinking

Heating a protein sufficiently may cause it to denature. Considering the definition of denaturation, what does this statement say about the strengths of peptide bonds in comparison to hydrogen bonds?

The image shown represents a tetrapeptide. (a) How many peptide bonds are in this molecule? (b) Identify the side groups of the four amino acids composing this peptide.



3.5: Using Biochemistry to Identify Microorganisms

Accurate identification of bacteria is essential in a clinical laboratory for diagnostic and management of epidemics, pandemics, and food poisoning caused by bacterial outbreaks. In this section, we discuss a few methods that use biochemical characteristics to identify microorganisms.

Multiple Choice

Which of the following characteristics/compounds is not considered to be a phenotypic biochemical characteristic used of microbial identification?

A. poly-β-hydroxybutyrate
B. small-subunit (16S) rRNA gene
C. carbon utilization
D. lipid composition

Answer

B

Proteomic analysis is a methodology that deals with which of the following?

A. the analysis of proteins functioning as enzymes within the cell
B. analysis of transport proteins in the cell
C. the analysis of integral proteins of the cell membrane
D. the study of all accumulated proteins of an organism

Answer

D

Which method involves the generation of gas phase ions from intact microorganisms?

A. FAME
B. PLFA
C. MALDI-TOF
D. Lancefield group testing

Answer

C

Which method involves the analysis of membrane-bound carbohydrates?

A. FAME
B. PLFA
C. MALDI-TOF
D. Lancefield group testing

Answer

D

Which method involves conversion of a microbe's lipids to volatile compounds for analysis by gas chromatography?

A. FAME
B. proteomic analysis
C. MALDI-TOF
D. Lancefield group testing

Answer

A

Fill in the Blank

A FAME analysis involves the conversion of _____ to more volatile _____ for analysis using _____.

Answer

fatty acids, methyl esters, gas chromatography

True/False

MALDI-TOF relies on obtaining a unique mass spectrum for the bacteria tested and then checking the acquired mass spectrum against the spectrum databases registered in the analysis software to identify the microorganism.

Answer

True

Lancefield group tests can identify microbes using antibodies that specifically bind cell-surface proteins.

Answer

False

Short Answer

Compare MALDI-TOF, FAME, and PLFA, and explain how each technique would be used to identify pathogens.

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CHAPTER OVERVIEW

4: The Prokaryotic Cell - Bacteria

Despite sharing certain characteristics, cells may vary significantly. The two main types of cells are prokaryotic cells (lacking a nucleus) and eukaryotic cells (containing a well-organized, membrane-bound nucleus). Each type of cell exhibits remarkable variety in structure, function, and metabolic activity. This chapter will focus on the historical discoveries that have shaped our current understanding of microbes, including their origins and their role in human disease. We will then explore the distinguishing structures found in prokaryotic and eukaryotic cells.

4.1: Spontaneous Generation

4.2: Foundations of Modern Cell Theory

4.3: Unique Characteristics of Prokaryotic Cells

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4.1: Spontaneous Generation

Learning Objectives

- Explain the theory of spontaneous generation and why people once accepted it as an explanation for the existence of certain types of organisms
- Explain how certain individuals (van Helmont, Redi, Needham, Spallanzani, and Pasteur) tried to prove or disprove spontaneous generation

Humans have been asking for millennia: Where does new life come from? Religion, philosophy, and science have all wrestled with this question. One of the oldest explanations was the theory of spontaneous generation, which can be traced back to the ancient Greeks and was widely accepted through the Middle Ages.

The Theory of Spontaneous Generation

The Greek philosopher **Aristotle** (384–322 BC) was one of the earliest recorded scholars to articulate the theory of **spontaneous generation**, the notion that life can arise from nonliving matter. Aristotle proposed that life arose from nonliving material if the material contained *pneuma* ("vital heat"). As evidence, he noted several instances of the appearance of animals from environments previously devoid of such animals, such as the seemingly sudden appearance of fish in a new puddle of water.[1]

This theory persisted into the 17th century, when scientists undertook additional experimentation to support or disprove it. By this time, the proponents of the theory cited how frogs simply seem to appear along the muddy banks of the Nile River in Egypt during the annual flooding. Others observed that mice simply appeared among grain stored in barns with thatched roofs. When the roof leaked and the grain molded, mice appeared. Jan Baptista van Helmont, a 17th century Flemish scientist, proposed that mice could arise from rags and wheat kernels left in an open container for 3 weeks. In reality, such habitats provided ideal food sources and shelter for mouse populations to flourish.

However, one of van Helmont's contemporaries, Italian physician **Francesco Redi** (1626–1697), performed an experiment in 1668 that was one of the first to refute the idea that maggots (the larvae of flies) spontaneously generate on meat left out in the open air. He predicted that preventing flies from having direct contact with the meat would also prevent the appearance of maggots. Redi left meat in each of six containers (Figure **4.1.1**). Two were open to the air, two were covered with gauze, and two were tightly sealed. His hypothesis was supported when maggots developed in the uncovered jars, but no maggots appeared in either the gauze-covered or the tightly sealed jars. He concluded that maggots could only form when flies were allowed to lay eggs in the meat, and that the maggots were the offspring of flies, not the product of spontaneous generation.

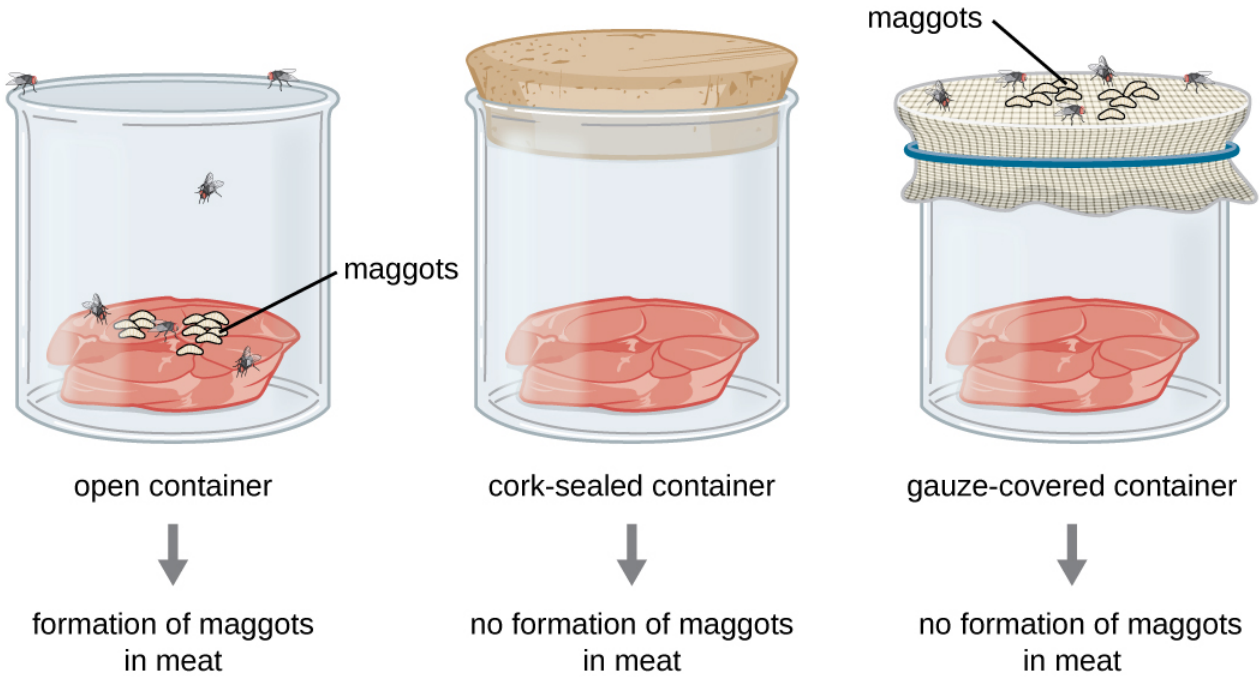


Figure **4.1.1**: Francesco Redi's experimental setup consisted of an open container, a container sealed with a cork top, and a container covered in mesh that let in air but not flies. Maggots only appeared on the meat in the open container. However, maggots were also found on the gauze of the gauze-covered container.

In 1745, **John Needham** (1713–1781) published a report of his own experiments, in which he briefly boiled broth infused with plant or animal matter, hoping to kill all preexisting microbes.[2] He then sealed the flasks. After a few days, Needham observed that the broth had become cloudy and a single drop contained numerous microscopic creatures. He argued that the new microbes must have arisen spontaneously. In reality, however, he likely did not boil the broth enough to kill all preexisting microbes.

Lazzaro Spallanzani (1729–1799) did not agree with Needham's conclusions, however, and performed hundreds of carefully executed experiments using heated broth.[3] As in Needham's experiment, broth in sealed jars and unsealed jars was infused with plant and animal matter. Spallanzani's results contradicted the findings of Needham: Heated but sealed flasks remained clear, without any signs of spontaneous growth, unless the flasks were subsequently opened to the air. This suggested that microbes were introduced into these flasks from the air. In response to Spallanzani's findings, Needham argued that life originates from a "life force" that was destroyed during Spallanzani's extended boiling. Any subsequent sealing of the flasks then prevented new life force from entering and causing spontaneous generation (Figure **4.1.2**).



Figure **4.1.2**: (a) Francesco Redi, who demonstrated that maggots were the offspring of flies, not products of spontaneous generation. (b) John Needham, who argued that microbes arose spontaneously in broth from a "life force." (c) Lazzaro Spallanzani, whose experiments with broth aimed to disprove those of Needham.

Disproving Spontaneous Generation

The debate over spontaneous generation continued well into the 19th century, with scientists serving as proponents of both sides. To settle the debate, the Paris Academy of Sciences offered a prize for resolution of the problem. **Louis Pasteur**, a prominent French chemist who had been studying microbial fermentation and the causes of wine spoilage, accepted the challenge. In 1858, Pasteur filtered air through a gun-cotton filter and, upon microscopic examination of the cotton, found it full of microorganisms, suggesting that the exposure of a broth to air was not introducing a "life force" to the broth but rather airborne microorganisms.

Later, Pasteur made a series of flasks with long, twisted necks ("swan-neck" flasks), in which he boiled broth to sterilize it (Figure **4.1.3**). His design allowed air inside the flasks to be exchanged with air from the outside, but prevented the introduction of any airborne microorganisms, which would get caught in the twists and bends of the flasks' necks. If a life force besides the airborne microorganisms were responsible for microbial growth within the sterilized flasks, it would have access to the broth, whereas the microorganisms would not. He correctly predicted that sterilized broth in his swan-neck flasks would remain sterile as long as the swan necks remained intact. However, should the necks be broken, microorganisms would be introduced, contaminating the flasks and allowing microbial growth within the broth.

Pasteur's set of experiments irrefutably disproved the theory of spontaneous generation and earned him the prestigious Alhumbert Prize from the Paris Academy of Sciences in 1862. In a subsequent lecture in 1864, Pasteur articulated "*Omne vivum ex vivo*" ("Life only comes from life"). In this lecture, Pasteur recounted his famous swan-neck flask experiment, stating that "…life is a germ and a germ is life. Never will the doctrine of spontaneous generation recover from the mortal blow of this simple experiment."[4] To Pasteur's credit, it never has.

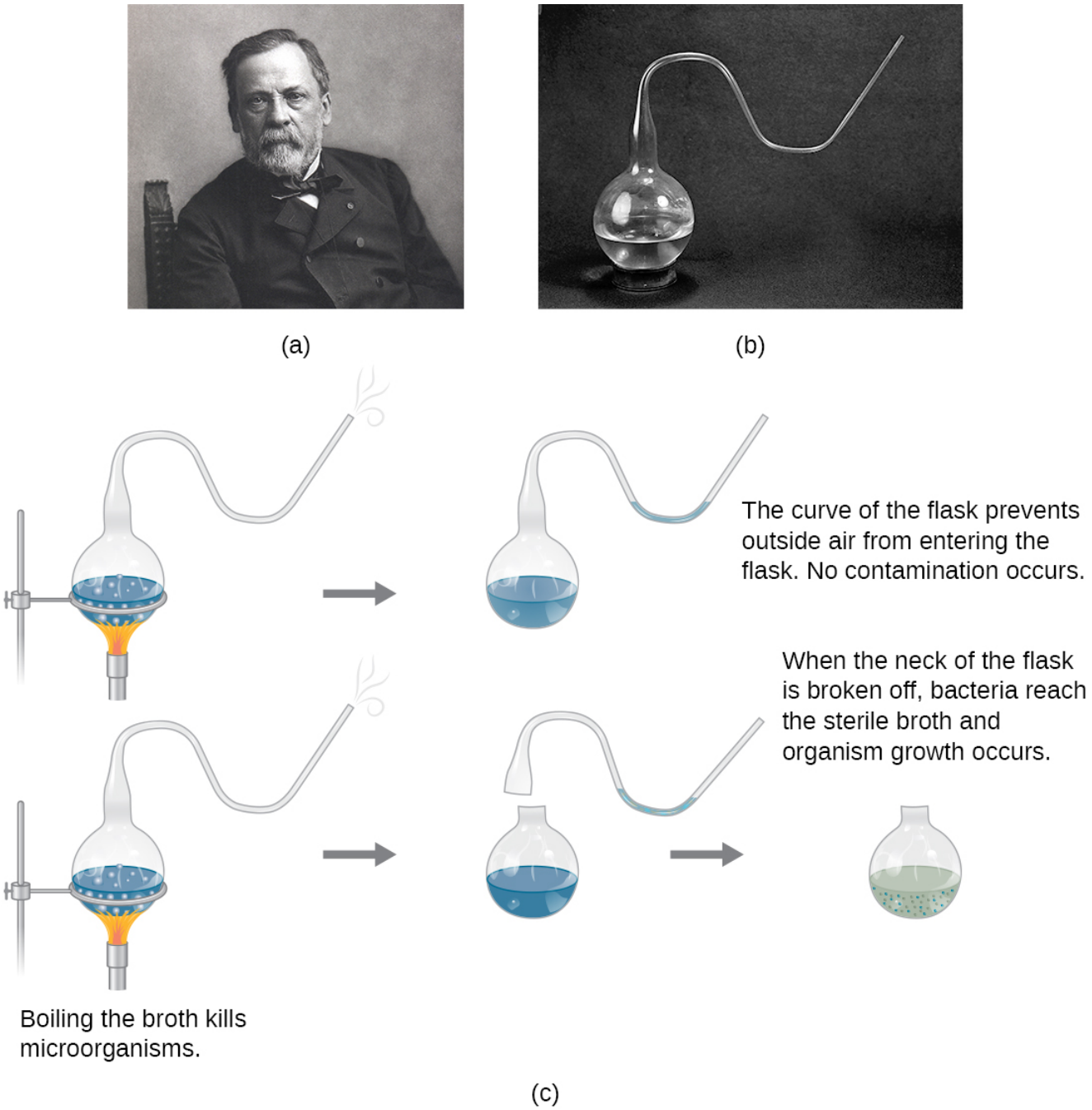


Figure **4.1.3**: (a) French scientist Louis Pasteur, who definitively refuted the long-disputed theory of spontaneous generation. (b) The unique swan-neck feature of the flasks used in Pasteur's experiment allowed air to enter the flask but prevented the entry of bacterial and fungal spores. (c) Pasteur's experiment consisted of two parts. In the first part, the broth in the flask was boiled to sterilize it. When this broth was cooled, it remained free of contamination. In the second part of the experiment, the flask was boiled and then the neck was broken off. The broth in this flask became contaminated. (credit b: modification of work by "Wellcome Images"/Wikimedia Commons)

Key Concepts and Summary

- The theory of **spontaneous generation** states that life arose from nonliving matter. It was a long-held belief dating back to Aristotle and the ancient Greeks.
- Experimentation by Francesco Redi in the 17th century presented the first significant evidence refuting spontaneous generation by showing that flies must have access to meat for maggots to develop on the meat. Prominent scientists designed experiments and argued both in support of (John Needham) and against (Lazzaro Spallanzani) spontaneous generation.

- Louis Pasteur is credited with conclusively disproving the theory of spontaneous generation with his famous swan-neck flask experiment. He subsequently proposed that “life only comes from life.”

Footnotes

1. 1 K. Zwier. “Aristotle on Spontaneous Generation.” www.sju.edu/int/academics/cas...R.%20Zwier.pdf
2. 2 E. Capanna. “Lazzaro Spallanzani: At the Roots of Modern Biology.” *Journal of Experimental Zoology* 285 no. 3 (1999):178–196.
3. 3 R. Mancini, M. Nigro, G. Ippolito. “Lazzaro Spallanzani and His Refutation of the Theory of Spontaneous Generation.” *Le Infezioni in Medicina* 15 no. 3 (2007):199–206.
4. 4 R. Vallery-Radot. *The Life of Pasteur*, trans. R.L. Devonshire. New York: McClure, Phillips and Co, 1902, 1:142.

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4.2: Foundations of Modern Cell Theory

Learning Objectives

- Explain the key points of cell theory and the individual contributions of Hooke, Schleiden, Schwann, Remak, and Virchow
- Explain the key points of endosymbiotic theory and cite the evidence that supports this concept
- Explain the contributions of Semmelweis, Snow, Pasteur, Lister, and Koch to the development of germ theory

While some scientists were arguing over the theory of spontaneous generation, other scientists were making discoveries leading to a better understanding of what we now call the cell theory. Modern cell theory has two basic tenets:

- All cells only come from other cells (the principle of biogenesis).
- Cells are the fundamental units of organisms.

Today, these tenets are fundamental to our understanding of life on earth. However, modern cell theory grew out of the collective work of many scientists.

The Origins of Cell Theory

The English scientist **Robert Hooke** first used the term “cells” in 1665 to describe the small chambers within cork that he observed under a microscope of his own design. To Hooke, thin sections of cork resembled “Honey-comb,” or “small Boxes or Bladders of Air.” He noted that each “Cavern, Bubble, or Cell” was distinct from the others (Figure **4.2.1**). At the time, Hooke was not aware that the cork cells were long dead and, therefore, lacked the internal structures found within living cells.

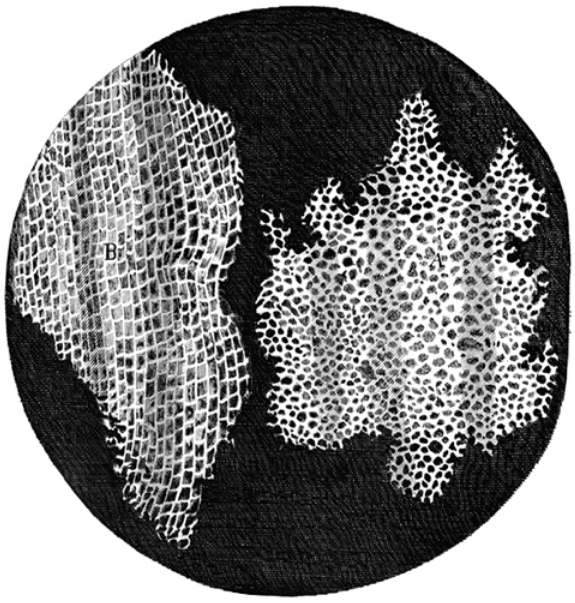


Figure **4.2.1**: Robert Hooke (1635–1703) was the first to describe cells based upon his microscopic observations of cork. This illustration was published in his work *Micrographia*.

Despite Hooke’s early description of cells, their significance as the fundamental unit of life was not yet recognized. Nearly 200 years later, in 1838, Matthias Schleiden (1804–1881), a German botanist who made extensive microscopic observations of plant tissues, described them as being composed of cells. Visualizing plant cells was relatively easy because plant cells are clearly separated by their thick cell walls. Schleiden believed that cells formed through crystallization, rather than cell division.

Theodor Schwann (1810–1882), a noted German physiologist, made similar microscopic observations of animal tissue. In 1839, after a conversation with Schleiden, Schwann realized that similarities existed between plant and animal tissues. This laid the foundation for the idea that cells are the fundamental components of plants and animals.

In the 1850s, two Polish scientists living in Germany pushed this idea further, culminating in what we recognize today as the modern cell theory. In 1852, **Robert Remak** (1815–1865), a prominent neurologist and embryologist, published convincing

evidence that cells are derived from other cells as a result of cell division. However, this idea was questioned by many in the scientific community. Three years later, **Rudolf Virchow** (1821–1902), a well-respected pathologist, published an editorial essay entitled "Cellular Pathology," which popularized the concept of cell theory using the Latin phrase *omnis cellula a cellula* ("all cells arise from cells"), which is essentially the second tenet of modern cell theory.[1]Given the similarity of Virchow's work to Remak's, there is some controversy as to which scientist should receive credit for articulating cell theory. See the following Eye on Ethics feature for more about this controversy.

Science and Plagiarism

Rudolf Virchow, a prominent, Polish-born, German scientist, is often remembered as the "Father of Pathology." Well known for innovative approaches, he was one of the first to determine the causes of various diseases by examining their effects on tissues and organs. He was also among the first to use animals in his research and, as a result of his work, he was the first to name numerous diseases and created many other medical terms. Over the course of his career, he published more than 2,000 papers and headed various important medical facilities, including the Charité – Universitätsmedizin Berlin, a prominent Berlin hospital and medical school. But he is, perhaps, best remembered for his 1855 editorial essay titled "Cellular Pathology," published in *Archiv für Pathologische Anatomie und Physiologie*, a journal that Virchow himself cofounded and still exists today.

Despite his significant scientific legacy, there is some controversy regarding this essay, in which Virchow proposed the central tenet of modern cell theory—that all cells arise from other cells. Robert Remak, a former colleague who worked in the same laboratory as Virchow at the University of Berlin, had published the same idea 3 years before. Though it appears Virchow was familiar with Remak's work, he neglected to credit Remak's ideas in his essay. When Remak wrote a letter to Virchow pointing out similarities between Virchow's ideas and his own, Virchow was dismissive. In 1858, in the preface to one of his books, Virchow wrote that his 1855 publication was just an editorial piece, not a scientific paper, and thus there was no need to cite Remak's work.



(a) (b)

Figure **4.2.2**: (a) Rudolf Virchow (1821–1902) popularized the cell theory in an 1855 essay entitled "Cellular Pathology." (b) The idea that all cells originate from other cells was first published in 1852 by his contemporary and former colleague Robert Remak (1815–1865).

By today's standards, Virchow's editorial piece would certainly be considered an act of plagiarism, since he presented Remak's ideas as his own. However, in the 19th century, standards for academic integrity were much less clear. Virchow's strong reputation, coupled with the fact that Remak was a Jew in a somewhat anti-Semitic political climate, shielded him from any significant repercussions. Today, the process of peer review and the ease of access to the scientific literature help discourage plagiarism. Although scientists are still motivated to publish original ideas that advance scientific knowledge, those who would consider plagiarizing are well aware of the serious consequences.

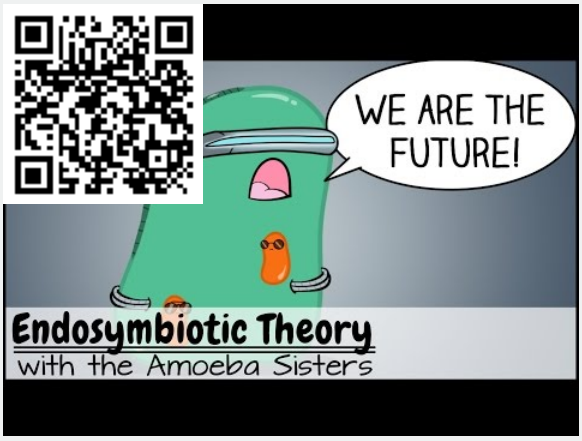
In academia, plagiarism represents the theft of both individual thought and research—an offense that can destroy reputations and end careers.[2 3 4 5]

Endosymbiotic Theory

Video: Endosymbiotic Theory

Explore the endosymbiotic theory with the Amoeba Sisters! This theory explains the development of the eukaryote cell from prokaryote cell symbiosis. Scientific theories are also briefly defined.





As scientists were making progress toward understanding the role of cells in plant and animal tissues, others were examining the structures within the cells themselves. In 1831, Scottish botanist Robert Brown (1773–1858) was the first to describe observations of nuclei, which he observed in plant cells. Then, in the early 1880s, German botanist Andreas Schimper (1856–1901) was the first to describe the chloroplasts of plant cells, identifying their role in starch formation during photosynthesis and noting that they divided independent of the nucleus.

Based upon the chloroplasts' ability to reproduce independently, Russian botanist Konstantin Mereschkowski (1855–1921) suggested in 1905 that chloroplasts may have originated from ancestral photosynthetic bacteria living symbiotically inside a eukaryotic cell. He proposed a similar origin for the nucleus of plant cells. This was the first articulation of the endosymbiotic hypothesis, and would explain how eukaryotic cells evolved from ancestral bacteria.

Mereschkowski's endosymbiotic hypothesis was furthered by American anatomist Ivan Wallin (1883–1969), who began to experimentally examine the similarities between mitochondria, chloroplasts, and bacteria—in other words, to put the endosymbiotic hypothesis to the test using objective investigation. Wallin published a series of papers in the 1920s supporting the endosymbiotic hypothesis, including a 1926 publication co-authored with Mereschkowski. Wallin claimed he could culture mitochondria outside of their eukaryotic host cells. Many scientists dismissed his cultures of mitochondria as resulting from bacterial contamination. Modern genome sequencing work supports the dissenting scientists by showing that much of the genome of mitochondria had been transferred to the host cell's nucleus, preventing the mitochondria from being able to live on their own.[6] [7]

Wallin's ideas regarding the endosymbiotic hypothesis were largely ignored for the next 50 years because scientists were unaware that these organelles contained their own DNA. However, with the discovery of mitochondrial and chloroplast DNA in the 1960s, the endosymbiotic hypothesis was resurrected. **Lynn Margulis** (1938–2011), an American geneticist, published her ideas regarding the endosymbiotic hypothesis of the origins of mitochondria and chloroplasts in 1967.[8] In the decade leading up to her publication, advances in microscopy had allowed scientists to differentiate prokaryotic cells from eukaryotic cells. In her publication, Margulis reviewed the literature and argued that the eukaryotic organelles such as mitochondria and chloroplasts are of prokaryotic origin. She presented a growing body of microscopic, genetic, molecular biology, fossil, and geological data to support her claims.

Again, this hypothesis was not initially popular, but mounting genetic evidence due to the advent of DNA sequencing supported the endosymbiotic theory, which is now defined as the theory that mitochondria and chloroplasts arose as a result of prokaryotic cells establishing a symbiotic relationship within a eukaryotic host (Figure **4.2.3**). With Margulis' initial endosymbiotic theory gaining wide acceptance, she expanded on the theory in her 1981 book *Symbiosis in Cell Evolution*. In it, she explains how endosymbiosis is a major driving factor in the evolution of organisms. More recent genetic sequencing and phylogenetic analysis show that mitochondrial DNA and chloroplast DNA are highly related to their bacterial counterparts, both in DNA sequence and chromosome structure. However, mitochondrial DNA and chloroplast DNA are reduced compared with nuclear DNA because many of the genes have moved from the organelles into the host cell's nucleus. Additionally, mitochondrial and chloroplast ribosomes are structurally similar to bacterial ribosomes, rather than to the eukaryotic ribosomes of their hosts. Last, the binary fission of these organelles strongly resembles the binary fission of bacteria, as compared with mitosis performed by eukaryotic cells. Since Margulis' original proposal, scientists have observed several examples of bacterial endosymbionts in modern-day eukaryotic cells. Examples include the endosymbiotic bacteria found within the guts of certain insects, such as cockroaches,[9] and photosynthetic bacteria-like organelles found in protists.[10]

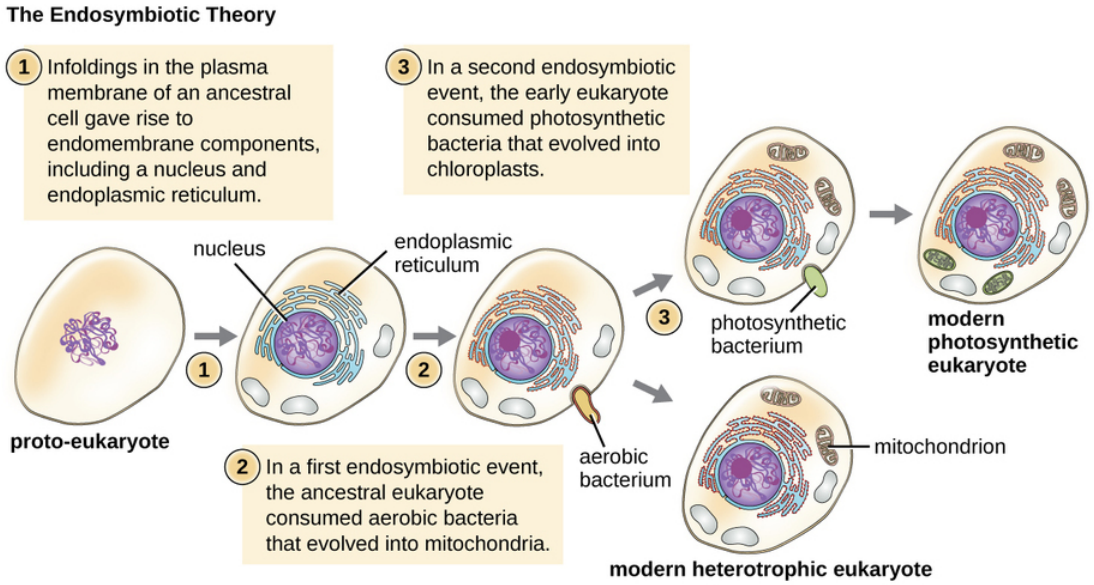


Figure **4.2.3**: According to the endosymbiotic theory, mitochondria and chloroplasts are each derived from the uptake of bacteria. These bacteria established a symbiotic relationship with their host cell that eventually led to the bacteria evolving into mitochondria and chloroplasts.

Key Concepts and Summary

- Although cells were first observed in the 1660s by Robert Hooke, **cell theory** was not well accepted for another 200 years. The work of scientists such as Schleiden, Schwann, Remak, and Virchow contributed to its acceptance.
- **Endosymbiotic theory** states that mitochondria and chloroplasts, organelles found in many types of organisms, have their origins in bacteria. Significant structural and genetic information support this theory.
- The **miasma theory of disease** was widely accepted until the 19th century, when it was replaced by the **germ theory of disease** thanks to the work of Semmelweis, Snow, Pasteur, Lister, and Koch, and others.

Footnotes

1. 1 M. Schultz. “Rudolph Virchow.” *Emerging Infectious Diseases* 14 no. 9 (2008):1480–1481.
2. 2 B. Kisch. “Forgotten Leaders in Modern Medicine, Valentin, Gouby, Remak, Auerbach.” *Transactions of the American Philosophical Society* 44 (1954):139–317.
3. 3 H. Harris. *The Birth of the Cell*. New Haven, CT: Yale University Press, 2000:133.
4. 4 C. Webster (ed.). *Biology, Medicine and Society 1840-1940*. Cambridge, UK; Cambridge University Press, 1981:118–119.
5. 5 C. Zuchora-Walske. *Key Discoveries in Life Science*. Minneapolis, MN: Lerner Publishing, 2015:12–13.
6. 6 T. Embley, W. Martin. “Eukaryotic Evolution, Changes, and Challenges.” *Nature* Vol. 440 (2006):623–630.
7. 7 O.G. Berg, C.G. Kurland. “Why Mitochondrial Genes Are Most Often Found in Nuclei.” *Molecular Biology and Evolution* 17 no. 6 (2000):951–961.
8. 8 L. *Sagan. “On the Origin of Mitosing Cells.” Journal of Theoretical Biology 14 no. 3 (1967):225–274.*
9. 9 A.E. Douglas. “The Microbial Dimension in Insect Nutritional Ecology.” *Functional Ecology* 23 (2009):38–47.
10. 10 J.M. Jaynes, L.P. Vernon. “The Cyanelle of *Cyanophora paradoxa*: Almost a Cyanobacterial Chloroplast.” *Trends in Biochemical Sciences* 7 no. 1 (1982):22–24.
11. 11 Alexander, J. Wesley. “The Contributions of Infection Control to a Century of Progress” *Annals of Surgery* 201:423-428, 1985.

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4.3: Unique Characteristics of Prokaryotic Cells

Learning Objectives

- Explain the distinguishing characteristics of prokaryotic cells
- Describe common cell morphologies and cellular arrangements typical of prokaryotic cells and explain how cells maintain their morphology
- Describe internal and external structures of prokaryotic cells in terms of their physical structure, chemical structure, and function
- Compare the distinguishing characteristics of bacterial and archaeal cells

Cell theory states that the cell is the fundamental unit of life. However, cells vary significantly in size, shape, structure, and function. At the simplest level of construction, all cells possess a few fundamental components. These include **cytoplasm** (a gel-like substance composed of water and dissolved chemicals needed for growth), which is contained within a **plasma membrane** (also called a cell membrane or cytoplasmic membrane); one or more **chromosomes**, which contain the genetic blueprints of the cell; and **ribosomes**, organelles used for the production of proteins.

Beyond these basic components, cells can vary greatly between organisms, and even within the same multicellular organism. The two largest categories of cells—prokaryotic cells and eukaryotic cells—are defined by major differences in several cell structures. Prokaryotic cells lack a nucleus surrounded by a complex nuclear membrane and generally have a single, circular chromosome located in a nucleoid. Eukaryotic cells have a nucleus surrounded by a complex nuclear membrane that contains multiple, rod-shaped chromosomes.[1]

All plant cells and animal cells are eukaryotic. Some microorganisms are composed of prokaryotic cells, whereas others are composed of eukaryotic cells. Prokaryotic microorganisms are classified within the domains Archaea and Bacteria, whereas eukaryotic organisms are classified within the domain Eukarya.

The structures inside a cell are analogous to the organs inside a human body, with unique structures suited to specific functions. Some of the structures found in prokaryotic cells are similar to those found in some eukaryotic cells; others are unique to prokaryotes. Although there are some exceptions, eukaryotic cells tend to be larger than prokaryotic cells. The comparatively larger size of eukaryotic cells dictates the need to compartmentalize various chemical processes within different areas of the cell, using complex membrane-bound organelles. In contrast, prokaryotic cells generally lack membrane-bound organelles; however, they often contain inclusions that compartmentalize their cytoplasm. Figure **4.3.1** illustrates structures typically associated with prokaryotic cells. These structures are described in more detail in the next section.

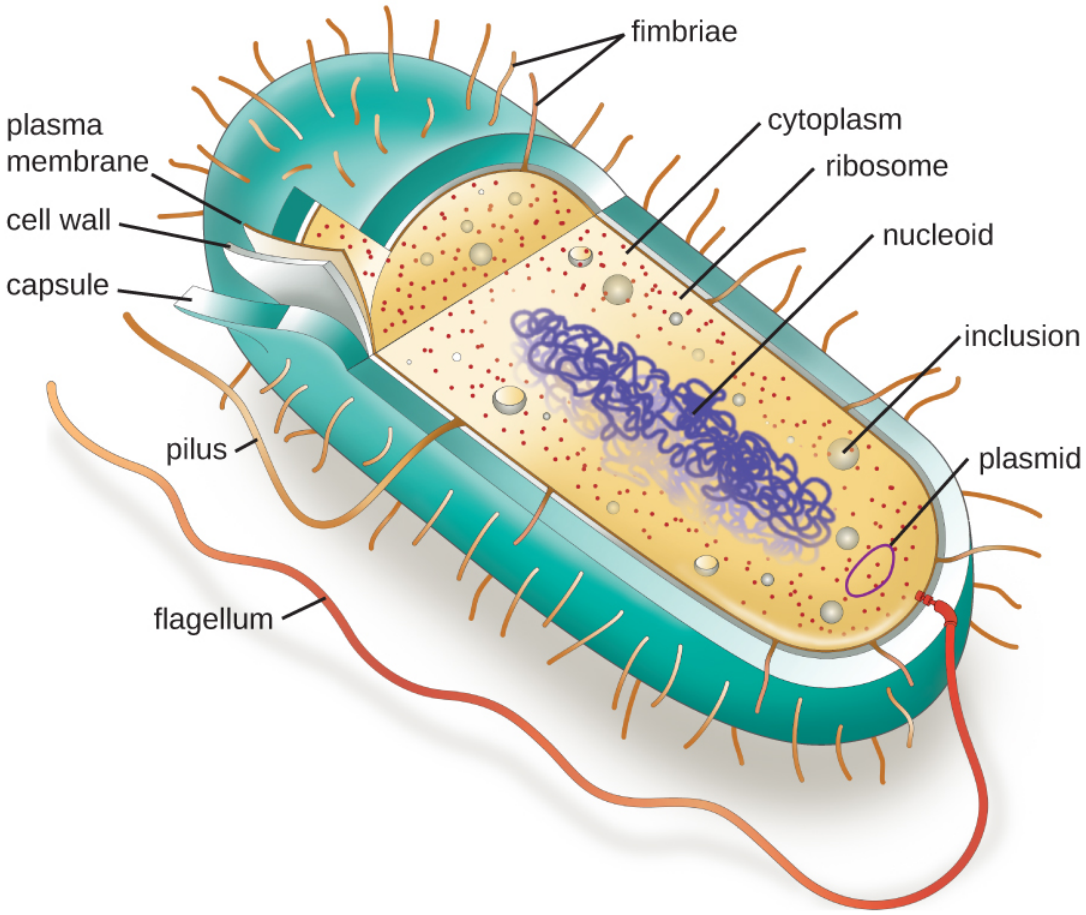


Figure **4.3.1**: A typical prokaryotic cell contains a cell membrane, chromosomal DNA that is concentrated in a nucleoid, ribosomes, and a cell wall. Some prokaryotic cells may also possess flagella, pili, fimbriae, and capsules.

Common Cell Morphologies and Arrangements

Individual cells of a particular prokaryotic organism are typically similar in shape, or **cell morphology**. Although thousands of prokaryotic organisms have been identified, only a handful of cell morphologies are commonly seen microscopically. Figure **4.3.2** names and illustrates cell morphologies commonly found in prokaryotic cells. In addition to cellular shape, prokaryotic cells of the same species may group together in certain distinctive arrangements depending on the plane of cell division. Some common arrangements are shown in Figure **4.3.3**.

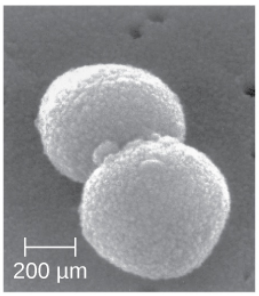
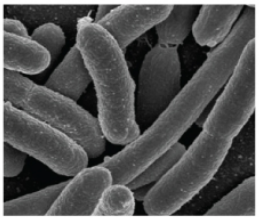
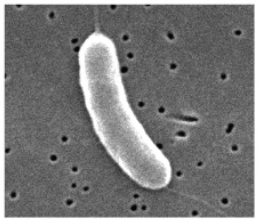
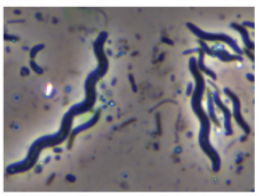
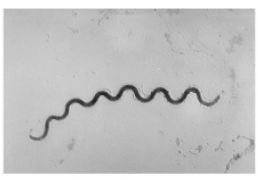
Common Prokaryotic Cell Shapes			
Name	**Description**	**Illustration**	**Image**
Coccus (pl. cocci)	Round		
Bacillus (pl. bacilli)	Rod		
Vibrio (pl. vibrios)	Curved rod		
Coccobacillus (pl. coccobacilli)	Short rod		
Spirillum (pl. spirilla)	Spiral		
Spirochete (pl. spirochetes)	Long, loose, helical spiral		

Figure 4.3.2: (credit “Coccus” micrograph: modification of work by Janice Haney Carr, Centers for Disease Control and Prevention; credit “Coccobacillus” micrograph: modification of work by Janice Carr, Centers for Disease Control and Prevention; credit “Spirochete” micrograph: modification of work by Centers for Disease Control and Prevention)

Common Prokaryotic Cell Arrangements

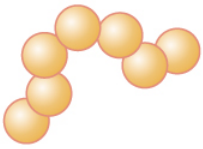
Name	Description	Illustration
Coccus (pl. cocci)	Single coccus	
Diplococcus (pl. diplococci)	Pair of two cocci	
Tetrad (pl. tetrads)	Grouping of four cells arranged in a square	
Streptococcus (pl. streptococci)	Chain of cocci	
Staphylococcus (pl. staphylococci)	Cluster of cocci	
Bacillus (pl. bacilli)	Single rod	
Streptobacillus (pl. streptobacilli)	Chain of rods	

Figure 4.3.3: Common cell arrangements.

Osmatic Pressure

In most prokaryotic cells, morphology is maintained by the **cell wall** in combination with **cytoskeletal elements**. The cell wall is a structure found in most prokaryotes and some eukaryotes; it envelopes the cell membrane, protecting the cell from changes in **osmotic pressure** (Figure 4.3.4). Osmotic pressure occurs because of differences in the concentration of solutes on opposing sides of a semipermeable membrane. Water is able to pass through a semipermeable membrane, but solutes (dissolved molecules like salts, sugars, and other compounds) cannot. When the concentration of solutes is greater on one side of the membrane, water diffuses across the membrane from the side with the lower concentration (more water) to the side with the higher concentration (less water) until the concentrations on both sides become equal. This diffusion of water is called **osmosis**, and it can cause extreme osmotic pressure on a cell when its external environment changes.

The external environment of a cell can be described as an isotonic, hypertonic, or hypotonic medium. In an **isotonic medium**, the solute concentrations inside and outside the cell are approximately equal, so there is no net movement of water across the cell membrane. In a **hypertonic medium**, the solute concentration outside the cell exceeds that inside the cell, so water diffuses out of the cell and into the external medium. In a **hypotonic medium**, the solute concentration inside the cell exceeds that outside of the cell, so water will move by osmosis into the cell. This causes the cell to swell and potentially lyse, or burst.

The degree to which a particular cell is able to withstand changes in osmotic pressure is called tonicity. Cells that have a cell wall are better able to withstand subtle changes in osmotic pressure and maintain their shape. In hypertonic environments, cells that lack a cell wall can become dehydrated, causing **crenation**, or shriveling of the cell; the plasma membrane contracts and appears scalloped or notched (Figure **4.3.4**). By contrast, cells that possess a cell wall undergo **plasmolysis** rather than crenation. In plasmolysis, the plasma membrane contracts and detaches from the cell wall, and there is a decrease in interior volume, but the cell wall remains intact, thus allowing the cell to maintain some shape and integrity for a period of time (Figure **4.3.5**). Likewise, cells that lack a cell wall are more prone to lysis in hypotonic environments. The presence of a cell wall allows the cell to maintain its shape and integrity for a longer time before lysing (Figure **4.3.5**).

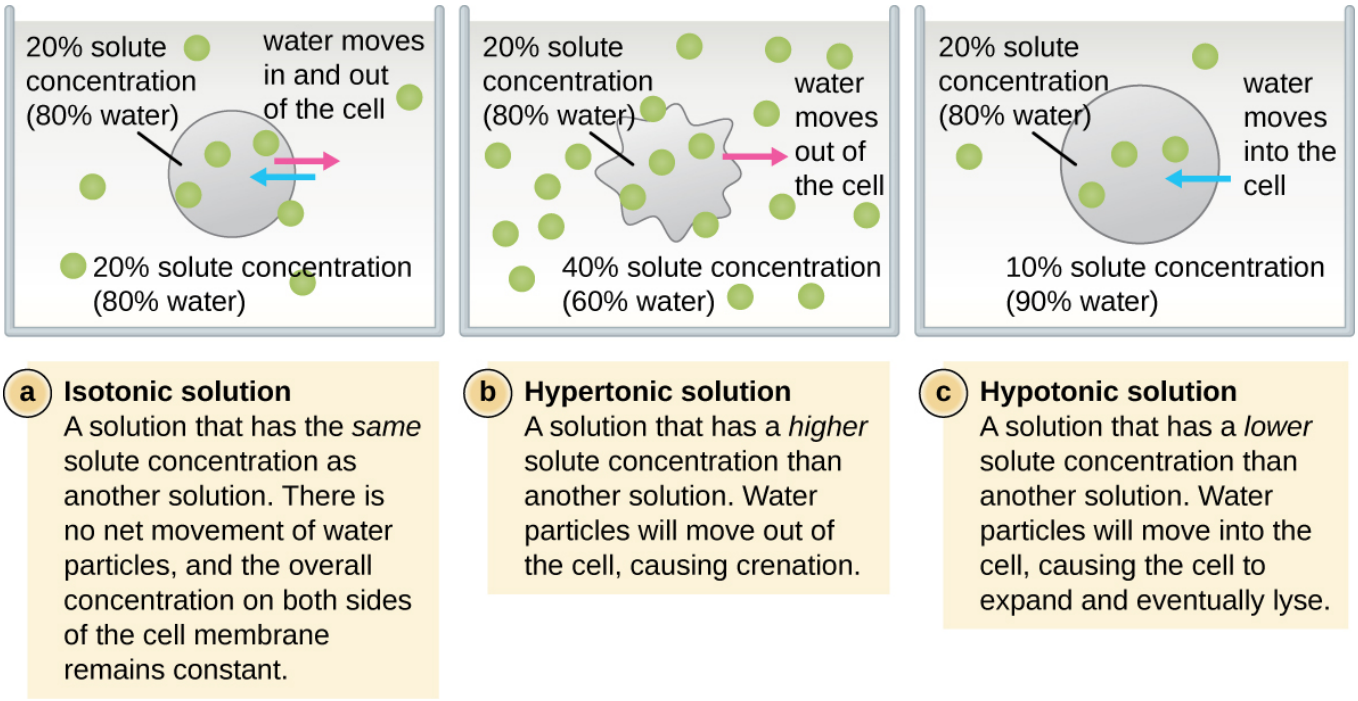


Figure **4.3.4**: In cells that lack a cell wall, changes in osmotic pressure can lead to crenation in hypertonic environments or cell lysis in hypotonic environments.

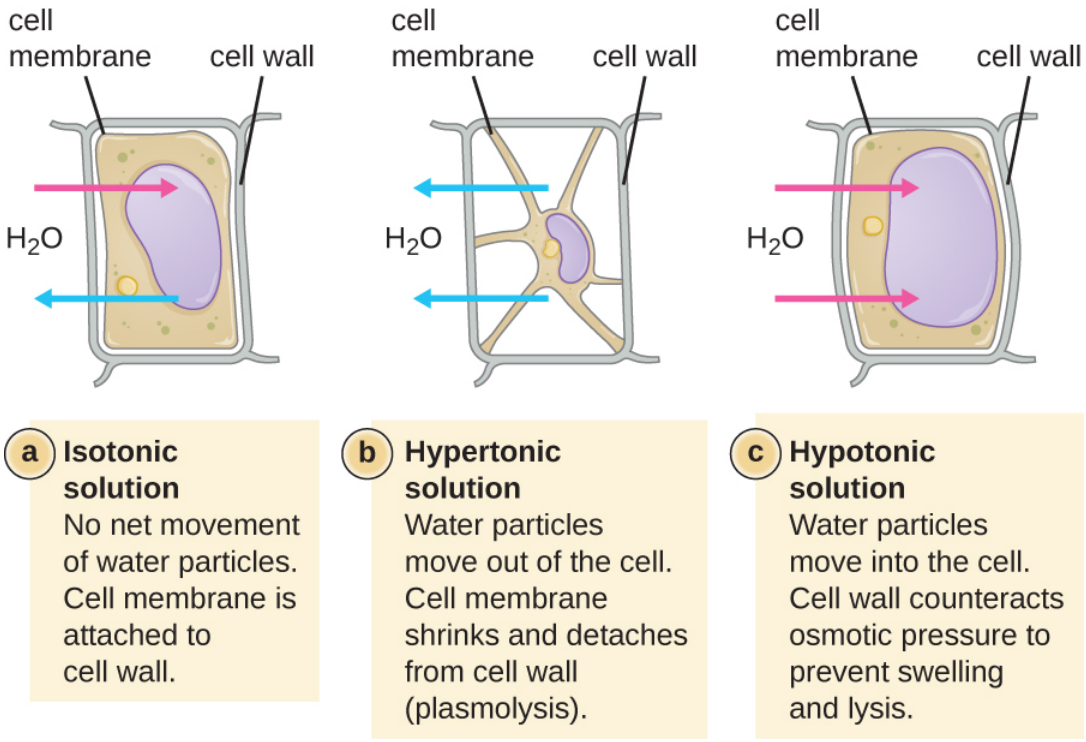


Figure **4.3.5**: In prokaryotic cells, the cell wall provides some protection against changes in osmotic pressure, allowing it to maintain its shape longer. The cell membrane is typically attached to the cell wall in an isotonic medium (left). In a hypertonic medium, the cell membrane detaches from the cell wall and contracts (plasmolysis) as water leaves the cell. In a hypotonic medium (right), the cell wall prevents the cell membrane from expanding to the point of bursting, although lysis will eventually occur if too much water is absorbed.

Key Concepts and Summary

- Prokaryotic cells differ from eukaryotic cells in that their genetic material is contained in a **nucleoid** rather than a membrane-bound nucleus. In addition, prokaryotic cells generally lack membrane-bound organelles.
- Prokaryotic cells of the same species typically share a similar **cell morphology** and **cellular arrangement**.
- Most prokaryotic cells have a **cell wall** that helps the organism maintain cellular morphology and protects it against changes in osmotic pressure.

Footnotes

1. 1 Y.-H.M. Chan, W.F. Marshall. “Scaling Properties of Cell and Organelle Size.” *Organogenesis* 6 no. 2 (2010):88–96.
2. 2 F. Rothfuss, M Bender, R Conrad. “Survival and Activity of Bacteria in a Deep, Aged Lake Sediment (Lake Constance).” *Microbial Ecology* 33 no. 1 (1997):69–77.
3. 3 R. Sinclair et al. “Persistence of Category A Select Agents in the Environment.” *Applied and Environmental Microbiology* 74 no. 3 (2008):555–563.
4. 4 T.J. Silhavy, D. Kahne, S. Walker. “The Bacterial Cell Envelope.” *Cold Spring Harbor Perspectives in Biology* 2 no. 5 (2010):a000414.
5. 5 B. Zuber et al. “Granular Layer in the Periplasmic Space of Gram-Positive Bacteria and Fine Structures of *Enterococcus gallinarum* and *Streptococcus gordonii* Septa Revealed by Cryo-Electron Microscopy of Vitreous Sections.” *Journal of Bacteriology* 188 no. 18 (2006):6652–6660
6. 6 L. Gana, S. Chena, G.J. Jensena. “Molecular Organization of Gram-Negative Peptidoglycan.” *Proceedings of the National Academy of Sciences of the United States of America* 105 no. 48 (2008):18953–18957.
7. 7 J.A. Garnetta et al. “Structural Insights Into the Biogenesis and Biofilm Formation by the *Escherichia coli* Common Pilus.” *Proceedings of the National Academy of Sciences of the United States of America* 109 no. 10 (2012):3950–3955.

8. 8 T. Proft, E.N. Baker. “Pili in Gram-Negative and Gram-Positive Bacteria—Structure, Assembly and Their Role in Disease.” *Cellular and Molecular Life Sciences* 66 (2009):613.

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CHAPTER OVERVIEW

5: The Cell

Life takes many forms, from giant redwood trees towering hundreds of feet in the air to the tiniest known microbes, which measure only a few billionths of a meter. Humans have long pondered life's origins and debated the defining characteristics of life, but our understanding of these concepts has changed radically since the invention of the microscope. In the 17th century, observations of microscopic life led to the development of the cell theory: the idea that the fundamental unit of life is the cell, that all organisms contain at least one cell, and that cells only come from other cells.

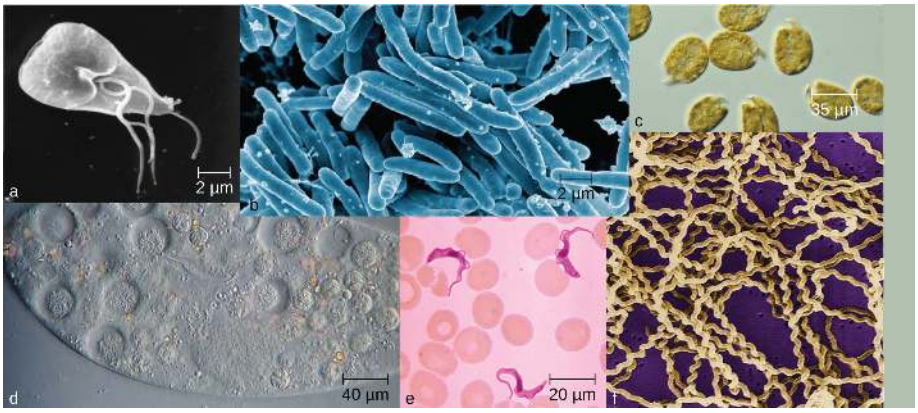


Figure **5.1**: Microorganisms vary visually in their size and shape, as can be observed microscopically; but they also vary in invisible ways, such as in their metabolic capabilities. (credit a, e, f: modification of work by Centers for Disease Control and Prevention; credit b: modification of work by NIAID; credit c: modification of work by CSIRO; credit d: modification of work by "Microscopic World"/YouTube)

Despite sharing certain characteristics, cells may vary significantly. The two main types of cells are prokaryotic cells (lacking a nucleus) and eukaryotic cells (containing a well-organized, membrane-bound nucleus). Each type of cell exhibits remarkable variety in structure, function, and metabolic activity. This chapter will focus on the historical discoveries that have shaped our current understanding of microbes, including their origins and their role in human disease. We will then explore the distinguishing structures found in prokaryotic and eukaryotic cells.

5.1: Unique Characteristics of Eukaryotic Cells
5.2: Eukaryotic Cell Anatomy
5.3: The Cytoplasmic Membrane
5.4: The Cell Wall
5.5: The Endomembrane System
- 5.5A: The Nucleus
- 5.5B: The Endoplasmic Reticulum
- 5.5C: The Golgi Complex

5.6: Other Internal Membrane-Bound Organelles
- 5.6A: Mitochondria
- 5.6B: Chloroplasts
- 5.6C: Lysosomes, Peroxisomes, Vacuoles, and Vesicles

5.7: Ribosomes
5.8: The Cytoskeleton
5.9: Flagella and Cilia
5.10: The Endosymbiotic Theory
5.E: The Cell (Exercises)

5.E: The Eukaryotic Cell (Exercises)

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5.1: Unique Characteristics of Eukaryotic Cells

Learning Objectives

- Explain the distinguishing characteristics of eukaryotic cells
- Describe internal and external structures of prokaryotic cells in terms of their physical structure, chemical structure, and function
- Identify and describe structures and organelles unique to eukaryotic cells
- Compare and contrast similar structures found in prokaryotic and eukaryotic cells

Eukaryotic organisms include protozoans, algae, fungi, plants, and animals. Some eukaryotic cells are independent, single-celled microorganisms, whereas others are part of multicellular organisms. The cells of eukaryotic organisms have several distinguishing characteristics. Above all, eukaryotic cells are defined by the presence of a **nucleus** surrounded by a complex nuclear membrane. Also, eukaryotic cells are characterized by the presence of **membrane-bound organelles** in the cytoplasm. Organelles such as mitochondria, the endoplasmic reticulum (ER), Golgi apparatus, lysosomes, and peroxisomes are held in place by the **cytoskeleton**, an internal network that supports transport of intracellular components and helps maintain cell shape (Figure **5.1.1**). The genome of eukaryotic cells is packaged in multiple, rod-shaped chromosomes as opposed to the single, circular-shaped chromosome that characterizes most prokaryotic cells. Table **5.1.1** compares the characteristics of eukaryotic cell structures with those of bacteria and archaea.

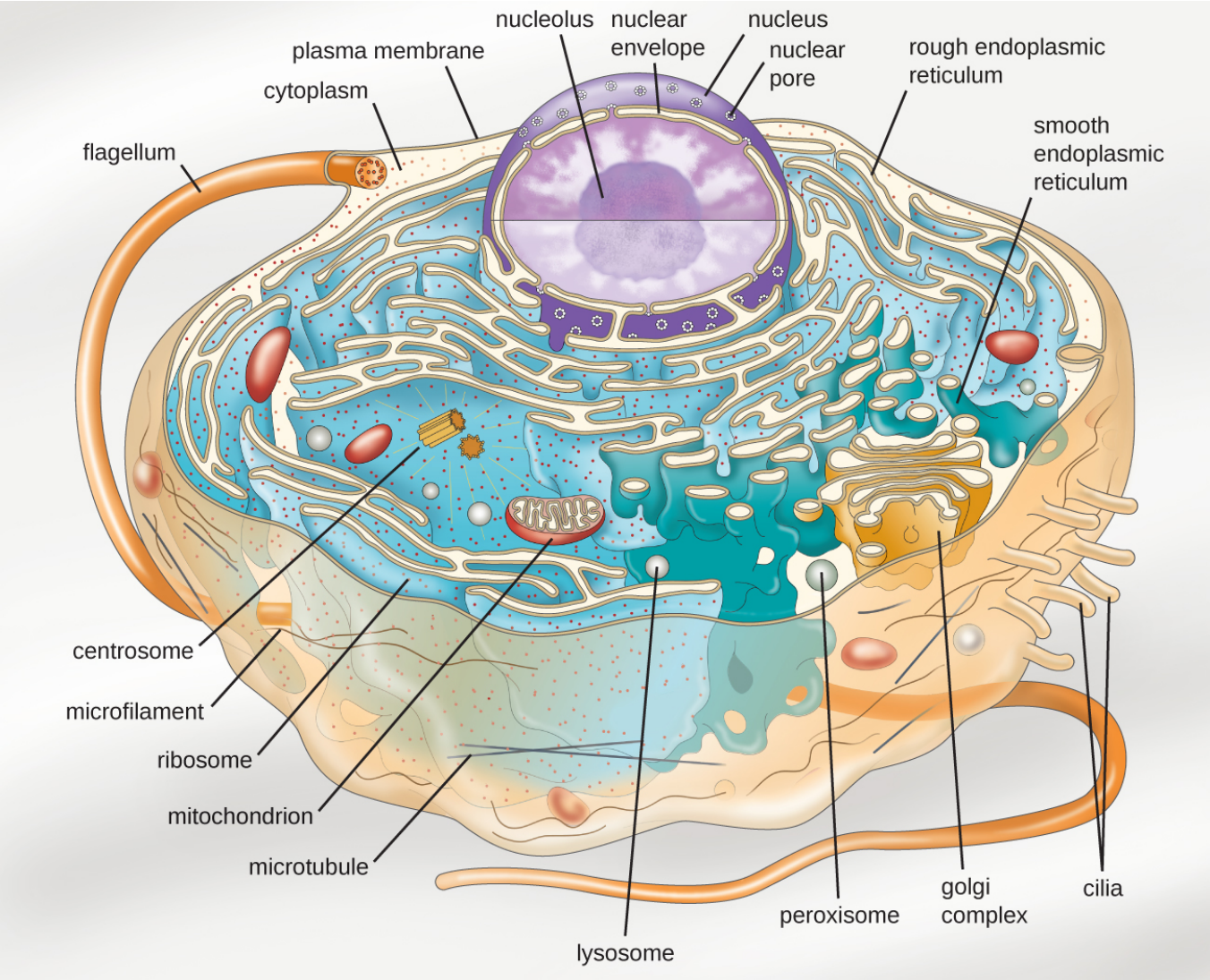


Figure 5.1.1: An illustration of a generalized, single-celled eukaryotic organism. Note that cells of eukaryotic organisms vary greatly in terms of structure and function, and a particular cell may not have all of the structures shown here.

Table 5.1.1: Summary of Cell Structures.

Cell Structure	Prokaryotes		Eukaryotes
	Bacteria	Archaea	
Size	~0.5–1 μM	~0.5–1 μM	~5–20 μM
Surface area-to-volume ratio	High	High	Low
Nucleus	No	No	Yes
Genome characteristics	• Single chromosome • Circular • Haploid • Lacks histones	• Single chromosome • Circular • Haploid • Contains histones	• Multiple chromosomes • Linear • Haploid or diploid • Contains histones
Cell division	Binary fission	Binary fission	Mitosis, meiosis
Membrane lipid composition	• Ester-linked • Straight-chain fatty acids • Bilayer	• Ether-linked • Branched isoprenoids • Bilayer or monolayer	• Ester-linked • Straight-chain fatty acids • Sterols • Bilayer
Cell wall composition	• Peptidoglycan, or • None	• Pseudopeptidoglycan, or • Glycopeptide, or • Polysaccharide, or • Protein (S-layer), or • None	• Cellulose (plants, some algae) • Chitin (molluscs, insects, crustaceans, and fungi) • Silica (some algae) • Most others lack cell walls
Motility structures	Rigid spiral flagella composed of flagellin	Rigid spiral flagella composed of archaeal flagellins	Flexible flagella and cilia composed of microtubules
Membrane-bound organelles	No	No	Yes
Endomembrane system	No	No	Yes (ER, Golgi, lysosomes)
Ribosomes	70S	70S	• 80S in cytoplasm and rough ER • 70S in mitochondria, chloroplasts

Cell Morphologies

Eukaryotic cells display a wide variety of different cell morphologies. Possible shapes include spheroid, ovoid, cuboidal, cylindrical, flat, lenticular, fusiform, discoidal, crescent, ring stellate, and polygonal (Figure **5.1.2**). Some eukaryotic cells are irregular in shape, and some are capable of changing shape. The shape of a particular type of eukaryotic cell may be influenced by factors such as its primary function, the organization of its cytoskeleton, the viscosity of its cytoplasm, the rigidity of its cell membrane or cell wall (if it has one), and the physical pressure exerted on it by the surrounding environment and/or adjoining cells.

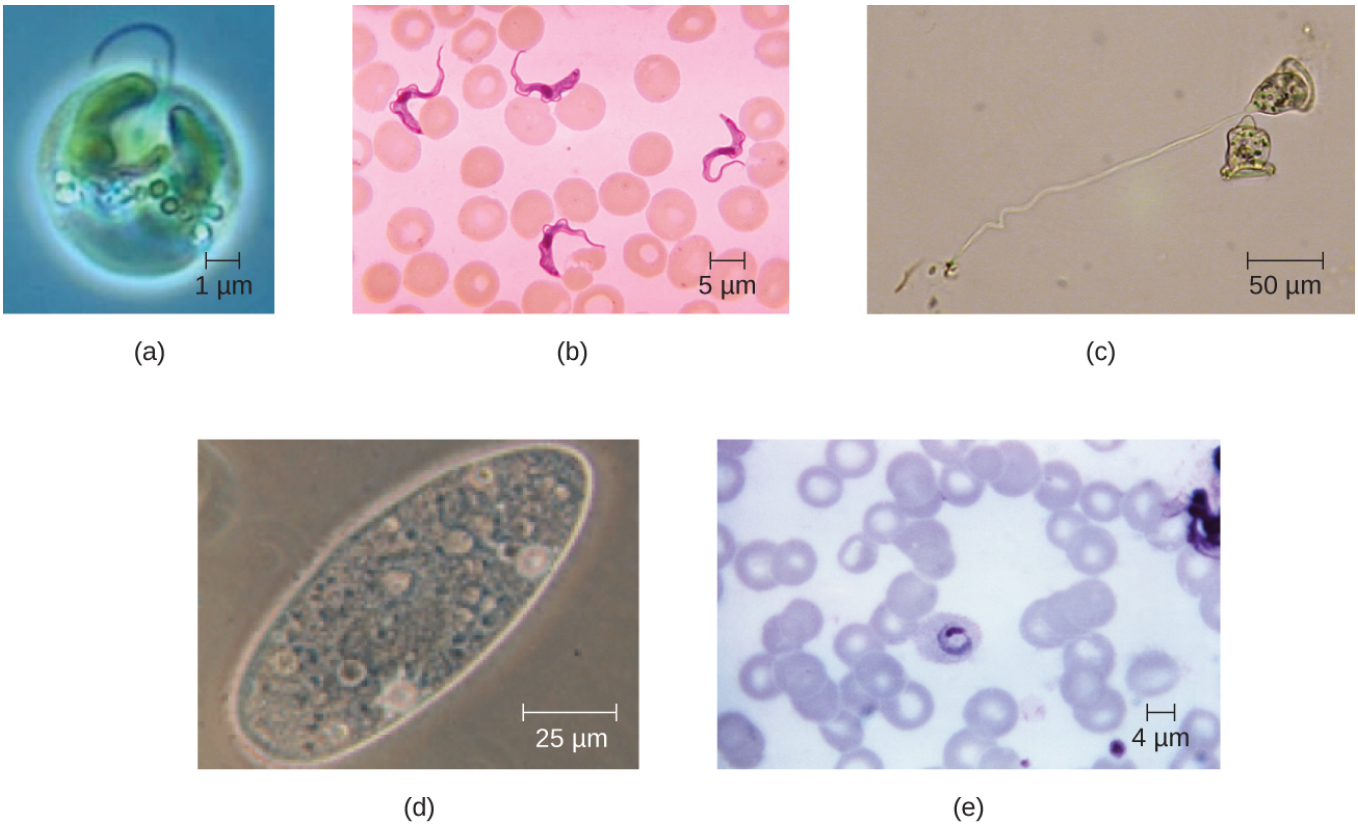


Figure **5.1.2**: Eukaryotic cells come in a variety of cell shapes. (a) Spheroid *Chromulina* alga. (b) Fusiform shaped *Trypanosoma*. (c) Bell-shaped *Vorticella*. (d) Ovoid *Paramecium*. (e) Ring-shaped *Plasmodium ovale*. (credit a: modification of work by NOAA; credit b, e: modification of work by Centers for Disease Control and Prevention).

Nucleus

Unlike prokaryotic cells, in which DNA is loosely contained in the nucleoid region, eukaryotic cells possess a **nucleus**, which is surrounded by a complex nuclear membrane that houses the DNA genome (Figure **5.1.3**). By containing the cell's DNA, the nucleus ultimately controls all activities of the cell and also serves an essential role in reproduction and heredity. Eukaryotic cells typically have their DNA organized into multiple linear chromosomes. The DNA within the nucleus is highly organized and condensed to fit inside the nucleus, which is accomplished by wrapping the DNA around proteins called histones.

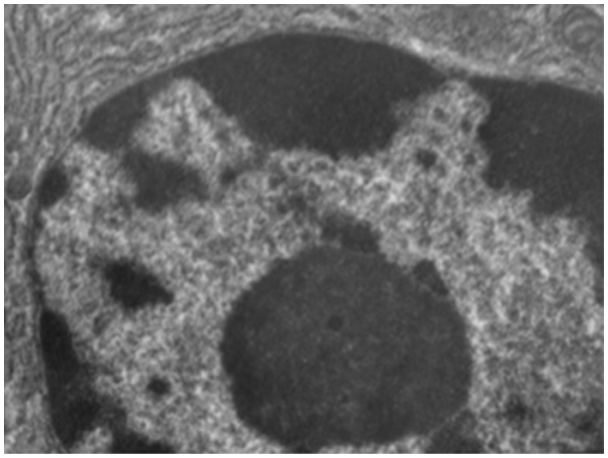


Figure **5.1.3**: Eukaryotic cells have a well-defined nucleus. The nucleus of this mammalian lung cell is the large, dark, oval-shaped structure in the lower half of the image.

Although most eukaryotic cells have only one nucleus, exceptions exist. For example, protozoans of the genus *Paramecium* typically have two complete nuclei: a small nucleus that is used for reproduction (micronucleus) and a large nucleus that directs

cellular metabolism (macronucleus). Additionally, some fungi transiently form cells with two nuclei, called heterokaryotic cells, during sexual reproduction. Cells whose nuclei divide, but whose cytoplasm does not, are called coenocytes.

The nucleus is bound by a complex **nuclear membrane**, often called the **nuclear envelope**, that consists of two distinct lipid bilayers that are contiguous with each other (Figure **5.1.4**). Despite these connections between the inner and outer membranes, each membrane contains unique lipids and proteins on its inner and outer surfaces. The nuclear envelope contains nuclear pores, which are large, rosette-shaped protein complexes that control the movement of materials into and out of the nucleus. The overall shape of the nucleus is determined by the **nuclear lamina**, a meshwork of intermediate filaments found just inside the nuclear envelope membranes. Outside the nucleus, additional intermediate filaments form a looser mesh and serve to anchor the nucleus in position within the cell.

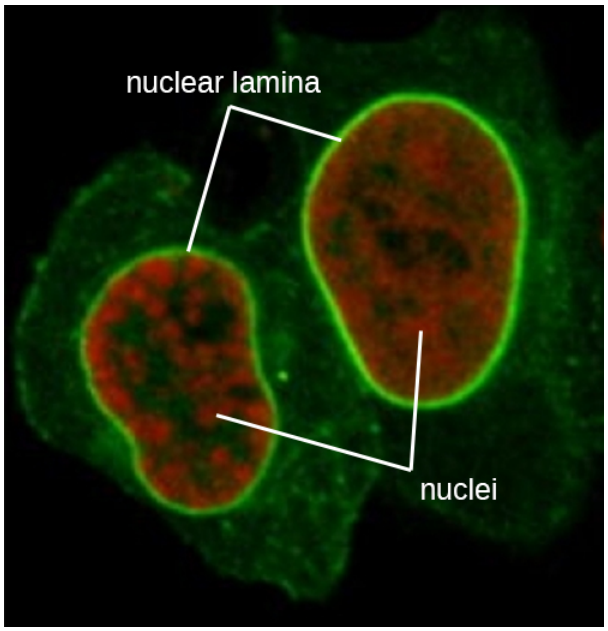


Figure **5.1.4**: In this fluorescent microscope image, all the intermediate filaments have been stained with a bright green fluorescent stain. The nuclear lamina is the intense bright green ring around the faint red nuclei.

Nucleolus

The **nucleolus** is a dense region within the nucleus where ribosomal RNA (rRNA) biosynthesis occurs. In addition, the nucleolus is also the site where assembly of ribosomes begins. Preribosomal complexes are assembled from rRNA and proteins in the nucleolus; they are then transported out to the cytoplasm, where ribosome assembly is completed (Figure **5.1.5**).

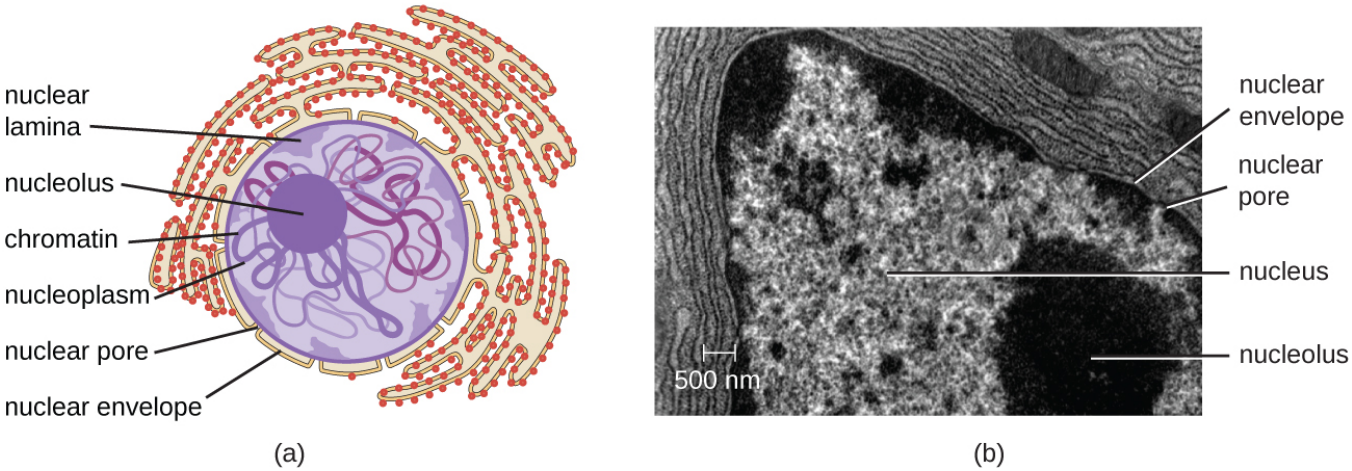


Figure **5.1.5**: (a) The nucleolus is the dark, dense area within the nucleus. It is the site of rRNA synthesis and preribosomal assembly. (b) Electron micrograph showing the nucleolus.

Ribosomes

Ribosomes found in eukaryotic organelles such as mitochondria or chloroplasts have 70S ribosomes—the same size as prokaryotic ribosomes. However, nonorganelle-associated ribosomes in eukaryotic cells are **80S ribosomes**, composed of a 40S small subunit and a 60S large subunit. In terms of size and composition, this makes them distinct from the ribosomes of prokaryotic cells.

The two types of nonorganelle-associated eukaryotic ribosomes are defined by their location in the cell: **free ribosomes** and **membrane-bound ribosomes**. Free ribosomes are found in the cytoplasm and serve to synthesize water-soluble proteins; membrane-bound ribosomes are found attached to the rough endoplasmic reticulum and make proteins for insertion into the cell membrane or proteins destined for export from the cell.

The differences between eukaryotic and prokaryotic ribosomes are clinically relevant because certain antibiotic drugs are designed to target one or the other. For example, cycloheximide targets eukaryotic action, whereas chloramphenicol targets prokaryotic ribosomes.[1] Since human cells are eukaryotic, they generally are not harmed by antibiotics that destroy the prokaryotic ribosomes in bacteria. However, sometimes negative side effects may occur because mitochondria in human cells contain prokaryotic ribosomes.

Endomembrane System

The **endomembrane system**, unique to eukaryotic cells, is a series of membranous tubules, sacs, and flattened disks that synthesize many cell components and move materials around within the cell (Figure **5.1.6**). Because of their larger cell size, eukaryotic cells require this system to transport materials that cannot be dispersed by diffusion alone. The endomembrane system comprises several organelles and connections between them, including the endoplasmic reticulum, Golgi apparatus, lysosomes, and vesicles.

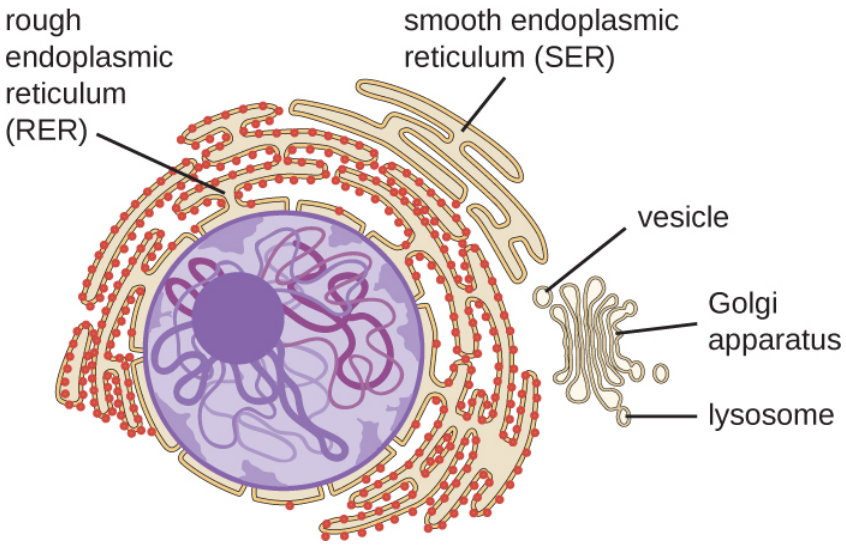


Figure **5.1.6**: The endomembrane system is composed of a series of membranous intracellular structures that facilitate movement of materials throughout the cell and to the cell membrane.

Endoplasmic Reticulum

The **endoplasmic reticulum (ER)** is an interconnected array of tubules and **cisternae** (flattened sacs) with a single lipid bilayer (Figure **5.1.7**). The spaces inside of the cisternae are called **lumen** of the ER. There are two types of ER, **rough endoplasmic reticulum (RER)** and **smooth endoplasmic reticulum (SER)**. These two different types of ER are sites for the synthesis of distinctly different types of molecules. RER is studded with ribosomes bound on the cytoplasmic side of the membrane. These ribosomes make proteins destined for the plasma membrane (Figure Runaway argument for \eatSpaces?). Following synthesis, these proteins are inserted into the membrane of the RER. Small sacs of the RER containing these newly synthesized proteins then bud off as **transport vesicles** and move either to the Golgi apparatus for further processing, directly to the plasma membrane, to the

membrane of another organelle, or out of the cell. Transport vesicles are single-lipid, bilayer, membranous spheres with hollow interiors that carry molecules. SER does not have ribosomes and, therefore, appears “smooth.” It is involved in biosynthesis of lipids, carbohydrate metabolism, and detoxification of toxic compounds within the cell.

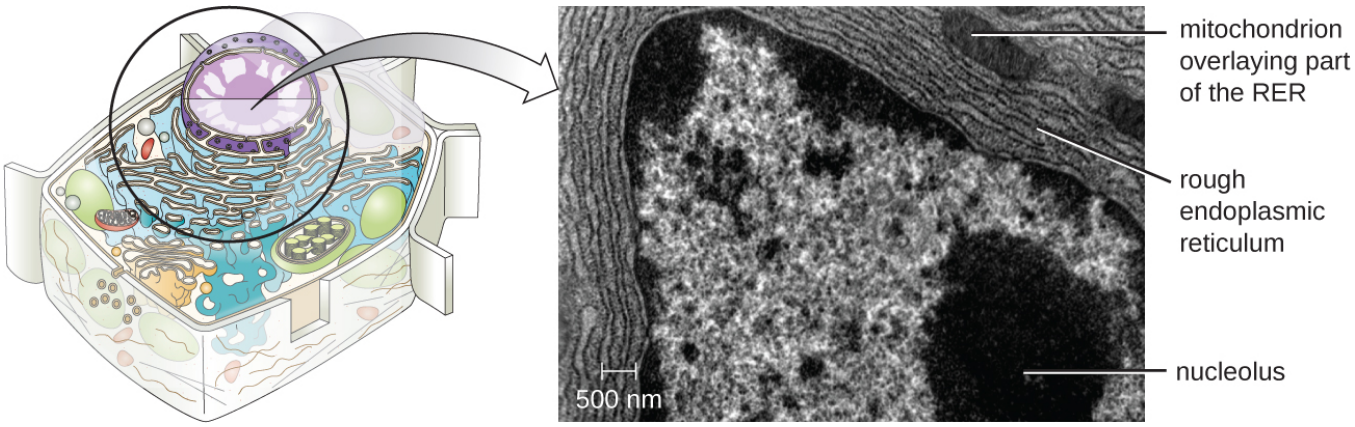


Figure **5.1.7**: The rough endoplasmic reticulum is studded with ribosomes for the synthesis of membrane proteins (which give it its rough appearance).

Golgi Apparatus

The **Golgi apparatus** was discovered within the endomembrane system in 1898 by Italian scientist Camillo Golgi (1843–1926), who developed a novel staining technique that showed stacked membrane structures within the cells of *Plasmodium*, the causative agent of malaria. The Golgi apparatus is composed of a series of membranous disks called dictyosomes, each having a single lipid bilayer, that are stacked together (Figure **5.1.8**).

Enzymes in the Golgi apparatus modify lipids and proteins transported from the ER to the Golgi, often adding carbohydrate components to them, producing glycolipids, glycoproteins, or proteoglycans. Glycolipids and glycoproteins are often inserted into the plasma membrane and are important for signal recognition by other cells or infectious particles. Different types of cells can be distinguished from one another by the structure and arrangement of the glycolipids and glycoproteins contained in their plasma membranes. These glycolipids and glycoproteins commonly also serve as cell surface receptors.

Transport vesicles leaving the ER fuse with a Golgi apparatus on its receiving, or *cis*, face. The proteins are processed within the Golgi apparatus, and then additional transport vesicles containing the modified proteins and lipids pinch off from the Golgi apparatus on its outgoing, or *trans*, face. These outgoing vesicles move to and fuse with the plasma membrane or the membrane of other organelles.

Exocytosis is the process by which **secretory vesicles** (spherical membranous sacs) release their contents to the cell’s exterior (Figure **5.1.8**). All cells have constitutive secretory pathways in which secretory vesicles transport soluble proteins that are released from the cell continually (constitutively). Certain specialized cells also have regulated secretory pathways, which are used to store soluble proteins in secretory vesicles. Regulated secretion involves substances that are only released in response to certain events or signals. For example, certain cells of the human immune system (e.g., mast cells) secrete histamine in response to the presence of foreign objects or pathogens in the body. Histamine is a compound that triggers various mechanisms used by the immune system to eliminate pathogens.

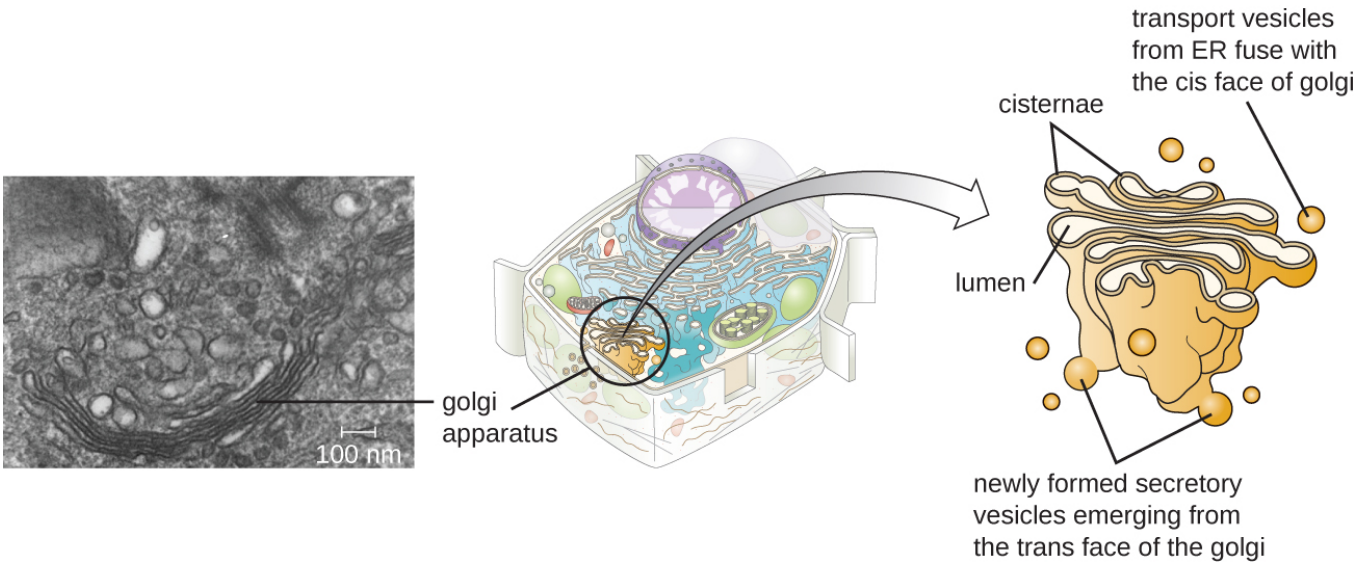


Figure 5.1.8: A transmission electron micrograph (left) of a Golgi apparatus in a white blood cell. The illustration (right) shows the cup-shaped, stacked disks and several transport vesicles. The Golgi apparatus modifies lipids and proteins, producing glycolipids and glycoproteins, respectively, which are commonly inserted into the plasma membrane.

Lysosomes

In the 1960s, Belgian scientist Christian de Duve (1917–2013) discovered **lysosomes**, membrane-bound organelles of the endomembrane system that contain digestive enzymes. Certain types of eukaryotic cells use lysosomes to break down various particles, such as food, damaged organelles or cellular debris, microorganisms, or immune complexes. Compartmentalization of the digestive enzymes within the lysosome allows the cell to efficiently digest matter without harming the cytoplasmic components of the cell.

Peroxisomes

Christian de Duve is also credited with the discovery of **peroxisomes**, membrane-bound organelles that are not part of the endomembrane system (Figure 5.1.9). Peroxisomes form independently in the cytoplasm from the synthesis of peroxin proteins by free ribosomes and the incorporation of these peroxin proteins into existing peroxisomes. Growing peroxisomes then divide by a process similar to binary fission.

Peroxisomes were first named for their ability to produce hydrogen peroxide, a highly reactive molecule that helps to break down molecules such as uric acid, amino acids, and fatty acids. Peroxisomes also possess the enzyme catalase, which can degrade hydrogen peroxide. Along with the SER, peroxisomes also play a role in lipid biosynthesis. Like lysosomes, the compartmentalization of these degradative molecules within an organelle helps protect the cytoplasmic contents from unwanted damage.

The peroxisomes of certain organisms are specialized to meet their particular functional needs. For example, glyoxysomes are modified peroxisomes of yeasts and plant cells that perform several metabolic functions, including the production of sugar molecules. Similarly, glycosomes are modified peroxisomes made by certain trypanosomes, the pathogenic protozoans that cause Chagas disease and African sleeping sickness.

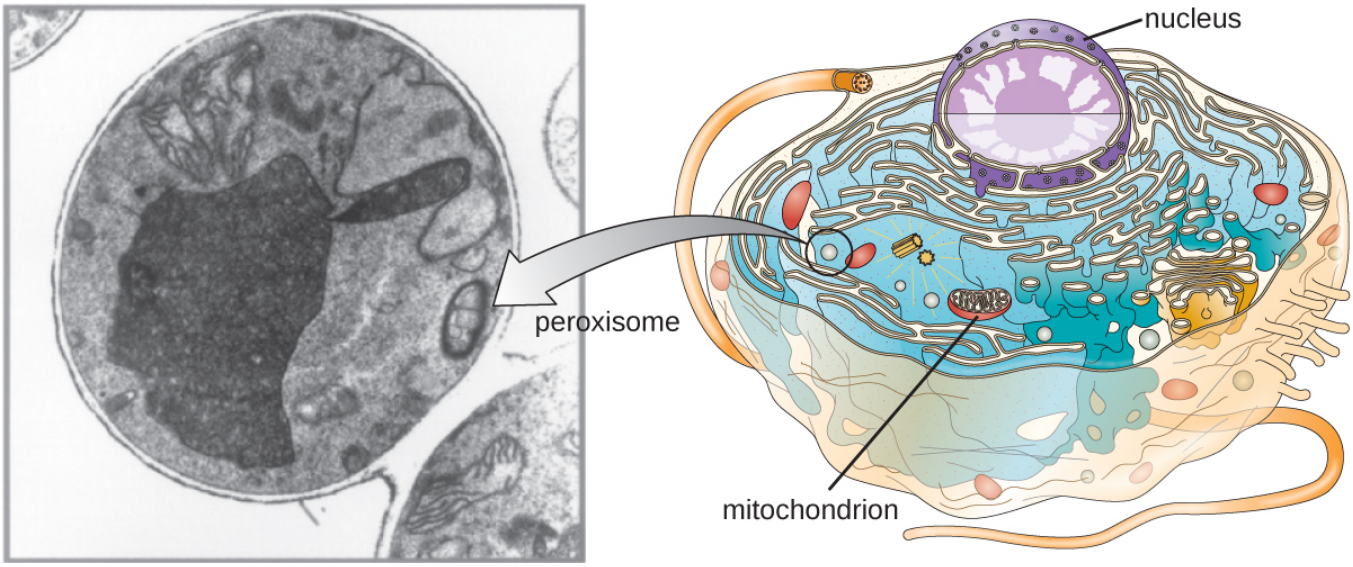


Figure **5.1.9**: A transmission electron micrograph (left) of a cell containing a peroxisome. The illustration (right) shows the location of peroxisomes in a cell. These eukaryotic structures play a role in lipid biosynthesis and breaking down various molecules. They may also have other specialized functions depending on the cell type. (credit "micrograph": modification of work by American Society for Microbiology).

Key Concepts and Summary

- Eukaryotic cells are defined by the presence of a **nucleus** containing the DNA genome and bound by a **nuclear membrane** (or **nuclear envelope**) composed of two lipid bilayers that regulate transport of materials into and out of the nucleus through nuclear pores.
- Eukaryotic cell morphologies vary greatly and may be maintained by various structures, including the cytoskeleton, the cell membrane, and/or the cell wall.
- The **nucleolus**, located in the nucleus of eukaryotic cells, is the site of ribosomal synthesis and the first stages of ribosome assembly.
- Eukaryotic cells contain **80S ribosomes** in the rough endoplasmic reticulum (**membrane bound-ribosomes**) and cytoplasm (**free ribosomes**). They contain 70s ribosomes in mitochondria and chloroplasts.
- Eukaryotic cells have evolved an **endomembrane** system, containing membrane-bound organelles involved in transport. These include vesicles, the endoplasmic reticulum, and the Golgi apparatus.
- The **smooth endoplasmic reticulum** plays a role in lipid biosynthesis, carbohydrate metabolism, and detoxification of toxic compounds. The **rough endoplasmic reticulum** contains membrane-bound 80S ribosomes that synthesize proteins destined for the cell membrane
- The **Golgi apparatus** processes proteins and lipids, typically through the addition of sugar molecules, producing glycoproteins or glycolipids, components of the plasma membrane that are used in cell-to-cell communication.
- **Lysosomes** contain digestive enzymes that break down small particles ingested by **endocytosis**, large particles or cells ingested by **phagocytosis**, and damaged intracellular components.
- The **cytoskeleton**, composed of **microfilaments**, **intermediate filaments**, and **microtubules**, provides structural support in eukaryotic cells and serves as a network for transport of intracellular materials.
- **Centrosomes** are microtubule-organizing centers important in the formation of the mitotic spindle in mitosis.
- **Mitochondria** are the site of cellular respiration. They have two membranes: an outer membrane and an inner membrane with cristae. The mitochondrial matrix, within the inner membrane, contains the mitochondrial DNA, 70S ribosomes, and metabolic enzymes.
- The plasma membrane of eukaryotic cells is structurally similar to that found in prokaryotic cells, and membrane components move according to the fluid mosaic model. However, eukaryotic membranes contain sterols, which alter membrane fluidity, as well as glycoproteins and glycolipids, which help the cell recognize other cells and infectious particles.

- In addition to active transport and passive transport, eukaryotic cell membranes can take material into the cell via **endocytosis**, or expel matter from the cell via **exocytosis.**
- Cells of fungi, algae, plants, and some protists have a **cell wall,** whereas cells of animals and some protozoans have a sticky **extracellular matrix** that provides structural support and mediates cellular signaling.
- Eukaryotic flagella are structurally distinct from prokaryotic flagella but serve a similar purpose (locomotion). **Cilia**are structurally similar to eukaryotic flagella, but shorter; they may be used for locomotion, feeding, or movement of extracellular particles.

Footnotes

1. 1 A.E. Barnhill, M.T. Brewer, S.A. Carlson. “Adverse Effects of Antimicrobials via Predictable or Idiosyncratic Inhibition of Host Mitochondrial Components.” *Antimicrobial Agents and Chemotherapy* 56 no. 8 (2012):4046–4051.
2. 2 Fuchs E, Cleveland DW. “A Structural Scaffolding of Intermediate Filaments in Health and Disease.” *Science* 279 no. 5350 (1998):514–519.
3. 3 E. Fuchs, D.W. Cleveland. “A Structural Scaffolding of Intermediate Filaments in Health and Disease.” *Science* 279 no. 5350 (1998):514–519.
4. 4 E. Fuchs, D.W. Cleveland. “A Structural Scaffolding of Intermediate Filaments in Health and Disease.” *Science* 279 no. 5350 (1998):514–519.
5. 5 N. Yarlett, J.H.P. Hackstein. “Hydrogenosomes: One Organelle, Multiple Origins.” *BioScience* 55 no. 8 (2005):657–658.
6. 6 M. Dudzick. “Protists.” OpenStax CNX. November 27, 2013. http://cnx.org/contents/f7048bb6-e46...ef291cf7049c@1

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5.2: Eukaryotic Cell Anatomy

5.02: Eukaryotic Cell Anatomy

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5.3: The Cytoplasmic Membrane

5.03: The Cytoplasmic Membrane

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5.4: The Cell Wall

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5.5: The Endomembrane System

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5.5A: The Nucleus

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5.5B: The Endoplasmic Reticulum

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5.5C: The Golgi Complex

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5.6: Other Internal Membrane-Bound Organelles

5.06: Other Internal Membrane-Bound Organelles

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5.6A: Mitochondria

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5.6B: Chloroplasts

5.6B: Chloroplasts

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5.6C: Lysosomes, Peroxisomes, Vacuoles, and Vesicles

5.6C: Lysosomes Peroxisomes Vacuoles and Vesicles

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5.7: Ribosomes

5.07: Ribosomes

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5.8: The Cytoskeleton

5.08: The Cytoskeleton

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5.9: Flagella and Cilia

5.09: Flagella and Cilia

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5.10: The Endosymbiotic Theory

5.10: The Endosymbiotic Theory

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5.E: The Cell (Exercises)

5.1: Unique Characteristics of Eukaryotic Cells

Eukaryotic cells are defined by the presence of a nucleus containing the DNA genome and bound by a nuclear membrane (or nuclear envelope) composed of two lipid bilayers that regulate transport of materials into and out of the nucleus through nuclear pores. Eukaryotic cell morphologies vary greatly and may be maintained by various structures, including the cytoskeleton, the cell membrane, and/or the cell wall. The nucleolus in the nucleus of eukaryotic cells is the site of ribosomal synthesis.

Multiple Choice

Which of the following organelles is not part of the endomembrane system?

A. endoplasmic reticulum
B. Golgi apparatus
C. lysosome
D. peroxisome

Answer

D

Which type of cytoskeletal fiber is important in the formation of the nuclear lamina?

A. microfilaments
B. intermediate filaments
C. microtubules
D. fibronectin

Answer

B

Sugar groups may be added to proteins in which of the following?

A. smooth endoplasmic reticulum
B. rough endoplasmic reticulum
C. Golgi apparatus
D. lysosome

Answer

C

Which of the following structures of a eukaryotic cell is not likely derived from endosymbiotic bacterium?

A. mitochondrial DNA
B. mitochondrial ribosomes
C. inner membrane
D. outer membrane

Answer

D

Which type of nutrient uptake involves the engulfment of small dissolved molecules into vesicles?

A. active transport
B. pinocytosis
C. receptor-mediated endocytosis

D. facilitated diffusion

Answer

B

Which of the following is not composed of microtubules?

A. desmosomes
B. centrioles
C. eukaryotic flagella
D. eukaryotic cilia

Answer

A

True/False

Mitochondria in eukaryotic cells contain ribosomes that are structurally similar to those found in prokaryotic cells.

Answer

True

Fill in the Blank

Peroxisomes typically produce _____, a harsh chemical that helps break down molecules.

Answer

hydrogen peroxide

Microfilaments are composed of _____ monomers.

Answer

actin

Short Answer

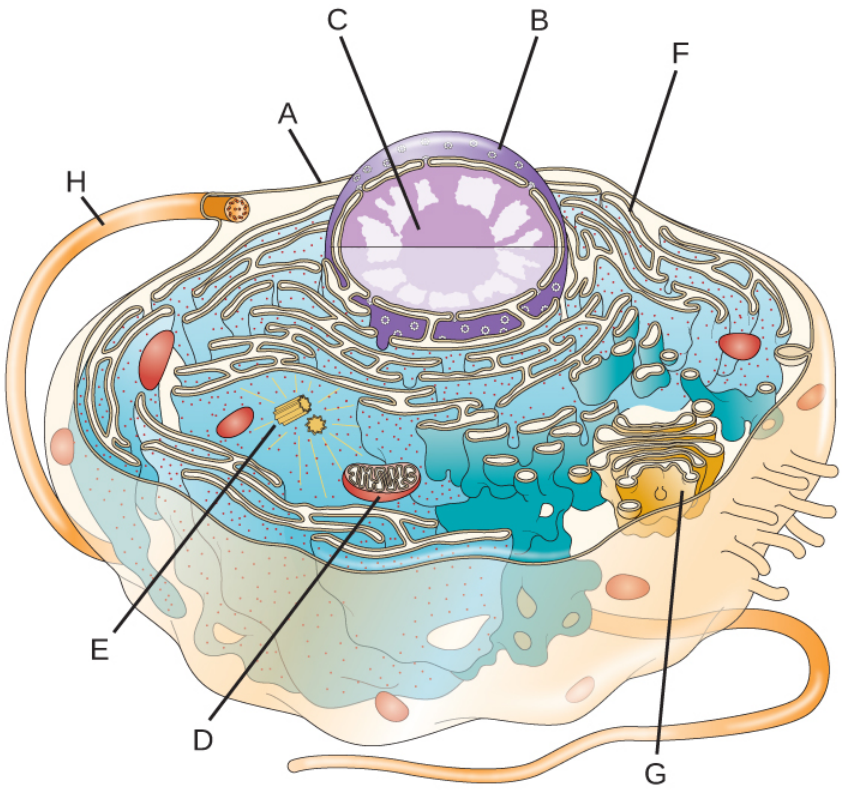
What existing evidence supports the theory that mitochondria are of prokaryotic origin?

Why do eukaryotic cells require an endomembrane system?

Name at least two ways that prokaryotic flagella are different from eukaryotic flagella.

Critical Thinking

Label the lettered parts of this eukaryotic cell.



How are peroxisomes more like mitochondria than like the membrane-bound organelles of the endomembrane system? How do they differ from mitochondria?

Why must the functions of both lysosomes and peroxisomes be compartmentalized?

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5.E: The Eukaryotic Cell (Exercises)

5.E: The Eukaryotic Cell (Exercises)

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CHAPTER OVERVIEW

6: Prokaryotic Pathogens

Scientists have studied prokaryotes for centuries, but it wasn't until 1966 that scientist Thomas Brock (1926–) discovered that certain bacteria can live in boiling water. This led many to wonder whether prokaryotes may also live in other extreme environments, such as at the bottom of the ocean, at high altitudes, or inside volcanoes, or even on other planets.

Prokaryotes have an important role in changing, shaping, and sustaining the entire biosphere. They can produce proteins and other substances used by molecular biologists in basic research and in medicine and industry. For example, the bacterium *Shewanella* lives in the deep sea, where oxygen is scarce. It grows long appendages, which have special sensors used to seek the limited oxygen in its environment. It can also digest toxic waste and generate electricity. Other species of prokaryotes can produce more oxygen than the entire Amazon rainforest, while still others supply plants, animals, and humans with usable forms of nitrogen; and inhabit our body, protecting us from harmful microorganisms and producing some vitally important substances. This chapter will examine the diversity, structure, and function of prokaryotes.

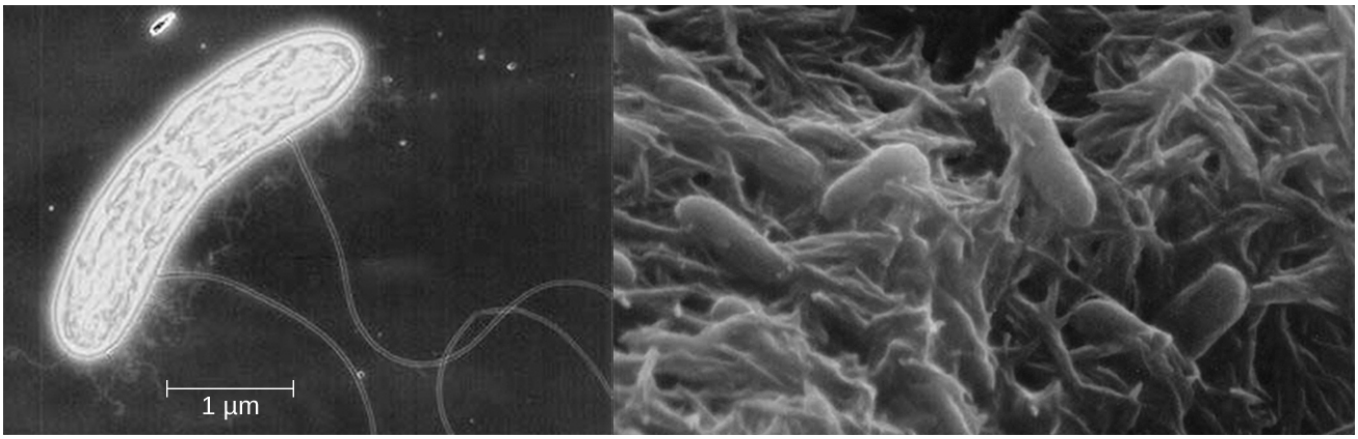


Figure **6.1**: The bacterium *Shewanella* lives in the deep sea, where there is little oxygen diffused in the water. It is able to survive in this harsh environment by attaching to the sea floor and using long appendages, called "nanocables," to sense oxygen. (credit a: modification of work by NASA; credit b: modification of work by Liza Gross)

6.1: Prokaryote Habitats, Relationships, and Microbiomes
6.2: Proteobacteria
6.3: Nonproteobacteria Gram-negative Bacteria and Phototrophic Bacteria
6.4: Gram-positive Bacteria
6.5: Deeply Branching Bacteria
6.6: Archaea
6.E: Prokaryotic Diversity (Exercises)

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6.1: Prokaryote Habitats, Relationships, and Microbiomes

Learning Objectives

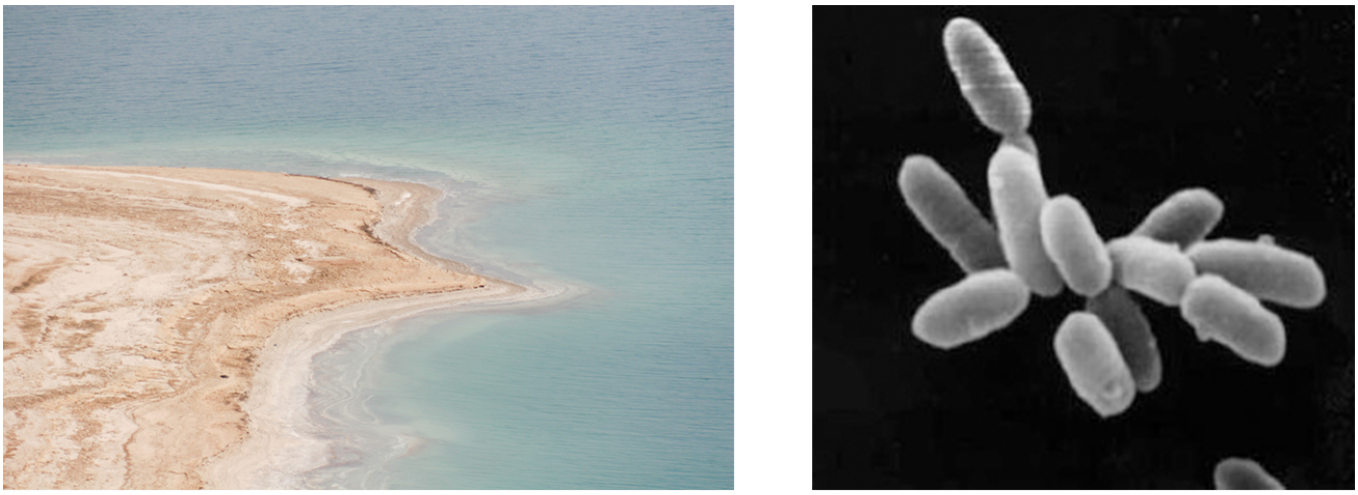
- Identify and describe unique examples of prokaryotes in various habitats on earth
- Identify and describe symbiotic relationships
- Compare normal/commensal/resident microbiota to transient microbiota
- Explain how prokaryotes are classified

Prokaryote Habitats and Functions

Prokaryotes are **ubiquitous**. They can be found everywhere on our planet, even in hot springs, in the Antarctic ice shield, and under extreme pressure two miles under water. One bacterium, *Paracoccus denitrificans*, has even been shown to survive when scientists removed it from its native environment (soil) and used a centrifuge to subject it to forces of gravity as strong as those found on the surface of Jupiter.

Prokaryotes also are abundant on and within the human body. According to a report by National Institutes of Health, prokaryotes, especially bacteria, outnumber human cells 10:1.[1] More recent studies suggest the ratio could be closer to 1:1, but even that ratio means that there are a great number of bacteria within the human body.[2] Bacteria thrive in the human mouth, nasal cavity, throat, ears, gastrointestinal tract, and vagina. Large colonies of bacteria can be found on healthy human skin, especially in moist areas (armpits, navel, and areas behind ears). However, even drier areas of the skin are not free from bacteria.

The existence of prokaryotes is very important for the stability and thriving of ecosystems. For example, they are a necessary part of soil formation and stabilization processes through the breakdown of organic matter and development of biofilms. One gram of soil contains up to 10 billion microorganisms (most of them prokaryotic) belonging to about 1,000 species. Many species of bacteria use substances released from plant roots, such as acids and carbohydrates, as nutrients. The bacteria metabolize these plant substances and release the products of bacterial metabolism back to the soil, forming humus and thus increasing the soil's fertility. In salty lakes such as the Dead Sea (Figure 6.1.1), salt-loving halobacteria decompose dead brine shrimp and nourish young brine shrimp and flies with the products of bacterial metabolism.



(a) (b)

Figure 6.1.1: (a) Some prokaryotes, called halophiles, can thrive in extremely salty environments such as the Dead Sea, pictured here. (b) The archaeon *Halobacterium salinarum*, shown here in an electron micrograph, is a halophile that lives in the Dead Sea. (credit a: modification of work by Jullen Menichini; credit b: modification of work by NASA)

In addition to living in the ground and the water, prokaryotic microorganisms are abundant in the air, even high in the atmosphere. There may be up to 2,000 different kinds of bacteria in the air, similar to their diversity in the soil.

Prokaryotes can be found everywhere on earth because they are extremely resilient and adaptable. They are often metabolically flexible, which means that they might easily switch from one energy source to another, depending on the availability of the sources, or from one metabolic pathway to another. For example, certain prokaryotic cyanobacteria can switch from a conventional type of lipid metabolism, which includes production of fatty aldehydes, to a different type of lipid metabolism that generates biofuel, such as fatty acids and wax esters. Groundwater bacteria store complex high-energy carbohydrates when grown in pure groundwater, but they metabolize these molecules when the groundwater is enriched with phosphates. Some bacteria get their energy by reducing sulfates into sulfides, but can switch to a different metabolic pathway when necessary, producing acids and free hydrogen ions.

Prokaryotes perform functions vital to life on earth by capturing (or “fixing”) and recycling elements like carbon and nitrogen. Organisms such as animals require organic carbon to grow, but, unlike prokaryotes, they are unable to use inorganic carbon sources like carbon dioxide. Thus, animals rely on prokaryotes to convert carbon dioxide into organic carbon products that they can use. This process of converting carbon dioxide to organic carbon products is called **carbon fixation**.

Plants and animals also rely heavily on prokaryotes for **nitrogen fixation**, the conversion of atmospheric nitrogen into ammonia, a compound that some plants can use to form many different biomolecules necessary to their survival. Bacteria in the genus *Rhizobium*, for example, are nitrogen-fixing bacteria; they live in the roots of legume plants such as clover, alfalfa, and peas (Figure 6.1.2). Ammonia produced by *Rhizobium* helps these plants to survive by enabling them to make building blocks of nucleic acids. In turn, these plants may be eaten by animals—sustaining their growth and survival—or they may die, in which case the products of nitrogen fixation will enrich the soil and be used by other plants.

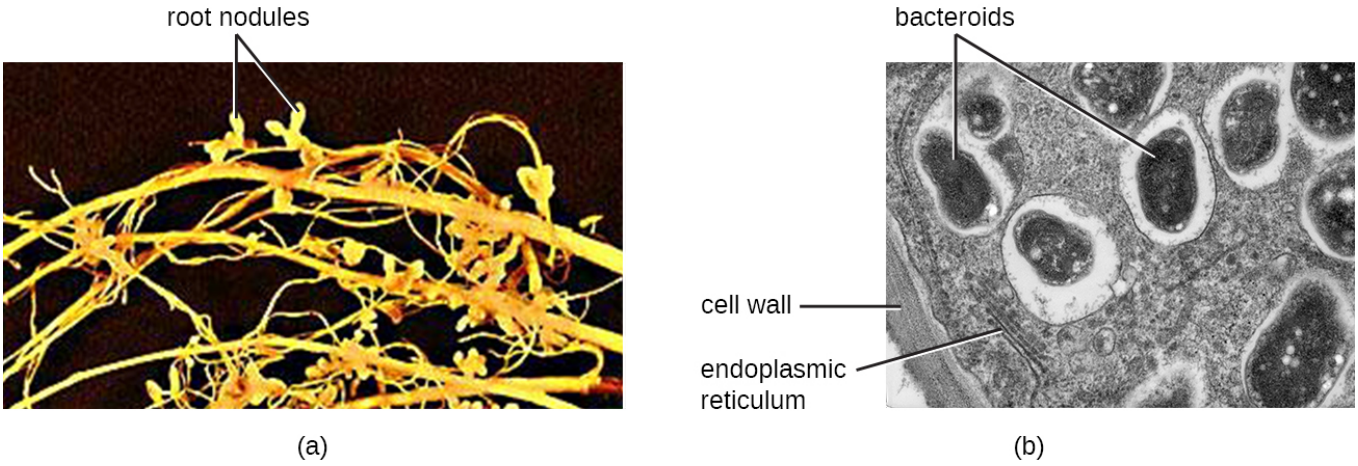


Figure **6.1.2**: (a) Nitrogen-fixing bacteria such as *Rhizobium* live in the root nodules of legumes such as clover. (b) This micrograph of the root nodule shows bacteroids (bacterium-like cells or modified bacterial cells) within the plant cells. The bacteroids are visible as darker ovals within the larger plant cell. (credit a: modification of work by USDA)

Another positive function of prokaryotes is in cleaning up the environment. Recently, some researchers focused on the diversity and functions of prokaryotes in manmade environments. They found that some bacteria play a unique role in degrading toxic chemicals that pollute water and soil.[3]

Despite all of the positive and helpful roles prokaryotes play, some are **human pathogens** that may cause illness or infection when they enter the body. In addition, some bacteria can contaminate food, causing spoilage or foodborne illness, which makes them subjects of concern in food preparation and safety. Less than 1% of prokaryotes (all of them bacteria) are thought to be human pathogens, but collectively these species are responsible for a large number of the diseases that afflict humans.

Besides pathogens, which have a direct impact on human health, prokaryotes also affect humans in many indirect ways. For example, prokaryotes are now thought to be key players in the processes of **climate change**. In recent years, as temperatures in the earth’s polar regions have risen, soil that was formerly frozen year-round (permafrost) has begun to thaw. Carbon trapped in the permafrost is gradually released and metabolized by prokaryotes. This produces massive amounts of carbon dioxide and methane, greenhouse gases that escape into the atmosphere and contribute to the greenhouse effect.

Key Concepts and Summary

- Prokaryotes are unicellular microorganisms whose cells have no nucleus.
- Prokaryotes can be found everywhere on our planet, even in the most extreme environments.
- Prokaryotes are very flexible metabolically, so they are able to adjust their feeding to the available natural resources.
- Prokaryotes live in **communities** that interact among themselves and with large organisms that they use as hosts (including humans).
- The totality of forms of prokaryotes (particularly bacteria) living on the human body is called the human microbiome, which varies between regions of the body and individuals, and changes over time.
- The totality of forms of prokaryotes (particularly bacteria) living in a certain region of the human body (e.g., mouth, throat, gut, eye, vagina) is called the **microbiota** of this region.
- Prokaryotes are classified into domains Archaea and Bacteria.
- In recent years, the traditional approaches to classification of prokaryotes have been supplemented by approaches based on molecular genetics.

Footnotes

1. 1 Medical Press. “Mouth Bacteria Can Change Their Diet, Supercomputers Reveal.” August 12, 2014. medicalxpress.com/news/2014-0...rs-reveal.html. Accessed February 24, 2015.
2. 2 A. Abbott. “Scientists Bust Myth That Our Bodies Have More Bacteria Than Human Cells: Decades-Old Assumption about Microbiota Revisited.” *Nature*. http://www.nature.com/news/scientist...-cells-1.19136. Accessed June 3, 2016.
3. 3 A.M. Kravetz “Unique Bacteria Fights Man-Made Chemical Waste.” 2012. www.livescience.com/25181-bac...s-nsf-bts.html. Accessed March 9, 2015.
4. 4 E.M. Bik et al. “Bacterial Diversity in the Oral Cavity of 10 Healthy Individuals.” *The ISME Journal* 4 no. 8 (2010):962–974.
5. 5 C.C. Booijink et al. “High Temporal and Intra-Individual Variation Detected in the Human Ileal Microbiota.” *Environmental Microbiology* 12 no. 12 (2010):3213–3227.
6. 6 National Institutes of Health. “Human Microbiome Project. Overview.” commonfund.nih.gov/hmp/overview. Accessed June 7, 2016.
7. 7 Q. Dong et al. “Diversity of Bacteria at Healthy Human Conjunctiva.” *Investigative Ophthalmology & Visual Science* 52 no. 8 (2011):5408–5413.
8. 8 F.E. Dewhirst et al. “The Human Oral Microbiome.” *Journal of Bacteriology* 192 no. 19 (2010):5002–5017.
9. 9 J.C. Lagier et al. “Microbial Culturomics: Paradigm Shift in the Human Gut Microbiome Study.” *Clinical Microbiology and Infection* 18 no. 12 (2012):1185–1193.

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6.2: Proteobacteria

Learning Objectives

- Describe the unique features of each class within the phylum Proteobacteria: Alphaproteobacteria, Betaproteobacteria, Gammaproteobacteria, Deltaproteobacteria, and Epsilonproteobacteria
- Give an example of a bacterium in each class of Proteobacteria

In 1987, the American microbiologist Carl Woese (1928–2012) suggested that a large and diverse group of bacteria that he called "purple bacteria and their relatives" should be defined as a separate phylum within the domain Bacteria based on the similarity of the nucleotide sequences in their genome.[1] This phylum of gram-negative bacteria subsequently received the name **Proteobacteria**. It includes many bacteria that are part of the normal human microbiota as well as many pathogens. The Proteobacteria are further divided into five classes: Alphaproteobacteria, Betaproteobacteria, Gammaproteobacteria, Deltaproteobacteria, and Epsilonproteobacteria.

Alphaproteobacteria

The first class of Proteobacteria is the **Alphaproteobacteria**. The unifying characteristic of this class is that they are **oligotrophs**, organisms capable of living in low-nutrient environments such as deep oceanic sediments, glacial ice, or deep undersurface soil.

Among the Alphaproteobacteria are two taxa, chlamydias and rickettsias, that are **obligate intracellular pathogens**, meaning that part of their life cycle must occur inside other cells called host cells. When not growing inside a host cell, *Chlamydia* and *Rickettsia* are metabolically inactive outside of the host cell. They cannot synthesize their own adenosine triphosphate (ATP), and, therefore, rely on cells for their energy needs.

Rickettsia spp. include a number of serious human pathogens. For example, *R. rickettsii* causes Rocky Mountain spotted fever, a life-threatening form of meningoencephalitis (inflammation of the membranes that wrap the brain). *R. rickettsii* infects ticks and can be transmitted to humans via a bite from an infected tick (Figure **6.2.1**).

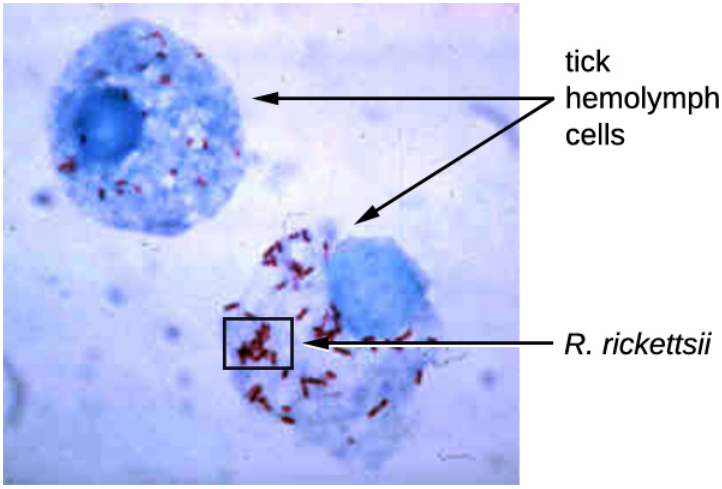


Figure **6.2.1**: Rickettsias require special staining methods to see them under a microscope. Here, *R. rickettsii*, which causes Rocky Mountain spotted fever, is shown infecting the cells of a tick. (credit: modification of work by Centers for Disease Control and Prevention)

Another species of *Rickettsia*, *R. prowazekii*, is spread by lice. It causes epidemic typhus, a severe infectious disease common during warfare and mass migrations of people. *R. prowazekii* infects human endothelium cells, causing inflammation of the inner lining of blood vessels, high fever, abdominal pain, and sometimes delirium. A relative, *R. typhi*, causes a less severe disease known as murine or endemic typhus, which is still observed in the southwestern United States during warm seasons.

Chlamydia is another taxon of the Alphaproteobacteria. Members of this genus are extremely resistant to the cellular defenses, giving them the ability to spread from host to host rapidly via elementary bodies. The metabolically and reproductively inactive

elementary bodies are the endospore-like form of intracellular bacteria that enter an epithelial cell, where they become active. Figure **6.2.2** illustrates the life cycle of *Chlamydia*.

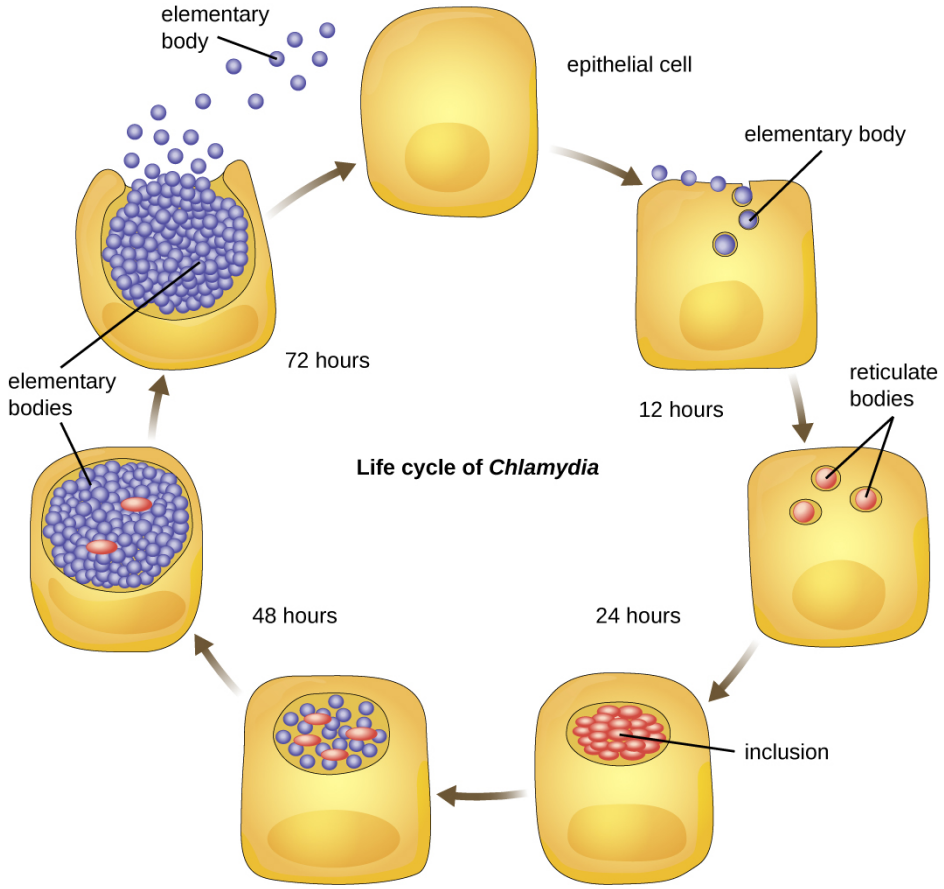


Figure **6.2.2**: Chlamydia begins infection of a host when the metabolically inactive elementary bodies enter an epithelial cell. Once inside the host cell, the elementary bodies turn into active reticulate bodies. The reticulate bodies multiply and release more elementary bodies when the cell dies after the *Chlamydia* uses all of the host cell's ATP. (credit: modification of work by Centers for Disease Control and Prevention)

C. trachomatis is a human pathogen that causes trachoma, a disease of the eyes, often leading to blindness. *C. trachomatis* also causes the sexually transmitted disease lymphogranuloma venereum (LGV). This disease is often mildly symptomatic, manifesting as regional lymph node swelling, or it may be asymptomatic, but it is extremely contagious and is common on college campuses. Table **6.2.1** summarizes the characteristics of important genera of Alphaproteobacteria.

Table **6.2.1**: Class Alphaproteobacteria

Genus	Microscopic Morphology	Unique Characteristics
Agrobacterium	Gram-negative bacillus	Plant pathogen; one species, *A. tumefaciens*, causes tumors in plants
Bartonella	Gram-negative, pleomorphic, flagellated coccobacillus	Facultative intracellular bacteria, transmitted by lice and fleas, cause trench fever and cat scratch disease in humans
Brucella	Gram-negative, small, flagellated coccobacillus	Facultative intracellular bacteria, transmitted by contaminated milk from infected cows, cause brucellosis in cattle and humans
Caulobacter	Gram-negative bacillus	Used in studies on cellular adaptation and differentiation because of its peculiar life cycle (during cell division, forms “swarm” cells and “stalked” cells)
Chlamydia	Gram-negative, coccoid or ovoid bacterium	Obligatory intracellular bacteria; some cause chlamydia, trachoma, and pneumonia
Coxiella	Small, gram-negative bacillus	Obligatory intracellular bacteria; cause Q fever; potential for use as biological weapon
Ehrlichia	Very small, gram-negative, coccoid or ovoid bacteria	Obligatory intracellular bacteria; can be transported from cell to cell; transmitted by ticks; cause ehrlichiosis (destruction of white blood cells and inflammation) in humans and dogs
Hyphomicrobium	Gram-negative bacilli; grows from a stalk	Similar to *Caulobacter*
Methylocystis	Gram-negative, coccoid or short bacilli	Nitrogen-fixing aerobic bacteria
Rhizobium	Gram-negative, rectangular bacilli with rounded ends forming clusters	Nitrogen-fixing bacteria that live in soil and form symbiotic relationship with roots of legumes (e.g., clover, alfalfa, and beans)
Rickettsia	Gram-negative, highly pleomorphic bacteria (may be cocci, rods, or threads)	Obligate intracellular bacteria; transmitted by ticks; may cause Rocky Mountain spotted fever and typhus

Betaproteobacteria

Unlike Alphaproteobacteria, which survive on a minimal amount of nutrients, the class **Betaproteobacteria** are eutrophs (or copiotrophs), meaning that they require a copious amount of organic nutrients. Betaproteobacteria often grow between aerobic and anaerobic areas (e.g., in mammalian intestines). Some genera include species that are human pathogens, able to cause severe, sometimes life-threatening disease. The genus *Neisseria*, for example, includes the bacteria *N. gonorrhoeae*, the causative agent of the STI gonorrhea, and *N. meningitides*, the causative agent of bacterial meningitis.

Neisseria are cocci that live on mucosal surfaces of the human body. They are fastidious, or difficult to culture, and they require high levels of moisture, nutrient supplements, and carbon dioxide. Also, *Neisseria* are microaerophilic, meaning that they require low levels of oxygen. For optimal growth and for the purposes of identification, *Neisseria* spp. are grown on chocolate agar (i.e., agar supplemented by partially hemolyzed red blood cells). Their characteristic pattern of growth in culture is diplococcal: pairs of cells resembling coffee beans (Figure **6.2.3**).

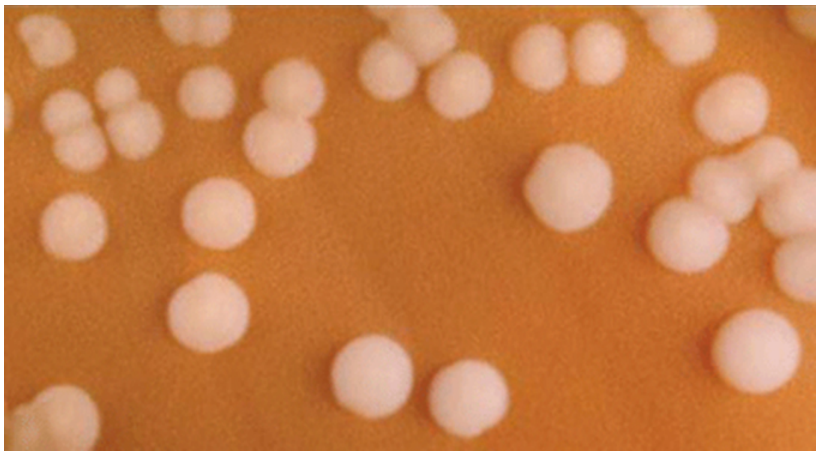


Figure **6.2.3**: Neisseria meningitidis growing in colonies on a chocolate agar plate. (credit: Centers for Disease Control and Prevention)

The pathogen responsible for pertussis (whooping cough) is also a member of Betaproteobacteria. The bacterium *Bordetella pertussis*, from the order Burkholderiales, produces several toxins that paralyze the movement of cilia in the human respiratory tract and directly damage cells of the respiratory tract, causing a severe cough. Table **6.2.2** summarizes the characteristics of important genera of Betaproteobacteria.

Table **6.2.2**: Class Betaproteobacteria

Example Genus	Microscopic Morphology	Unique Characteristics
Bordetella	A small, gram-negative coccobacillus	Aerobic, very fastidious; *B. pertussis* causes pertussis (whooping cough)
Burkholderia	Gram-negative bacillus	Aerobic, aquatic, cause diseases in horses and humans (especially patients with cystic fibrosis); agents of nosocomial infections
Leptothrix	Gram-negative, sheathed, filamentous bacillus	Aquatic; oxidize iron and manganese; can live in wastewater treatment plants and clog pipes
Neisseria	Gram-negative, coffee bean-shaped coccus forming pairs	Require moisture and high concentration of carbon dioxide; oxidase positive, grow on chocolate agar; pathogenic species cause gonorrhea and meningitis
Thiobacillus	Gram-negative bacillus	Thermophilic, acidophilic, strictly aerobic bacteria; oxidize iron and sulfur

Key Concepts and Summary

- **Proteobacteria** is a phylum of gram-negative bacteria discovered by Carl Woese in the 1980s based on nucleotide sequence homology.
- Proteobacteria are further classified into the classes alpha-, beta-, gamma-, delta- and epsilonproteobacteria, each class having separate orders, families, genera, and species.
- **Alphaproteobacteria** are **oligotrophs**. The taxa chlamydias and rickettsias are **obligate intracellular pathogens,** feeding on cells of host organisms; they are metabolically inactive outside of the host cell. Some Alphaproteobacteria can convert atmospheric nitrogen to nitrites, making nitrogen usable by other forms of life.
- **Betaproteobacteria** are **eutrophs**. They include human pathogens of the genus *Neisseria* and the species *Bordetella pertussis*.

Footnotes

1. C.R. Woese. “Bacterial Evolution.” *Microbiological Review* 51 no. 2 (1987):221–271.
2. H. Reichenbach. “Myxobacteria, Producers of Novel Bioactive Substances.” *Journal of Industrial Microbiology & Biotechnology* 27 no. 3 (2001):149–156.
3. S. Suerbaum, P. Michetti. “*Helicobacter pylori* infection.” *New England Journal of Medicine* 347 no. 15 (2002):1175–1186.

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6.3: Nonproteobacteria Gram-negative Bacteria and Phototrophic Bacteria

Learning Objectives

- Describe the unique features of nonproteobacteria gram-negative bacteria
- Give an example of a nonproteobacteria bacterium in each category
- Describe the unique features of phototrophic bacteria
- Identify phototrophic bacteria

The majority of the gram-negative bacteria belong to the phylum Proteobacteria, discussed in the previous section. Those that do not are called the nonproteobacteria. In this section, we will describe three classes of gram-negative nonproteobacteria: the spirochetes, the CFB group, and the Planctomycetes. A diverse group of phototrophic bacteria that includes Proteobacteria and nonproteobacteria will be discussed at the end of this section.

Spirochetes

Spirochetes are characterized by their long (up to 250 µm), spiral-shaped bodies. Most spirochetes are also very thin, which makes it difficult to examine gram-stained preparations under a conventional brightfield microscope. Darkfield fluorescent microscopy is typically used instead. Spirochetes are also difficult or even impossible to culture. They are highly motile, using their axial filament to propel themselves. The axial filament is similar to a flagellum, but it wraps around the cell and runs inside the cell body of a spirochete in the periplasmic space between the outer membrane and the plasma membrane (Figure **6.3.1**).

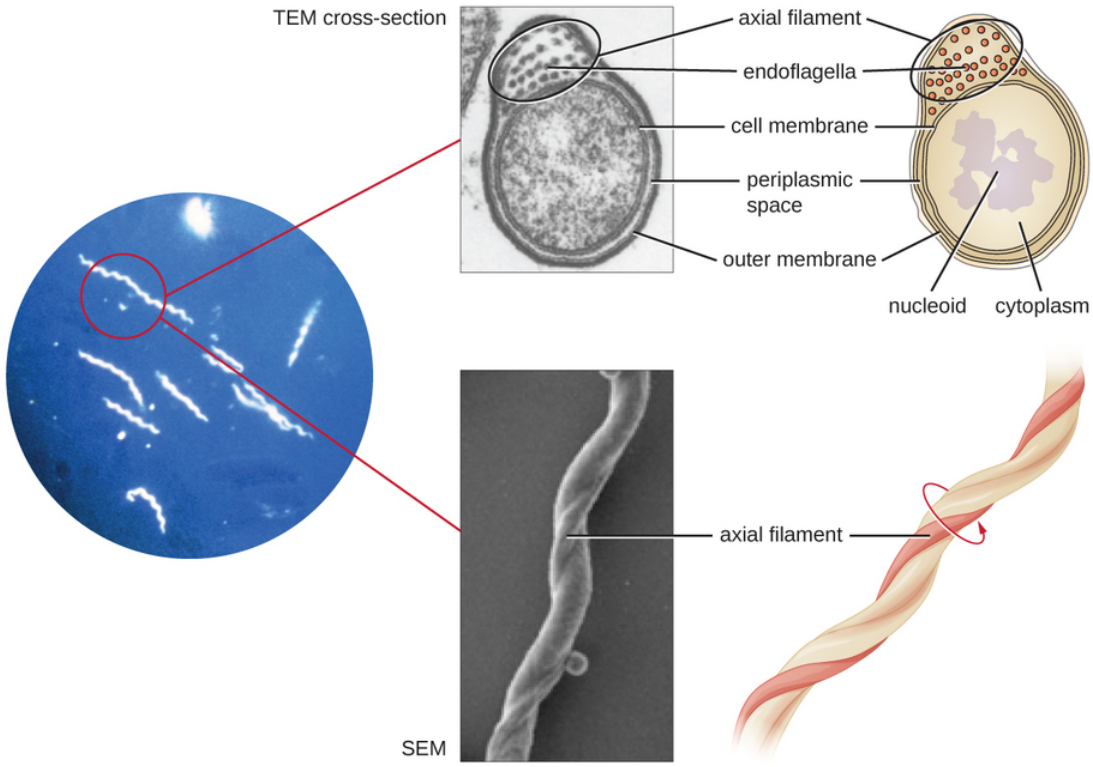


Figure **6.3.1**: Spirochetes are typically observed using darkfield microscopy (left). However, electron microscopy (top center, bottom center) provides a more detailed view of their cellular morphology. The flagella found between the inner and outer membranes of spirochetes wrap around the bacterium, causing a twisting motion used for locomotion. (credit "spirochetes" micrograph: modification of work by Centers for Disease Control and Prevention; credit "SEM/TEM": modification of work by Guyard C, Raffel SJ, Schrumpf ME, Dahlstrom E, Sturdevant D, Ricklefs SM, Martens C, Hayes SF, Fischer ER, Hansen BT, Porcella SF, Schwan TG)

Several genera of spirochetes include human pathogens. For example, the genus *Treponema* includes a species *T. pallidum*, which is further classified into four subspecies: *T. pallidum pallidum*, *T. pallidum pertenue*, *T. pallidum carateum*, and *T. pallidum endemicum*. The subspecies *T. pallidum pallidum* causes the sexually transmitted infection known as syphilis, the third most

prevalent sexually transmitted bacterial infection in the United States, after chlamydia and gonorrhea. The other subspecies of *T. pallidum* cause tropical infectious diseases of the skin, bones, and joints.

Another genus of spirochete, *Borrelia*, contains a number of pathogenic species. *B. burgdorferi* causes Lyme disease, which is transmitted by several genera of ticks (notably *Ixodes* and *Amblyomma*) and often produces a "bull's eye" rash, fever, fatigue, and, sometimes, debilitating arthritis. *B. recurrens* causes a condition known as relapsing fever.

Cytophaga, Fusobacterium, and Bacteroides

The gram-negative nonproteobacteria of the genera *Cytophaga*, *Fusobacterium*, and *Bacteroides* are classified together as a phylum and called the CFB group. Although they are phylogenetically diverse, bacteria of the **CFB group** share some similarities in the sequence of nucleotides in their DNA. They are rod-shaped bacteria adapted to anaerobic environments, such as the tissue of the gums, gut, and rumen of ruminating animals. CFB bacteria are avid fermenters, able to process cellulose in rumen, thus enabling ruminant animals to obtain carbon and energy from grazing.

Cytophaga are motile aquatic bacteria that glide. *Fusobacteria* inhabit the human mouth and may cause severe infectious diseases. The largest genus of the CFB group is *Bacteroides*, which includes dozens of species that are prevalent inhabitants of the human large intestine, making up about 30% of the entire gut microbiome (Figure 6.3.2). One gram of human feces contains up to 100 billion *Bacteroides* cells. Most *Bacteroides* are mutualistic. They benefit from nutrients they find in the gut, and humans benefit from their ability to prevent pathogens from colonizing the large intestine. Indeed, when populations of *Bacteroides* are reduced in the gut—as often occurs when a patient takes antibiotics—the gut becomes a more favorable environment for pathogenic bacteria and fungi, which can cause secondary infections.

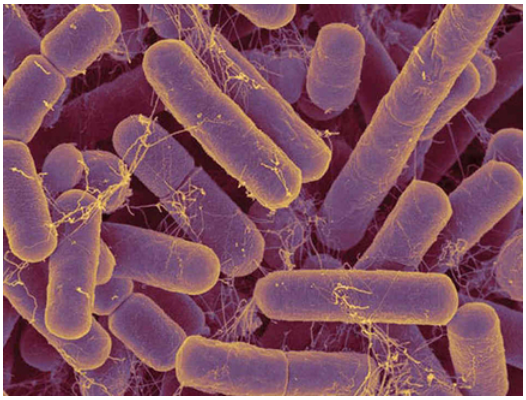


Figure 6.3.2: Bacteroides comprise up to 30% of the normal microbiota in the human gut. (credit: NOAA)

Only a few species of *Bacteroides* are pathogenic. *B. melaninogenicus*, for example, can cause wound infections in patients with weakened immune systems.

Key Concepts and Summary

- Gram-negative nonproteobacteria include the taxa **spirochetes**; the *Cytophaga*, *Fusobacterium*, *Bacteroides* group; Planctomycetes; and many representatives of **phototrophic bacteria.**
- Spirochetes are motile, spiral bacteria with a long, narrow body; they are difficult or impossible to culture.
- Several genera of spirochetes contain human pathogens that cause such diseases as syphilis and Lyme disease.
- *Cytophaga*, *Fusobacterium*, and *Bacteroides* are classified together as a phylum called the **CFB group**. They are rod-shaped anaerobic organoheterotrophs and avid fermenters. *Cytophaga* are aquatic bacteria with the gliding motility. *Fusobacteria* inhabit the human mouth and may cause severe infectious diseases. *Bacteroides* are present in vast numbers in the human gut, most of them being mutualistic but some are pathogenic.

Footnotes

1. 1 R.C. Fuller et al. "Carbon Metabolism in *Chromatium*." *Journal of Biological Chemistry* 236 (1961):2140–2149.

2. 2 T.T. Selao et al. “Comparative Proteomic Studies in *Rhodospirillum rubrum* Grown Under Different Nitrogen Conditions.” *Journal of Proteome Research* 7 no. 8 (2008):3267–3275.
3. 3 A. De los Rios et al. “Ultrastructural and Genetic Characteristics of Endolithic Cyanobacterial Biofilms Colonizing Antarctic Granite Rocks.” *FEMS Microbiology Ecology* 59 no. 2 (2007):386–395.
4. 4 T. Cavalier-Smith. “Membrane Heredity and Early Chloroplast Evolution.” *Trends in Plant Science* 5 no. 4 (2000):174–182.
5. 5 S. Zhang, D.A. Bryant. “The Tricarboxylic Acid Cycle in Cyanobacteria.” *Science* 334 no. 6062 (2011):1551–1553.
6. 6 A. Cain et al. “Cyanobacteria as a Biosorbent for Mercuric Ion.” Bioresource Technology 99 no. 14 (2008):6578–6586.
7. 7 C.S. Ku et al. “Edible Blue-Green Algae Reduce the Production of Pro-Inflammatory Cytokines by Inhibiting NF-κB Pathway in Macrophages and Splenocytes.” *Biochimica et Biophysica Acta* 1830 no. 4 (2013):2981–2988.
8. 8 I. Stewart et al. Cyanobacterial Poisoning in Livestock, Wild Mammals and Birds – an Overview. *Advances in Experimental Medicine and Biology* 619 (2008):613–637.

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6.4: Gram-positive Bacteria

Learning Objectives

- Describe the unique features of each category of high G+C and low G+C gram-positive bacteria
- Identify similarities and differences between high G+C and low G+C bacterial groups
- Give an example of a bacterium of high G+C and low G+C group commonly associated with each category

Prokaryotes are identified as gram-positive if they have a multiple layer matrix of peptidoglycan forming the cell wall. Crystal violet, the primary stain of the Gram stain procedure, is readily retained and stabilized within this matrix, causing gram-positive prokaryotes to appear purple under a brightfield microscope after Gram staining. For many years, the retention of Gram stain was one of the main criteria used to classify prokaryotes, even though some prokaryotes did not readily stain with either the primary or secondary stains used in the Gram stain procedure.

Advances in nucleic acid biochemistry have revealed additional characteristics that can be used to classify gram-positive prokaryotes, namely the guanine to cytosine ratios (G+C) in DNA and the composition of 16S rRNA subunits. Microbiologists currently recognize two distinct groups of gram-positive, or weakly staining gram-positive, prokaryotes. The class Actinobacteria comprises the **high G+C gram-positive bacteria**, which have more than 50% guanine and cytosine nucleotides in their DNA. The class Bacilli comprises **low G+C gram-positive bacteria**, which have less than 50% of guanine and cytosine nucleotides in their DNA.

Actinobacteria: High G+C Gram-Positive Bacteria

The name Actinobacteria comes from the Greek words for *rays* and *small rod*, but Actinobacteria are very diverse. Their microscopic appearance can range from thin filamentous branching rods to coccobacilli. Some Actinobacteria are very large and complex, whereas others are among the smallest independently living organisms. Most Actinobacteria live in the soil, but some are aquatic. The vast majority are aerobic. One distinctive feature of this group is the presence of several different peptidoglycans in the cell wall.

The genus *Actinomyces* is a much studied representative of Actinobacteria. *Actinomyces* spp. play an important role in soil ecology, and some species are human pathogens. A number of *Actinomyces* spp. inhabit the human mouth and are opportunistic pathogens, causing infectious diseases like periodontitis (inflammation of the gums) and oral abscesses. The species *A. israelii* is an anaerobe notorious for causing endocarditis (inflammation of the inner lining of the heart) (Figure **6.4.1**).



Figure **6.4.1**: (a) *Actinomyces israelii* (false-color scanning electron micrograph [SEM]) has a branched structure. (b) *Corynebacterium diphtheria* causes the deadly disease diphtheria. Note the distinctive palisades. (c) The gram-variable bacterium *Gardnerella vaginalis* causes bacterial vaginosis in women. This micrograph shows a Pap smear from a woman with vaginosis. (credit a: modification of work by "GrahamColm"/Wikimedia Commons; credit b: modification of work by Centers for Disease Control and Prevention; credit c: modification of work by Mwakigonja AR, Torres LM, Mwakyoma HA, Kaaya EE)

The genus *Mycobacterium* is represented by bacilli covered with a mycolic acid coat. This waxy coat protects the bacteria from some antibiotics, prevents them from drying out, and blocks penetration by Gram stain reagents (see Staining Microscopic Specimens). Because of this, a special acid-fast staining procedure is used to visualize these bacteria. The genus *Mycobacterium* is

an important cause of a diverse group of infectious diseases. *M. tuberculosis* is the causative agent of tuberculosis, a disease that primarily impacts the lungs but can infect other parts of the body as well. It has been estimated that one-third of the world's population has been infected with *M. tuberculosis* and millions of new infections occur each year. Treatment of *M. tuberculosis* is challenging and requires patients to take a combination of drugs for an extended time. Complicating treatment even further is the development and spread of multidrug-resistant strains of this pathogen.

Another pathogenic species, *M. leprae*, is the cause of Hansen's disease (leprosy), a chronic disease that impacts peripheral nerves and the integrity of the skin and mucosal surface of the respiratory tract. Loss of pain sensation and the presence of skin lesions increase susceptibility to secondary injuries and infections with other pathogens.

Bacteria in the genus *Corynebacterium* contain diaminopimelic acid in their cell walls, and microscopically often form *palisades*, or pairs of rod-shaped cells resembling the letter V. Cells may contain metachromatic granules, intracellular storage of inorganic phosphates that are useful for identification of *Corynebacterium*. The vast majority of *Corynebacterium* spp. are nonpathogenic; however, *C. diphtheria* is the causative agent of diphtheria, a disease that can be fatal, especially in children (Figure **6.4.1**). *C. diphtheria* produces a toxin that forms a pseudomembrane in the patient's throat, causing swelling, difficulty breathing, and other symptoms that can become serious if untreated.

The genus *Bifidobacterium* consists of filamentous anaerobes, many of which are commonly found in the gastrointestinal tract, vagina, and mouth. In fact, *Bifidobacterium* spp. constitute a substantial part of the human gut microbiota and are frequently used as probiotics and in yogurt production.

The genus *Gardnerella*, contains only one species, *G. vaginalis*. This species is defined as "gram-variable" because its small coccobacilli do not show consistent results when Gram stained (Figure **6.4.1**). Based on its genome, it is placed into the high G+C gram-positive group. *G. vaginalis* can cause bacterial vaginosis in women; symptoms are typically mild or even undetectable, but can lead to complications during pregnancy.

Table **6.4.1** summarizes the characteristics of some important genera of Actinobacteria.

Table **6.4.1**: Actinobacteria: High G+C Gram-Positive

Example Genus	Microscopic Morphology	Unique Characteristics
Actinomyces	Gram-positive bacillus; in colonies, shows fungus-like threads (hyphae)	Facultative anaerobes; in soil, decompose organic matter; in the human mouth, may cause gum disease
Arthrobacter	Gram-positive bacillus (at the exponential stage of growth) or coccus (in stationary phase)	Obligate aerobes; divide by "snapping," forming V-like pairs of daughter cells; degrade phenol, can be used in bioremediation
Bifidobacterium	Gram-positive, filamentous actinobacterium	Anaerobes commonly found in human gut microbiota
Corynebacterium	Gram-positive bacillus	Aerobes or facultative anaerobes; form palisades; grow slowly; require enriched media in culture; *C. diphtheriae* causes diphtheria
Frankia	Gram-positive, fungus-like (filamentous) bacillus	Nitrogen-fixing bacteria; live in symbiosis with legumes
Gardnerella	Gram-variable coccobacillus	Colonize the human vagina, may alter the microbial ecology, thus leading to vaginosis
Micrococcus	Gram-positive coccus, form microscopic clusters	Ubiquitous in the environment and on the human skin; oxidase-positive (as opposed to morphologically similar *S. aureus*); some are opportunistic pathogens
Mycobacterium	Gram-positive, acid-fast bacillus	Slow growing, aerobic, resistant to drying and phagocytosis; covered with a waxy coat made of mycolic acid; *M. tuberculosis* causes tuberculosis; *M. leprae* causes leprosy
Nocardia	Weakly gram-positive bacillus; forms acid-fast branches	May colonize the human gingiva; may cause severe pneumonia and inflammation of the skin
Propionibacterium	Gram-positive bacillus	Aerotolerant anaerobe; slow-growing; *P. acnes* reproduces in the human sebaceous glands and may cause or contribute to acne
Rhodococcus	Gram-positive bacillus	Strict aerobe; used in industry for biodegradation of pollutants; *R. fascians* is a plant pathogen, and *R. equi* causes pneumonia in foals
Streptomyces	Gram-positive, fungus-like (filamentous) bacillus	Very diverse genus (>500 species); aerobic, spore-forming bacteria; scavengers, decomposers found in soil (give the soil its "earthy" odor); used in pharmaceutical industry as antibiotic producers (more than two-thirds of clinically useful antibiotics)

Low G+C Gram-positive Bacteria

The low G+C gram-positive bacteria have less than 50% guanine and cytosine in their DNA, and this group of bacteria includes a number of genera of bacteria that are pathogenic.

Clostridia

One large and diverse class of low G+C gram-positive bacteria is Clostridia. The best studied genus of this class is *Clostridium*. These rod-shaped bacteria are generally obligate anaerobes that produce endospores and can be found in anaerobic habitats like soil and aquatic sediments rich in organic nutrients. The endospores may survive for many years.

Clostridium spp. produce more kinds of protein toxins than any other bacterial genus, and several species are human pathogens. *C. perfringens* is the third most common cause of food poisoning in the United States and is the causative agent of an even more serious disease called gas gangrene. Gas gangrene occurs when *C. perfringens* endospores enter a wound and germinate, becoming viable bacterial cells and producing a toxin that can cause the necrosis (death) of tissue. *C. tetani*, which causes tetanus, produces a neurotoxin that is able to enter neurons, travel to regions of the central nervous system where it blocks the inhibition of nerve impulses involved in muscle contractions, and cause a life-threatening spastic paralysis. *C. botulinum* produces botulinum neurotoxin, the most lethal biological toxin known. Botulinum toxin is responsible for rare but frequently fatal cases of botulism. The toxin blocks the release of acetylcholine in neuromuscular junctions, causing flaccid paralysis. In very small concentrations, botulinum toxin has been used to treat muscle pathologies in humans and in a cosmetic procedure to eliminate wrinkles. *C. difficile* is a common source of hospital-acquired infections (Figure **6.4.2**) that can result in serious and even fatal cases of colitis (inflammation of the large intestine). Infections often occur in patients who are immunosuppressed or undergoing antibiotic therapy that alters the normal microbiota of the gastrointestinal tract.

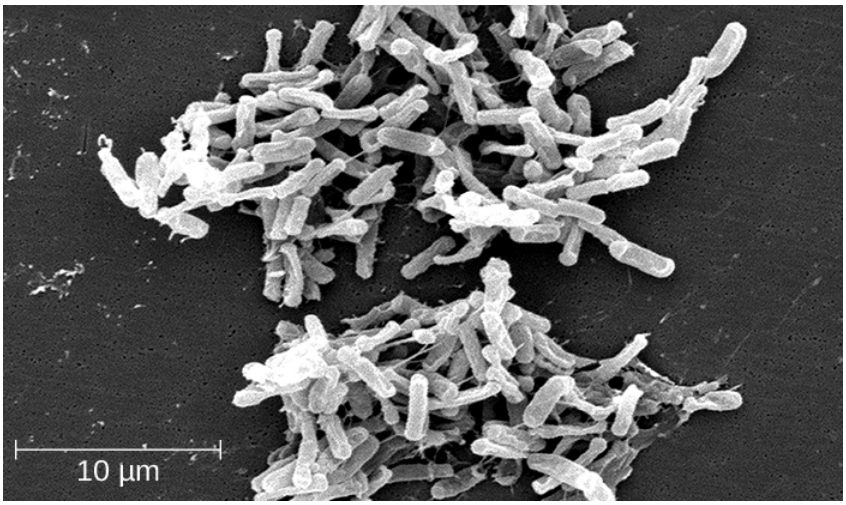


Figure **6.4.2**: Clostridium difficile, a gram-positive, rod-shaped bacterium, causes severe colitis and diarrhea, often after the normal gut microbiota is eradicated by antibiotics. (credit: modification of work by Centers for Disease Control and Prevention)

Lactobacillales

The order Lactobacillales comprises low G+C gram-positive bacteria that include both bacilli and cocci in the genera *Lactobacillus*, *Leuconostoc*, *Enterococcus*, and *Streptococcus*. Bacteria of the latter three genera typically are spherical or ovoid and often form chains.

Streptococcus, the name of which comes from the Greek word for *twisted chain*, is responsible for many types of infectious diseases in humans. Species from this genus, often referred to as streptococci, are usually classified by serotypes called Lancefield groups, and by their ability to lyse red blood cells when grown on blood agar.

S. pyogenes belongs to the Lancefield group A, β-hemolytic *Streptococcus*. This species is considered a pyogenic pathogen because of the associated pus production observed with infections it causes (Figure **6.4.3**). *S. pyogenes* is the most common cause of bacterial pharyngitis (strep throat); it is also an important cause of various skin infections that can be relatively mild (e.g., impetigo) or life threatening (e.g., necrotizing fasciitis, also known as flesh eating disease), life threatening.

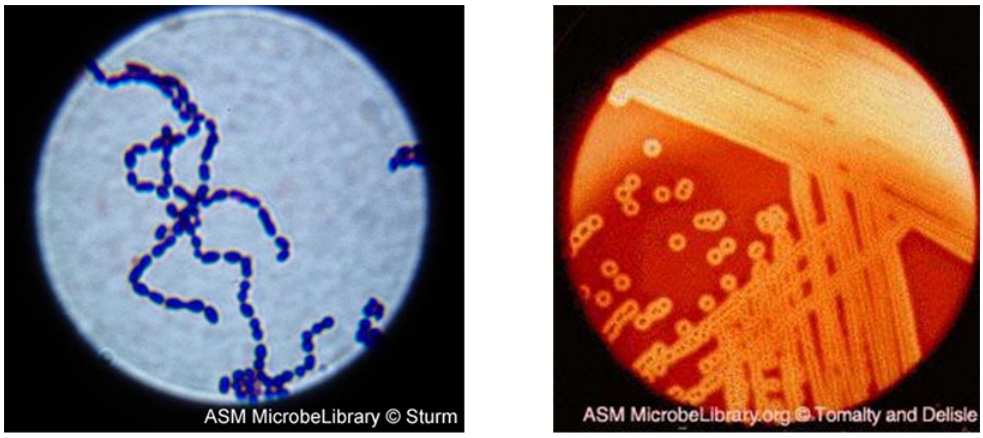


Figure **6.4.3**: (a) A gram-stained specimen of *Streptococcus pyogenes* shows the chains of cocci characteristic of this organism's morphology. (b) *S. pyogenes* on blood agar shows characteristic lysis of red blood cells, indicated by the halo of clearing around colonies. (credit a, b: modification of work by American Society for Microbiology)

The nonpyogenic (i.e., not associated with pus production) streptococci are a group of streptococcal species that are not a taxon but are grouped together because they inhabit the human mouth. The nonpyogenic streptococci do not belong to any of the Lancefield groups. Most are commensals, but a few, such as *S. mutans*, are implicated in the development of dental caries.

S. pneumoniae (commonly referred to as pneumococcus), is a *Streptococcus* species that also does not belong to any Lancefield group. *S. pneumoniae* cells appear microscopically as diplococci, pairs of cells, rather than the long chains typical of most streptococci. Scientists have known since the 19th century that *S. pneumoniae* causes pneumonia and other respiratory infections. However, this bacterium can also cause a wide range of other diseases, including meningitis, septicemia, osteomyelitis, and endocarditis, especially in newborns, the elderly, and patients with immunodeficiency.

Bacilli

The name of the class Bacilli suggests that it is made up of bacteria that are bacillus in shape, but it is a morphologically diverse class that includes bacillus-shaped and cocccus-shaped genera. Among the many genera in this class are two that are very important clinically: *Bacillus* and *Staphylococcus*.

Bacteria in the genus *Bacillus* are bacillus in shape and can produce endospores. They include aerobes or facultative anaerobes. A number of *Bacillus* spp. are used in various industries, including the production of antibiotics (e.g., barnase), enzymes (e.g., alpha-amylase, BamH1 restriction endonuclease), and detergents (e.g., subtilisin).

Two notable pathogens belong to the genus *Bacillus*. *B. anthracis* is the pathogen that causes anthrax, a severe disease that affects wild and domesticated animals and can spread from infected animals to humans. Anthrax manifests in humans as charcoal-black ulcers on the skin, severe enterocolitis, pneumonia, and brain damage due to swelling. If untreated, anthrax is lethal. *B. cereus*, a closely related species, is a pathogen that may cause food poisoning. It is a rod-shaped species that forms chains. Colonies appear milky white with irregular shapes when cultured on blood agar (Figure **6.4.4**). One other important species is *B. thuringiensis*. This bacterium produces a number of substances used as insecticides because they are toxic for insects.

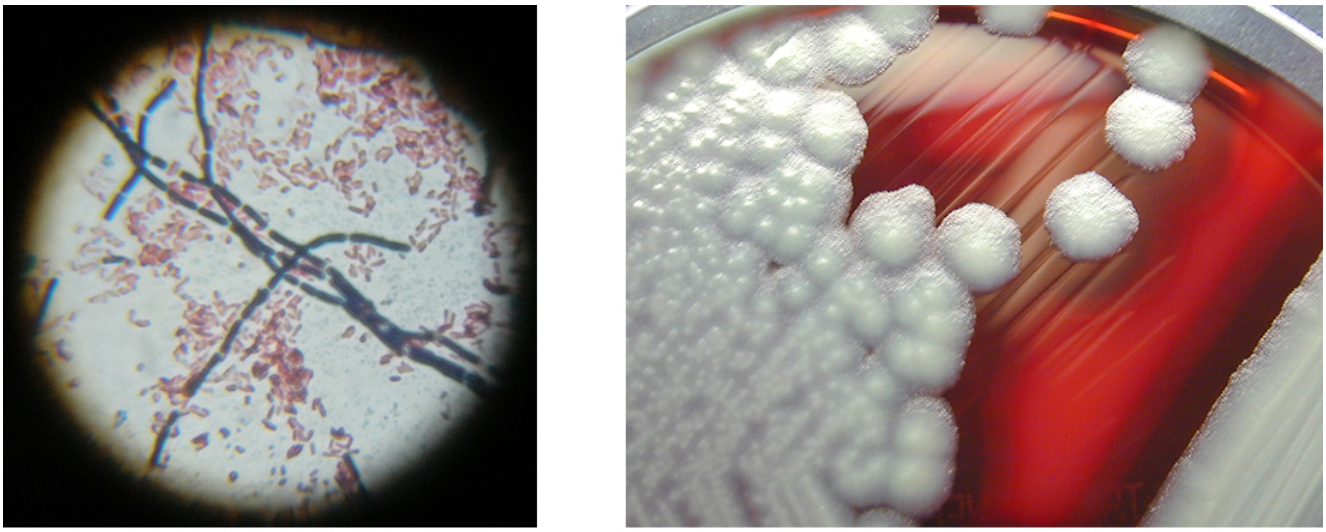


Figure 6.4.4: (a) In this gram-stained specimen, the violet rod-shaped cells forming chains are the gram-positive bacteria *Bacillus cereus*. The small, pink cells are the gram-negative bacteria *Escherichia coli*. (b) In this culture, white colonies of *B. cereus* have been grown on sheep blood agar. (credit a: modification of work by "Bibliomaniac 15"/Wikimedia Commons; credit b: modification of work by Centers for Disease Control and Prevention)

The genus *Staphylococcus* also belongs to the class Bacilli, even though its shape is coccus rather than a bacillus. The name *Staphylococcus* comes from a Greek word for *bunches of grapes*, which describes their microscopic appearance in culture (Figure 6.4.5). *Staphylococcus* spp. are facultative anaerobic, halophilic, and nonmotile. The two best-studied species of this genus are *S. epidermidis* and *S. aureus*.

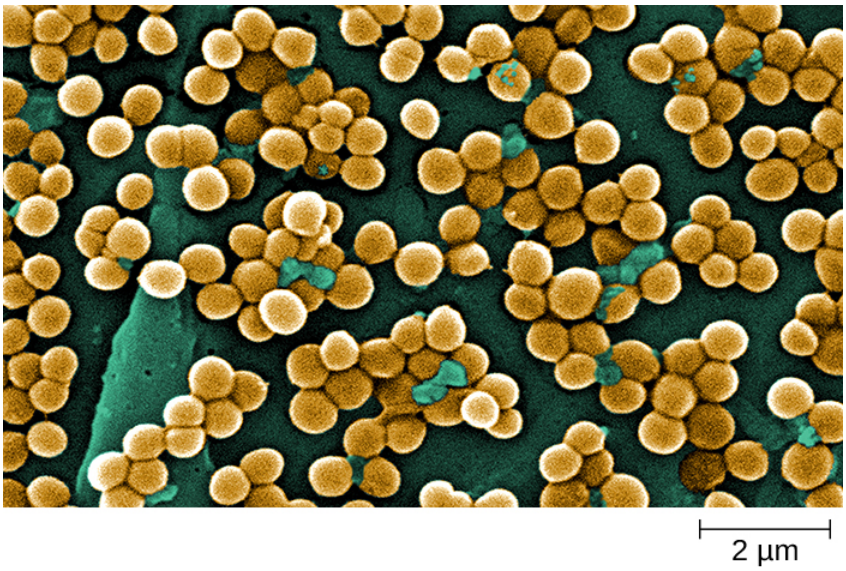


Figure 6.4.5: This SEM of *Staphylococcus aureus* illustrates the typical "grape-like" clustering of cells. (credit: modification of work by Centers for Disease Control and Prevention)

S. epidermidis, whose main habitat is the human skin, is thought to be nonpathogenic for humans with healthy immune systems, but in patients with immunodeficiency, it may cause infections in skin wounds and prostheses (e.g., artificial joints, heart valves). *S. epidermidis* is also an important cause of infections associated with intravenous catheters. This makes it a dangerous pathogen in hospital settings, where many patients may be immunocompromised.

Strains of *S. aureus* cause a wide variety of infections in humans, including skin infections that produce boils, carbuncles, cellulitis, or impetigo. Certain strains of *S. aureus* produce a substance called enterotoxin, which can cause severe enteritis, often called staph

food poisoning. Some strains of *S. aureus* produce the toxin responsible for toxic shock syndrome, which can result in cardiovascular collapse and death.

Many strains of *S. aureus* have developed resistance to antibiotics. Some antibiotic-resistant strains are designated as methicillin-resistant *S. aureus* (MRSA) and vancomycin-resistant *S. aureus* (VRSA). These strains are some of the most difficult to treat because they exhibit resistance to nearly all available antibiotics, not just methicillin and vancomycin. Because they are difficult to treat with antibiotics, infections can be lethal. MRSA and VRSA are also contagious, posing a serious threat in hospitals, nursing homes, dialysis facilities, and other places where there are large populations of elderly, bedridden, and/or immunocompromised patients.

Biopiracy and Bioprospecting

In 1969, an employee of a Swiss pharmaceutical company was vacationing in Norway and decided to collect some soil samples. He took them back to his lab, and the Swiss company subsequently used the fungus *Tolypocladium inflatum* in those samples to develop cyclosporine A, a drug widely used in patients who undergo tissue or organ transplantation. The Swiss company earns more than $1 billion a year for production of cyclosporine A, yet Norway receives nothing in return—no payment to the government or benefit for the Norwegian people. Despite the fact the cyclosporine A saves numerous lives, many consider the means by which the soil samples were obtained to be an act of "biopiracy," essentially a form of theft. Do the ends justify the means in a case like this?

Nature is full of as-yet-undiscovered bacteria and other microorganisms that could one day be used to develop new life-saving drugs or treatments.[1] Pharmaceutical and biotechnology companies stand to reap huge profits from such discoveries, but ethical questions remain. To whom do biological resources belong? Should companies who invest (and risk) millions of dollars in research and development be required to share revenue or royalties for the right to access biological resources?

Compensation is not the only issue when it comes to bioprospecting. Some communities and cultures are philosophically opposed to bioprospecting, fearing unforeseen consequences of collecting genetic or biological material. Native Hawaiians, for example, are very protective of their unique biological resources.

For many years, it was unclear what rights government agencies, private corporations, and citizens had when it came to collecting samples of microorganisms from public land. Then, in 1993, the Convention on Biological Diversity granted each nation the rights to any genetic and biological material found on their own land. Scientists can no longer collect samples without a prior arrangement with the land owner for compensation. This convention now ensures that companies act ethically in obtaining the samples they use to create their products.

Key Concepts and Summary

- Gram-positive bacteria are a very large and diverse group of microorganisms. Understanding their taxonomy and knowing their unique features is important for diagnostics and treatment of infectious diseases.
- Gram-positive bacteria are classified into **high G+C gram-positive** and **low G+C gram-positive** bacteria, based on the prevalence of guanine and cytosine nucleotides in their genome
- Actinobacteria is the taxonomic name of the class of high G+C gram-positive bacteria. This class includes the genera *Actinomyces*, *Arthrobacter*, *Corynebacterium*, *Frankia*, *Gardnerella*, *Micrococcus*, *Mycobacterium*, *Nocardia*, *Propionibacterium*, *Rhodococcus*, and *Streptomyces*. Some representatives of these genera are used in industry; others are human or animal pathogens.
- Examples of high G+C gram-positive bacteria that are human pathogens include *Mycobacterium tuberculosis*, which causes tuberculosis; *M. leprae*, which causes leprosy (Hansen's disease); and *Corynebacteriumdiphtheriae*, which causes diphtheria.
- *Clostridia* spp. are low G+C gram-positive bacteria that are generally obligate anaerobes and can form endospores. Pathogens in this genus include *C. perfringens* (gas gangrene), *C. tetani* (tetanus), and *C. botulinum* (botulism).
- Lactobacillales include the genera *Enterococcus*, *Lactobacillus*, *Leuconostoc*, and *Streptococcus*. *Streptococcus* is responsible for many human diseases, including pharyngitis (strep throat), scarlet fever, rheumatic fever, glomerulonephritis, pneumonia, and other respiratory infections.
- Bacilli is a taxonomic class of low G+C gram-positive bacteria that include rod-shaped and coccus-shaped species, including the genera *Bacillus* and *Staphylococcus*. *B. anthracis* causes anthrax, *B. cereus* may cause opportunistic infections of the

gastrointestinal tract, and *S. aureus* strains can cause a wide range of infections and diseases, many of which are highly resistant to antibiotics.

- *Mycoplasma* spp. are very small, **pleomorphic** low G+C gram-positive bacteria that lack cell walls. *M. pneumoniae* causes atypical pneumonia.

Footnotes

1. 1 J. Andre. *Bioethics as Practice*. Chapel Hill, NC: University of North Carolina Press, 2002.

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6.5: Deeply Branching Bacteria

6.05: Deeply Branching Bacteria

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6.6: Archaea

6.06: Archaea

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6.E: Prokaryotic Diversity (Exercises)

6.1: Prokaryote Habitats, Relationships, and Microbiomes

Prokaryotes are unicellular microorganisms whose cells have no nucleus. Prokaryotes can be found everywhere on our planet, even in the most extreme environments. Prokaryotes are very flexible metabolically, so they are able to adjust their feeding to the available natural resources. Prokaryotes live in communities that interact among themselves and with large organisms that they use as hosts (including humans).

Multiple Choice

The term prokaryotes refers to which of the following?

A. very small organisms
B. unicellular organisms that have no nucleus
C. multicellular organisms
D. cells that resemble animal cells more than plant cells

Answer

B

The term microbiota refers to which of the following?

A. all microorganisms of the same species
B. all of the microorganisms involved in a symbiotic relationship
C. all microorganisms in a certain region of the human body
D. all microorganisms in a certain geographic region

Answer

C

Which of the following refers to the type of interaction between two prokaryotic populations in which one population benefits and the other is not affected?

A. mutualism
B. commensalism
C. parasitism
D. neutralism

Answer

B

True/False

Among prokaryotes, there are some that can live in every environment on earth.

Answer

True

Fill in the Blank

When prokaryotes live as interacting communities in which one population benefits to the harm of the other, the type of symbiosis is called _____.

Answer

parasitism

The domain _____ does not include prokaryotes.

Answer

Eukarya

Pathogenic bacteria that are part of the transient microbiota can sometimes be eliminated by _____ therapy.

Answer

antibiotic

Nitrogen-fixing bacteria provide other organisms with usable nitrogen in the form of _____.

Answer

ammonia

Short Answer

Compare commensalism and amensalism.

Give an example of the changes of human microbiota that result from medical intervention.

6.2: Proteobacteria

Proteobacteria is a phylum of gram-negative bacteria and are classified into the classes alpha-, beta-, gamma-, delta- and epsilonproteobacteria, each class having separate orders, families, genera, and species. Alphaproteobacteria are oligotrophs. The taxa chlamydias and rickettsias are obligate intracellular pathogens, feeding on cells of host organisms; they are metabolically inactive outside of the host cell. Some Alphaproteobacteria can convert atmospheric nitrogen to nitrites.

Multiple Choice

Which of the following describes Proteobacteria in domain Bacteria?

A. phylum
B. class
C. species
D. genus

Answer

A

All Alphaproteobacteria are which of the following?

A. oligotrophs
B. intracellular
C. pathogenic
D. all of the above
E. none of the above

Answer

A

Class Betaproteobacteria includes all but which of the following genera?

A. *Neisseria*.
B. *Bordetella*.
C. *Leptothrix*.
D. *Campylobacter*.

Answer

D

Haemophilus influenzae is a common cause of which of the following?

A. influenza
B. dysentery
C. upper respiratory tract infections
D. hemophilia

Answer

C

Fill in the Blank

Rickettsias are _____ intracellular bacteria.

Answer

obligate

The species _____, which belongs to Epsilonproteobacteria, causes peptic ulcers of the stomach and duodenum.

Answer

Helicobacter pylori

The genus *Salmonella* belongs to the class _____ and includes pathogens that cause salmonellosis and typhoid fever.

Answer

Gammaproteobacteria

Short Answer

What is the metabolic difference between coliforms and noncoliforms? Which category contains several species of intestinal pathogens?

Why are *Mycoplasma* and *Chlamydia* classified as obligate intracellular pathogens?

Critical Thinking

The cell shown is found in the human stomach and is now known to cause peptic ulcers. What is the name of this bacterium?



(credit: American Society for Microbiology)

6.3: Nonproteobacteria Gram-negative Bacteria and Phototrophic Bacteria

Gram-negative nonproteobacteria include the taxa spirochetes; the Cytophaga, Fusobacterium, Bacteroides group; Planctomycetes; and many representatives of phototrophic bacteria. Spirochetes are motile, spiral bacteria with a long, narrow body; they are difficult or impossible to culture. Several genera of spirochetes contain human pathogens that cause such diseases as syphilis and Lyme disease. Cytophaga, Fusobacterium, and Bacteroides are classified together as a phylum called the CFB group.

Multiple Choice

Which of the following is the organelle that spirochetes use to propel themselves?

A. plasma membrane
B. axial filament
C. pilum
D. fimbria

Answer

B

Which of the following bacteria are the most prevalent in the human gut?

A. cyanobacteria
B. staphylococci
C. *Borrelia*
D. *Bacteroides*

Answer

D

Which of the following refers to photosynthesis performed by bacteria with the use of water as the donor of electrons?

A. oxygenic
B. anoxygenic
C. heterotrophic
D. phototrophic

Answer

A

Fill in the Blank

The bacterium that causes syphilis is called _____.

Answer

Treponema pallidum pallidum

Bacteria in the genus *Rhodospirillum* that use hydrogen for oxidation and fix nitrogen are _____ bacteria.

Answer

purple nonsulfur

Short Answer

Explain the term CFB group and name the genera that this group includes.

Name and briefly describe the bacterium that causes Lyme disease.

Characterize the phylum Cyanobacteria.

6.4: Gram-positive Bacteria

Gram-positive bacteria are a very large and diverse group of microorganisms. Understanding their taxonomy and knowing their unique features is important for diagnostics and treatment of infectious diseases. Gram-positive bacteria are classified into high G+C gram-positive and low G+C gram-positive bacteria, based on the prevalence of guanine and cytosine nucleotides in their genome.

Multiple Choice

Which of the following bacterial species is classified as high G+C gram-positive?

A. *Corynebacterium diphtheriae*
B. *Staphylococcus aureus*
C. *Bacillus anthracis*
D. *Streptococcus pneumonia*

Answer

A

Fill in the Blank

Streptococcus is the _____ of bacteria that is responsible for many human diseases.

Answer

genus

One species of *Streptococcus*, *S. pyogenes*, is a classified as a _____ pathogen due to the characteristic production of pus in infections it causes.

Answer

pyogenic

Propionibacterium belongs to _____ G+C gram-positive bacteria. One of its species is used in the food industry and another causes acne.

Answer

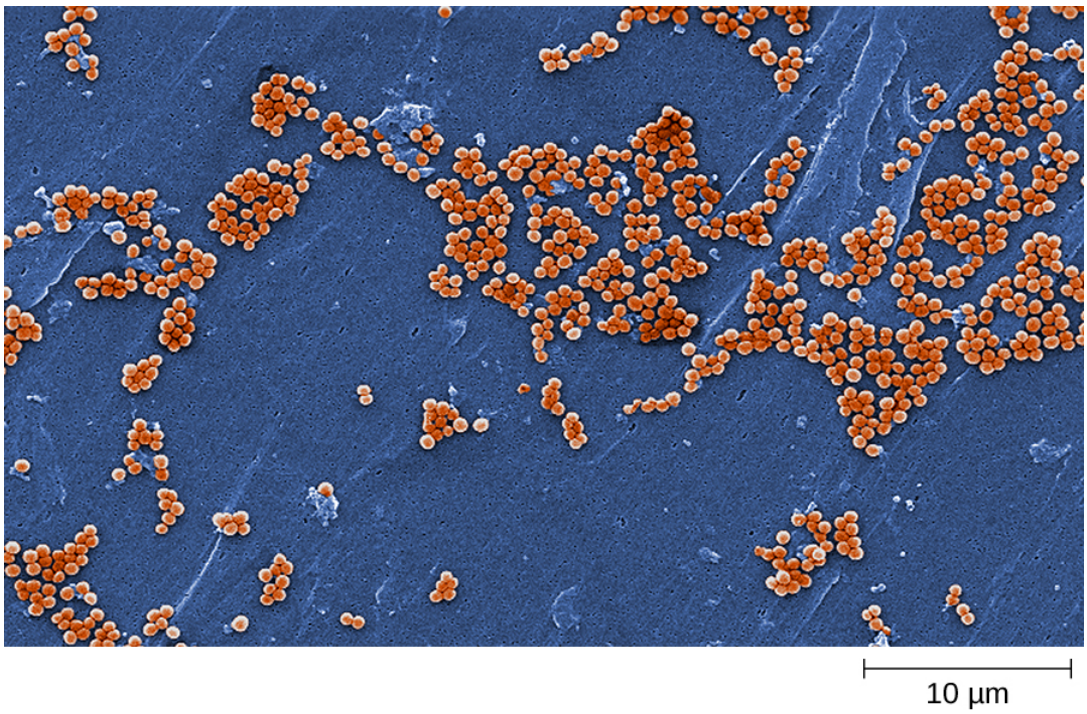
high

Short Answer

Name and describe two types of *S. aureus* that show multiple antibiotic resistance.

Critical Thinking

The microscopic growth pattern shown is characteristic of which genus of bacteria?



(credit: modification of work by Janice Haney Carr/Centers for Disease Control and Prevention)

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CHAPTER OVERVIEW

7: Viruses

Public health measures in the developed world have dramatically reduced mortality from viral epidemics. But when epidemics do occur, they can spread quickly with global air travel. In 2009, an outbreak of H1N1 influenza spread across various continents. In early 2014, cases of Ebola in Guinea led to a massive epidemic in western Africa. This included the case of an infected man who traveled to the United States, sparking fears the epidemic might spread beyond Africa.

Until the late 1930s and the advent of the electron microscope, no one had seen a virus. Yet treatments for preventing or curing viral infections were used and developed long before that. Historical records suggest that by the 17th century, and perhaps earlier, inoculation (also known as variolation) was being used to prevent the viral disease smallpox in various parts of the world. By the late 18th century, Englishman Edward Jenner was inoculating patients with cowpox to prevent smallpox, a technique he coined *vaccination*.[1]

Today, the structure and genetics of viruses are well defined, yet new discoveries continue to reveal their complexities. In this chapter, we will learn about the structure, classification, and cultivation of viruses, and how they impact their hosts. In addition, we will learn about other infective particles such as viroids and prions.

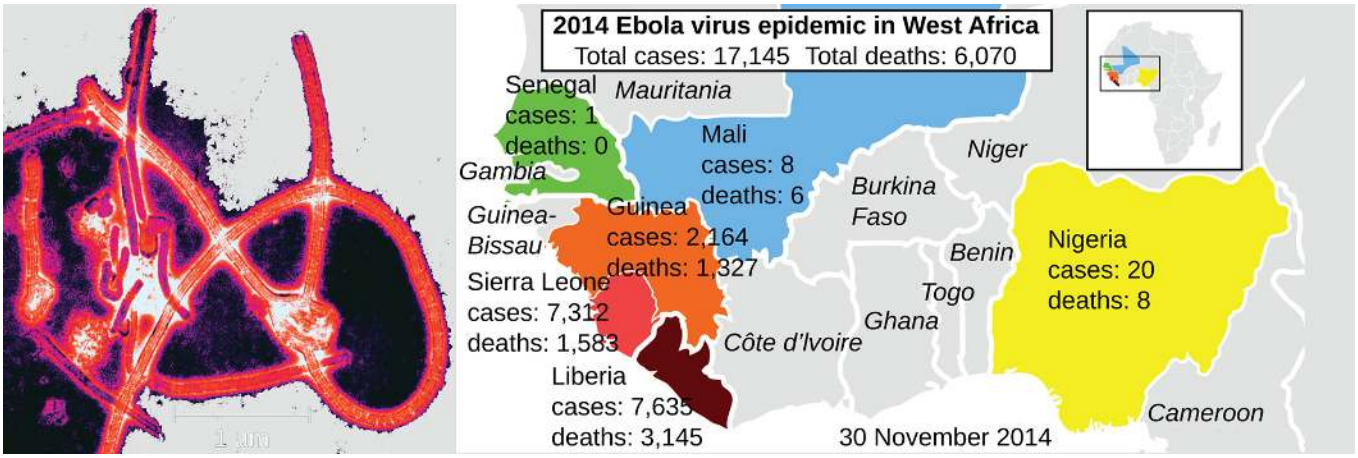


Figure 7.1: The year 2014 saw the first large-scale outbreak of Ebola virus (electron micrograph, left) in human populations in West Africa (right). Such epidemics are now widely reported and documented, but viral epidemics are sure to have plagued human populations since the origin of our species. (credit left: modification of work by Thomas W. Geisbert)

7.1: Viruses
7.2: The Viral Life Cycle
7.3: Viruses
7.3.1: General Characteristics of Viruses
7.3.2: Size and Shapes of Viruses
7.3.3: Viral Structure
7.3.4: Classification of Viruses
7.3.5: Other Acellular Infectious Agents- Viroids and Prions
7.3.6: Animal Virus Life Cycles
7.3.6A: The Productive Life Cycle of Animal Viruses
7.3.6B: Productive Life Cycle with Possible Latency
7.3.6C: The Life Cycle of HIV
7.3.6D: Natural History of a Typical HIV Infection
7.3.6E: The Role of Viruses in Tumor Production

7.3.7: Bacteriophage Life Cycles- An Overview
7.3.7A: The Lytic Life Cycle of Bacteriophages
7.3.7B: The Lysogenic Life Cycle of Bacteriophages
7.3.8: Pathogenicity of Animal Viruses
7.3.9: Bacteriophage-Induced Alterations of Bacteria
7.3.10: Antiviral Agents
7.3.11: General Categories of Viral Infections
7.3.E: Viruses (Exercises)
7.E: Acellular Pathogens (Exercises)

Footnotes

1. 1 S. Riedel “Edward Jenner and the History of Smallpox and Vaccination.” *Baylor University Medical Center Proceedings* 18, no. 1 (January 2005): 21–25.

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7.1: Viruses

Learning Objectives

- Describe the general characteristics of viruses as pathogens
- Identify the range of organisms that viruses can infect
- Explain the concept vectors
- Distinguish between mechanical and biological vectors

Despite their small size, which prevented them from being seen with light microscopes, the discovery of a filterable component smaller than a bacterium that causes tobacco mosaic disease (TMD) dates back to 1892.[1] At that time, Dmitri Ivanovski, a Russian botanist, discovered the source of TMD by using a porcelain filtering device first invented by Charles Chamberland and Louis Pasteur in Paris in 1884. Porcelain Chamberland filters have a pore size of 0.1 µm, which is small enough to remove all bacteria ≥0.2 µm from any liquids passed through the device. An extract obtained from TMD-infected tobacco plants was made to determine the cause of the disease. Initially, the source of the disease was thought to be bacterial. It was surprising to everyone when Ivanovski, using a Chamberland filter, found that the cause of TMD was not removed after passing the extract through the porcelain filter. So if a bacterium was not the cause of TMD, what could be causing the disease? Ivanovski concluded the cause of TMD must be an extremely small bacterium or bacterial spore. Other scientists, including Martinus Beijerinck, continued investigating the cause of TMD. It was Beijerinck, in 1899, who eventually concluded the causative agent was not a bacterium but, instead, possibly a chemical, like a biological poison we would describe today as a toxin. As a result, the word *virus*, Latin for poison, was used to describe the cause of TMD a few years after Ivanovski's initial discovery. Even though he was not able to see the virus that caused TMD, and did not realize the cause was not a bacterium, Ivanovski is credited as the original discoverer of viruses and a founder of the field of virology.

Today, we can see viruses using electron microscopes (Figure **7.1.1**) and we know much more about them. Viruses are distinct biological entities; however, their evolutionary origin is still a matter of speculation. In terms of taxonomy, they are not included in the tree of life because they are **acellular** (not consisting of cells). In order to survive and reproduce, viruses must infect a cellular host, making them obligate intracellular parasites. The genome of a virus enters a host cell and directs the production of the viral components, proteins and nucleic acids, needed to form new virus particles called **virions**. New virions are made in the host cell by assembly of viral components. The new virions transport the viral genome to another host cell to carry out another round of infection. Table **7.1.1** summarizes the properties of viruses.

Table **7.1.1**: Properties of viruses.

Characteristics of Viruses
Infectious, acellular pathogens
Obligate intracellular parasites with host and cell-type specificity
DNA or RNA genome (never both)
Genome is surrounded by a protein capsid and, in some cases, a phospholipid membrane studded with viral glycoproteins
Lack genes for many products needed for successful reproduction, requiring exploitation of host-cell genomes to reproduce

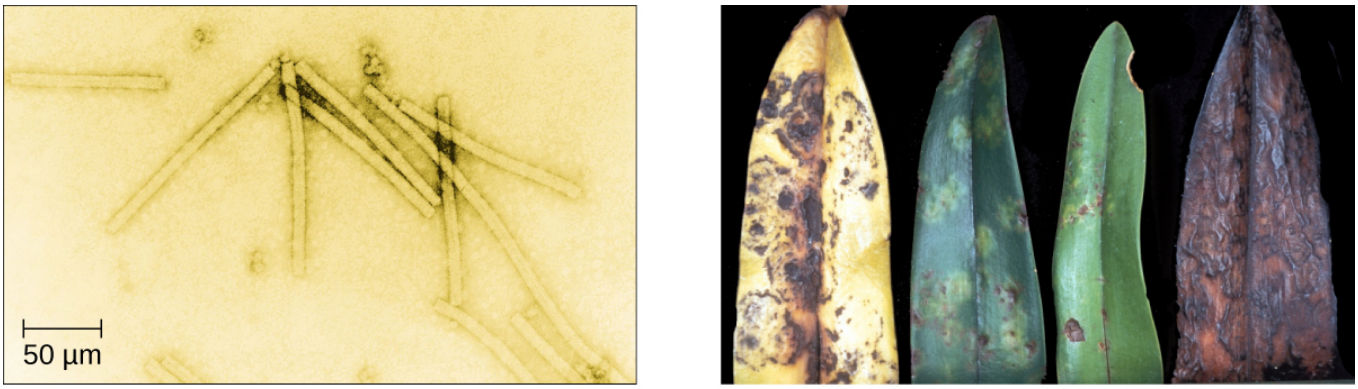
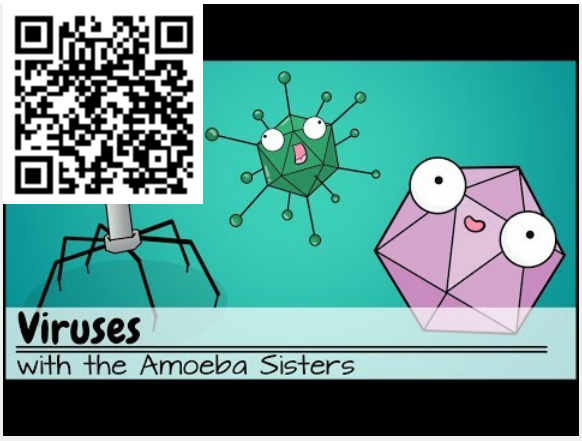


Figure **7.1.1**: (a) Tobacco mosaic virus (TMV) viewed with transmission electron microscope. (b) Plants infected with tobacco mosaic disease (TMD), caused by TMV. (credit a: modification of work by USDA Agricultural Research Service—scale-bar data from Matt Russell; credit b: modification of work by USDA Forest Service, Department of Plant Pathology Archive North Carolina State University)

Video: Viruses

Explore the lytic and lysogenic viral replication cycles with the Amoeba Sisters! This video also discusses virus structures and why a host is critical for viral reproduction.





Hosts and Viral Transmission

Viruses can infect every type of host cell, including those of plants, animals, fungi, protists, bacteria, and archaea. Most viruses will only be able to infect the cells of one or a few species of organism. This is called the **host range**. However, having a wide host range is not common and viruses will typically only infect specific hosts and only specific cell types within those hosts. The viruses that infect bacteria are called **bacteriophages**, or simply phages. The word *phage* comes from the Greek word for devour. Other viruses are just identified by their host group, such as animal or plant viruses. Once a cell is infected, the effects of the virus can vary depending on the type of virus. Viruses may cause abnormal growth of the cell or cell death, alter the cell's genome, or cause little noticeable effect in the cell.

Viruses can be transmitted through direct contact, indirect contact with fomites, or through a **vector**: an animal that transmits a pathogen from one host to another. Arthropods such as mosquitoes, ticks, and flies, are typical vectors for viral diseases, and they may act as **mechanical vectors** or **biological vectors**. Mechanical transmission occurs when the arthropod carries a viral pathogen on the outside of its body and transmits it to a new host by physical contact. Biological transmission occurs when the arthropod carries the viral pathogen inside its body and transmits it to the new host through biting.

In humans, a wide variety of viruses are capable of causing various infections and diseases. Some of the deadliest emerging pathogens in humans are viruses, yet we have few treatments or drugs to deal with viral infections, making them difficult to eradicate.

Viruses that can be transmitted from an animal host to a human host can cause zoonoses. For example, the avian influenza virus originates in birds, but can cause disease in humans. Reverse zoonoses are caused by infection of an animal by a virus that originated in a human.

Fighting Bacteria With Viruses

The emergence of superbugs, or multidrug resistant bacteria, has become a major challenge for pharmaceutical companies and a serious health-care problem. According to a 2013 report by the US Centers for Disease Control and Prevention (CDC), more than 2 million people are infected with drug-resistant bacteria in the US annually, resulting in at least 23,000 deaths.[2] The continued use and overuse of antibiotics will likely lead to the evolution of even more drug-resistant strains.

One potential solution is the use of phage therapy, a procedure that uses bacteria-killing viruses (bacteriophages) to treat bacterial infections. Phage therapy is not a new idea. The discovery of bacteriophages dates back to the early 20th century, and phage therapy was first used in Europe in 1915 by the English bacteriologist Frederick Twort.[3] However, the subsequent discovery of penicillin and other antibiotics led to the near abandonment of this form of therapy, except in the former Soviet Union and a few countries in Eastern Europe. Interest in phage therapy outside of the countries of the former Soviet Union is only recently re-emerging because of the rise in antibiotic-resistant bacteria.[4]

Phage therapy has some advantages over antibiotics in that phages kill only one specific bacterium, whereas antibiotics kill not only the pathogen but also beneficial bacteria of the normal microbiota. Development of new antibiotics is also expensive for drug companies and for patients, especially for those who live in countries with high poverty rates.

Phages have also been used to prevent food spoilage. In 2006, the US Food and Drug Administration approved the use of a solution containing six bacteriophages that can be sprayed on lunch meats such as bologna, ham, and turkey to kill *Listeria monocytogenes*, a bacterium responsible for listeriosis, a form of food poisoning. Some consumers have concerns about the use of phages on foods, however, especially given the rising popularity of organic products. Foods that have been treated with phages must declare "bacteriophage preparation" in the list of ingredients or include a label declaring that the meat has been "treated with antimicrobial solution to reduce microorganisms."[5]

Key Concepts and Summary

- Viruses are generally ultramicroscopic, typically from 20 nm to 900 nm in length. Some large viruses have been found.
- Viruses are obligate intracellular parasites.
- Viruses are known to infect various types of cells found in plants, animals, fungi, protists, bacteria, and archaea. Viruses typically have limited **host ranges** and infect specific cell types.

Footnotes

1. 1 H. Lecoq. "[Discovery of the First Virus, the Tobacco Mosaic Virus: 1892 or 1898?]." *Comptes Rendus de l'Academie des Sciences – Serie III – Sciences de la Vie* 324, no. 10 (2001): 929–933.
2. 2 US Department of Health and Human Services, Centers for Disease Control and Prevention. "Antibiotic Resistance Threats in the United States, 2013." www.cdc.gov/drugresistance/pd...s-2013-508.pdf (accessed September 22, 2015).
3. 3 M. Clokie et al. "Phages in Nature." *Bacteriophage* 1, no. 1 (2011): 31–45.
4. 4 A. Sulakvelidze et al. "Bacteriophage Therapy." *Antimicrobial Agents and Chemotherapy* 45, no. 3 (2001): 649–659.
5. 5 US Food and Drug Administration. "FDA Approval of *Listeria*-specific Bacteriophage Preparation on Ready-to-Eat (RTE) Meat and Poultry Products." www.fda.gov/food/ingredientsp.../ucm083572.htm (accessed September 22, 2015).
6. 6 N. *Philippe et al. "Pandoraviruses: Amoeba Viruses with Genomes up to 2.5 Mb Reaching that of Parasitic Eukaryotes." Science 341, no. 6143 (2013): 281–286.*
7. 7 J. Cohen. "What's Old Is New: 1918 Virus Matches 2009 H1N1 Strain. *Science* 327, no. 5973 (2010): 1563–1564.

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7.2: The Viral Life Cycle

Learning Objectives

- Describe the major steps of the bacteriophage lytic cycle: attachment, penetration, biosynthesis, maturation and release
- Explain the lysogenic cycle including the role of prophage integration and environmental triggers for induction
- Compare and contrast generalized and specialized transduction

All viruses depend on cells for reproduction and metabolic processes. By themselves, viruses do not encode for all of the enzymes necessary for viral replication. But within a host cell, a virus can commandeer cellular machinery to produce more viral particles. Bacteriophages replicate only in the cytoplasm, since prokaryotic cells do not have a nucleus or organelles. In eukaryotic cells, most DNA viruses can replicate inside the nucleus, with an exception observed in the large DNA viruses, such as the poxviruses, that can replicate in the cytoplasm. RNA viruses that infect animal cells often replicate in the cytoplasm.

The Life Cycle of Viruses with Prokaryote Hosts

The life cycle of bacteriophages has been a good model for understanding how viruses affect the cells they infect, since similar processes have been observed for eukaryotic viruses, which can cause immediate death of the cell or establish a latent or chronic infection. Virulent phages typically lead to the death of the cell through cell lysis. **Temperate phages**, on the other hand, can become part of a host chromosome and are replicated with the cell genome until such time as they are induced to make newly assembled viruses, or **progeny viruses**.

Video: Lytic v. Lysogenic Cycles of Bacteriophages

McGraw Hill Video.





The Lytic Cycle

During the **lytic cycle** of virulent phage, the bacteriophage takes over the cell, reproduces new phages, and destroys the cell. **T-even phage** is a good example of a well-characterized class of virulent phages. There are five stages in the bacteriophage lytic cycle (see Figure **7.2.1**). **Attachment** is the first stage in the infection process in which the phage interacts with specific bacterial surface receptors (e.g., lipopolysaccharides and OmpC protein on host surfaces). Most phages have a narrow host range and may infect one species of bacteria or one strain within a species. This unique recognition can be exploited for targeted treatment of bacterial infection by phage therapy or for phage typing to identify unique bacterial subspecies or strains. The second stage of infection is **entry** or **penetration**. This occurs through contraction of the tail sheath, which acts like a hypodermic needle to inject the viral genome through the cell wall and membrane. The phage head and remaining components remain outside the bacteria.

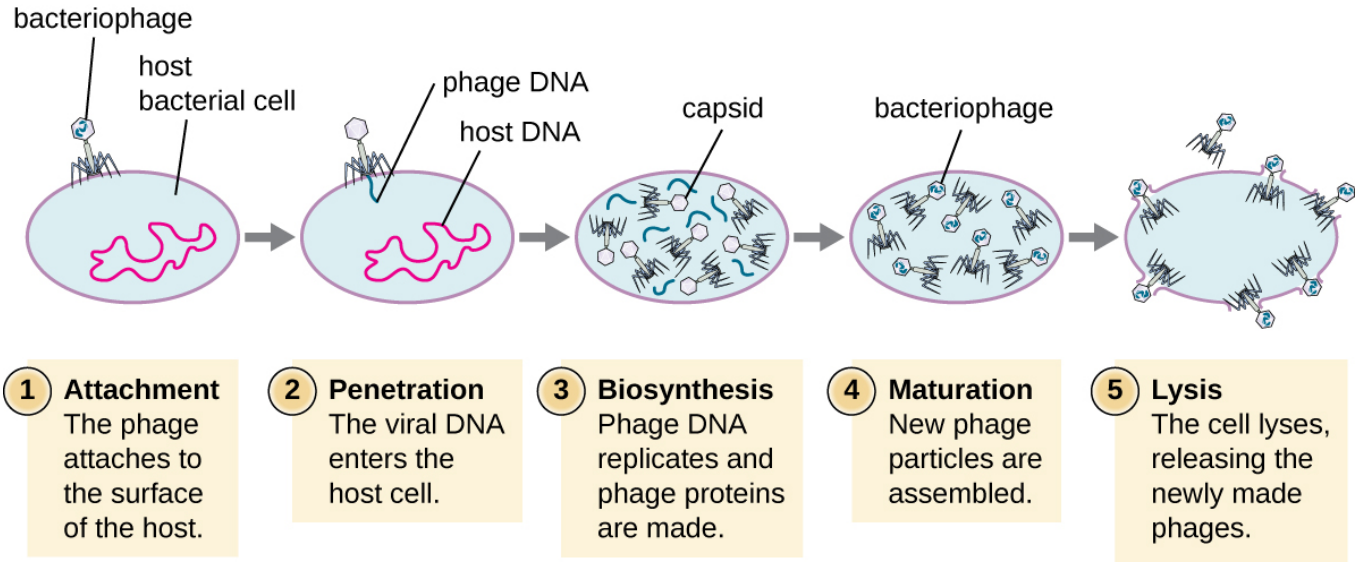


Figure **7.2.1**: A virulent phage shows only the lytic cycle pictured here. In the lytic cycle, the phage replicates and lyses the host cell.

The third stage of infection is **biosynthesis** of new viral components. After entering the host cell, the virus synthesizes virus-encoded endonucleases to degrade the bacterial chromosome. It then hijacks the host cell to replicate, transcribe, and translate the necessary viral components (capsomeres, sheath, base plates, tail fibers, and viral enzymes) for the assembly of new viruses. Polymerase genes are usually expressed early in the cycle, while capsid and tail proteins are expressed later. During the **maturation** phase, new virions are created. To liberate free phages, the bacterial cell wall is disrupted by phage proteins such as

holin or lysozyme. The final stage is release. Mature viruses burst out of the host cell in a process called **lysis** and the progeny viruses are liberated into the environment to infect new cells.

The Lysogenic Cycle

In a **lysogenic cycle**, the phage genome also enters the cell through attachment and penetration. A prime example of a phage with this type of life cycle is the lambda phage. During the lysogenic cycle, instead of killing the host, the phage genome integrates into the bacterial chromosome and becomes part of the host. The integrated phage genome is called a **prophage**. A bacterial host with a prophage is called a **lysogen**. The process in which a bacterium is infected by a temperate phage is called **lysogeny**. It is typical of temperate phages to be latent or inactive within the cell. As the bacterium replicates its chromosome, it also replicates the phage's DNA and passes it on to new daughter cells during reproduction. The presence of the phage may alter the phenotype of the bacterium, since it can bring in extra genes (e.g., toxin genes that can increase bacterial virulence). This change in the host phenotype is called **lysogenic conversion** or **phage conversion**. Some bacteria, such as *Vibrio cholerae* and *Clostridium botulinum*, are less virulent in the absence of the prophage. The phages infecting these bacteria carry the toxin genes in their genome and enhance the virulence of the host when the toxin genes are expressed. In the case of *V. cholera*, phage encoded toxin can cause severe diarrhea; in *C. botulinum*, the toxin can cause paralysis. During lysogeny, the prophage will persist in the host chromosome until **induction**, which results in the excision of the viral genome from the host chromosome. After induction has occurred the temperate phage can proceed through a lytic cycle and then undergo lysogeny in a newly infected cell (see Figure 7.2.2).

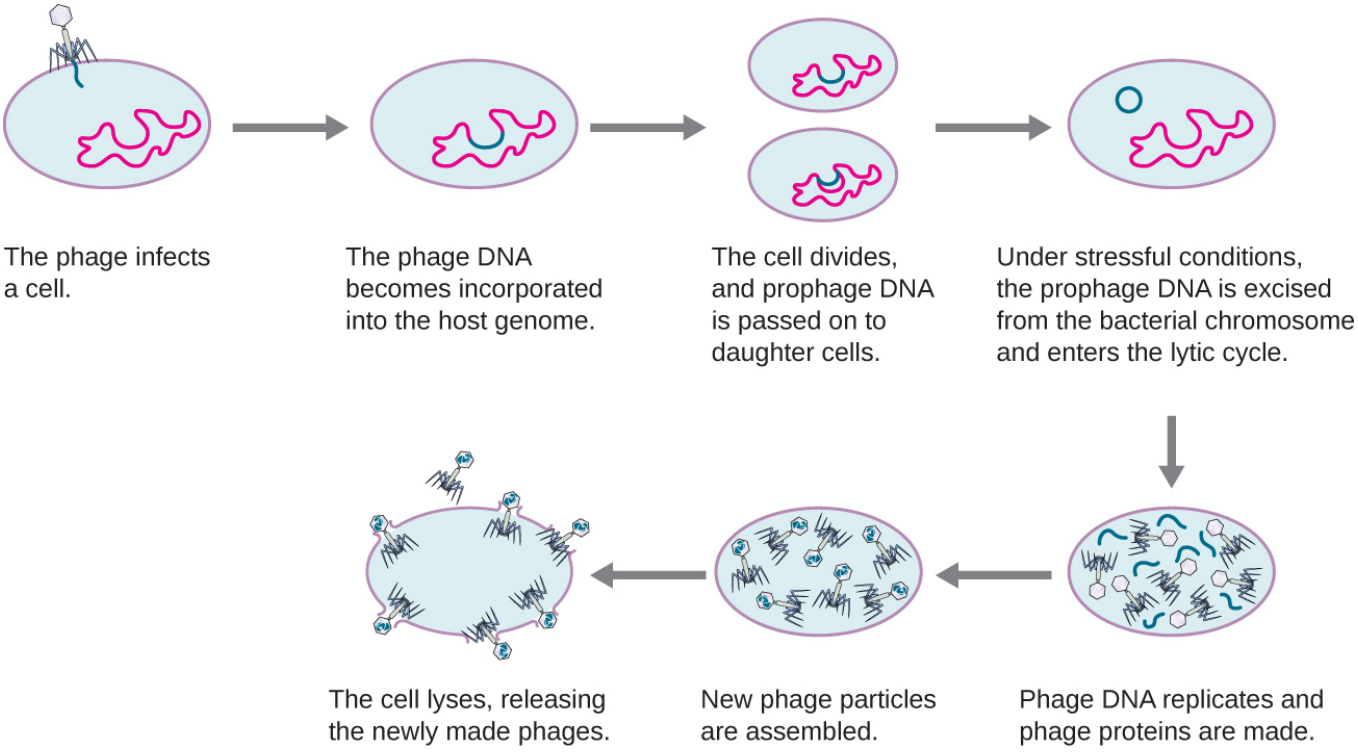


Figure 7.2.2: A temperate bacteriophage has both lytic and lysogenic cycles. In the lysogenic cycle, phage DNA is incorporated into the host genome, forming a prophage, which is passed on to subsequent generations of cells. Environmental stressors such as starvation or exposure to toxic chemicals may cause the prophage to be excised and enter the lytic cycle.

Link to Learning

This video illustrates the stages of the lysogenic life cycle of a bacteriophage and the transition to a lytic phase.

Transduction

Transduction occurs when a bacteriophage transfers bacterial DNA from one bacterium to another during sequential infections. There are two types of transduction: generalized and specialized transduction. During the lytic cycle of viral replication, the virus hijacks the host cell, degrades the host chromosome, and makes more viral genomes. As it assembles and packages DNA into the phage head, packaging occasionally makes a mistake. Instead of packaging viral DNA, it takes a random piece of host DNA and inserts it into the capsid. Once released, this virion will then inject the former host's DNA into a newly infected host. The asexual transfer of genetic information can allow for DNA recombination to occur, thus providing the new host with new genes (e.g., an antibiotic-resistance gene, or a sugar-metabolizing gene). **Generalized transduction** occurs when a random piece of bacterial chromosomal DNA is transferred by the phage during the lytic cycle. **Specialized transduction** occurs at the end of the lysogenic cycle, when the prophage is excised and the bacteriophage enters the lytic cycle. Since the phage is integrated into the host genome, the prophage can replicate as part of the host. However, some conditions (e.g., ultraviolet light exposure or chemical exposure) stimulate the prophage to undergo induction, causing the phage to excise from the genome, enter the lytic cycle, and produce new phages to leave host cells. During the process of excision from the host chromosome, a phage may occasionally remove some bacterial DNA near the site of viral integration. The phage and host DNA from one end or both ends of the integration site are packaged within the capsid and are transferred to the new, infected host. Since the DNA transferred by the phage is not randomly packaged but is instead a specific piece of DNA near the site of integration, this mechanism of gene transfer is referred to as specialized transduction (see Figure **7.2.3**). The DNA can then recombine with host chromosome, giving the latter new characteristics. Transduction seems to play an important role in the evolutionary process of bacteria, giving them a mechanism for asexual exchange of genetic information.

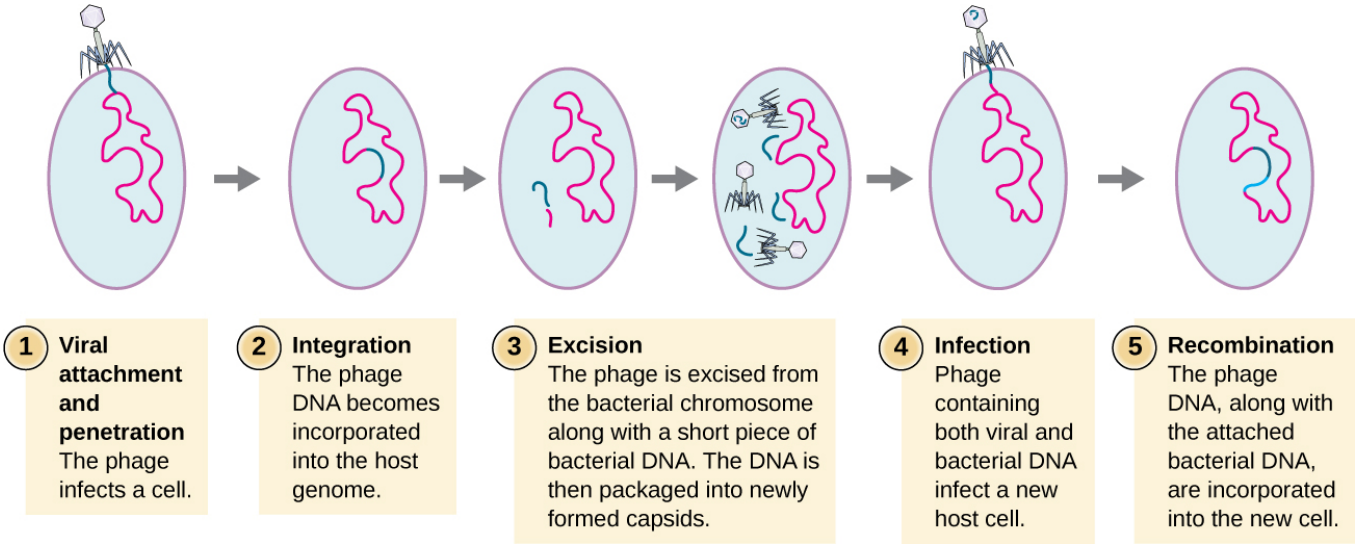


Figure **7.2.3**: This flowchart illustrates the mechanism of specialized transduction. An integrated phage excises, bringing with it a piece of the DNA adjacent to its insertion point. On reinfection of a new bacterium, the phage DNA integrates along with the genetic material acquired from the previous host.

Key Concepts and Summary

- Many viruses target specific hosts or tissues. Some may have more than one host.
- Many viruses follow several stages to infect host cells. These stages include **attachment, penetration, uncoating, biosynthesis, maturation,** and **release**.
- Bacteriophages have a **lytic** or **lysogenic cycle**. The lytic cycle leads to the death of the host, whereas the lysogenic cycle leads to integration of phage into the host genome.
- Bacteriophages inject DNA into the host cell, whereas animal viruses enter by endocytosis or membrane fusion.
- Bacteriophages transfer genetic information between hosts using either **generalized** or **specialized transduction**.

Footnotes

1. 1 World Health Organization. “WHO Ebola Data and Statistics.” March 18, 2005. http://apps.who.int/gho/data/view.eb...150318?lang=en

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7.3: Viruses

7.04: Viruses

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7.3.1: General Characteristics of Viruses

7.4.01: General Characteristics of Viruses

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7.3.2: Size and Shapes of Viruses

7.4.02: Size and Shapes of Viruses

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7.3.3: Viral Structure

7.4.03: Viral Structure

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7.3.4: Classification of Viruses

7.4.04: Classification of Viruses

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7.3.5: Other Acellular Infectious Agents- Viroids and Prions

7.4.05: Other Acellular Infectious Agents- Viroids and Prions

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7.3.6: Animal Virus Life Cycles

7.4.06: Animal Virus Life Cycles

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7.3.6A: The Productive Life Cycle of Animal Viruses

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7.3.6B: Productive Life Cycle with Possible Latency

7.4.6B: Productive Life Cycle with Possible Latency

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7.3.6C: The Life Cycle of HIV

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7.3.6D: Natural History of a Typical HIV Infection

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7.3.6E: The Role of Viruses in Tumor Production

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7.3.7: Bacteriophage Life Cycles- An Overview

7.4.07: Bacteriophage Life Cycles- An Overview

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7.3.7A: The Lytic Life Cycle of Bacteriophages

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7.3.7B: The Lysogenic Life Cycle of Bacteriophages

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7.3.8: Pathogenicity of Animal Viruses

7.4.08: Pathogenicity of Animal Viruses

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7.3.9: Bacteriophage-Induced Alterations of Bacteria

7.4.09: Bacteriophage-Induced Alterations of Bacteria

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7.3.10: Antiviral Agents

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7.3.11: General Categories of Viral Infections

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7.3.E: Viruses (Exercises)

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7.E: Acellular Pathogens (Exercises)

7.1: Viruses

Viruses are generally ultramicroscopic, typically from 20 nm to 900 nm in length. Some large viruses have been found. Virions are acellular and consist of a nucleic acid, DNA or RNA, but not both, surrounded by a protein capsid. There may also be a phospholipid membrane surrounding the capsid. Viruses are obligate intracellular parasites.

Multiple Choice

The component(s) of a virus that is/are extended from the envelope for attachment is/are the:

A. capsomeres
B. spikes
C. nucleic acid
D. viral whiskers

Answer

B

Which of the following does a virus lack? Select all that apply.

A. ribosomes
B. metabolic processes
C. nucleic acid
D. glycoprotein

Answer

A and B

The envelope of a virus is derived from the host's

A. nucleic acids
B. membrane structures
C. cytoplasm
D. genome

Answer

B

In naming viruses, the family name ends with _____ and genus name ends with _____.

A. *−virus; −viridae*
B. *−viridae; −virus*
C. *−virion; virus*
D. *−virus; virion*

Answer

B

What is another name for a nonenveloped virus?

A. enveloped virus
B. provirus
C. naked virus
D. latent virus

Answer

C

True/False

True or False: Scientists have identified viruses that are able to infect fungal cells.

Answer

True

Fill in the Blank

A virus that infects a bacterium is called a/an _____.

Answer

bacteriophage

A/an _____ virus possesses characteristics of both a polyhedral and helical virus.

Answer

complex

A virus containing only nucleic acid and a capsid is called a/an _____ virus or _____ virus.

Answer

naked or nonenveloped

The _____ _____ on the bacteriophage allow for binding to the bacterial cell.

Answer

tail fibers

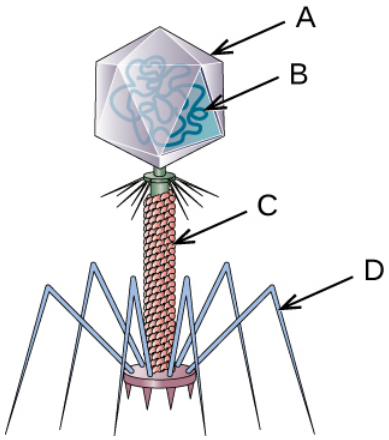
Short Answer

Discuss the geometric differences among helical, polyhedral, and complex viruses.

What was the meaning of the word “virus” in the 1880s and why was it used to describe the cause of tobacco mosaic disease?

Critical Thinking

Name each labeled part of the illustrated bacteriophage.



In terms of evolution, which do you think arises first? The virus or the host? Explain your answer.

Do you think it is possible to create a virus in the lab? Imagine that you are a mad scientist. Describe how you would go about creating a new virus.

7.2: The Viral Life Cycle

Many viruses target specific hosts or tissues. Some may have more than one host. Many viruses follow several stages to infect host cells. These stages include attachment, penetration, uncoating, biosynthesis, maturation, and release. Bacteriophages have a lytic or lysogenic cycle. The lytic cycle leads to the death of the host, whereas the lysogenic cycle leads to integration of phage into the host genome.

Multiple Choice

Which of the following leads to the destruction of the host cells?

A. lysogenic cycle
B. lytic cycle
C. prophage
D. temperate phage

Answer

B

A virus obtains its envelope during which of the following phases?

A. attachment
B. penetration
C. assembly
D. release

Answer

D

Which of the following components is brought into a cell by HIV?

A. a DNA-dependent DNA polymerase
B. RNA polymerase
C. ribosome
D. reverse transcriptase

Answer

D

A positive-strand RNA virus:

A. must first be converted to a mRNA before it can be translated.
B. can be used directly to translate viral proteins.
C. will be degraded by host enzymes.
D. is not recognized by host ribosomes.

Answer

B

What is the name for the transfer of genetic information from one bacterium to another bacterium by a phage?

A. transduction
B. penetration

C. excision
D. translation

Answer

A

Fill in the Blank

An enzyme from HIV that can make a copy of DNA from RNA is called _____.

Answer

reverse transcriptase

For lytic viruses, _____ is a phase during a viral growth curve when the virus is not detected.

Answer

eclipse

Short Answer

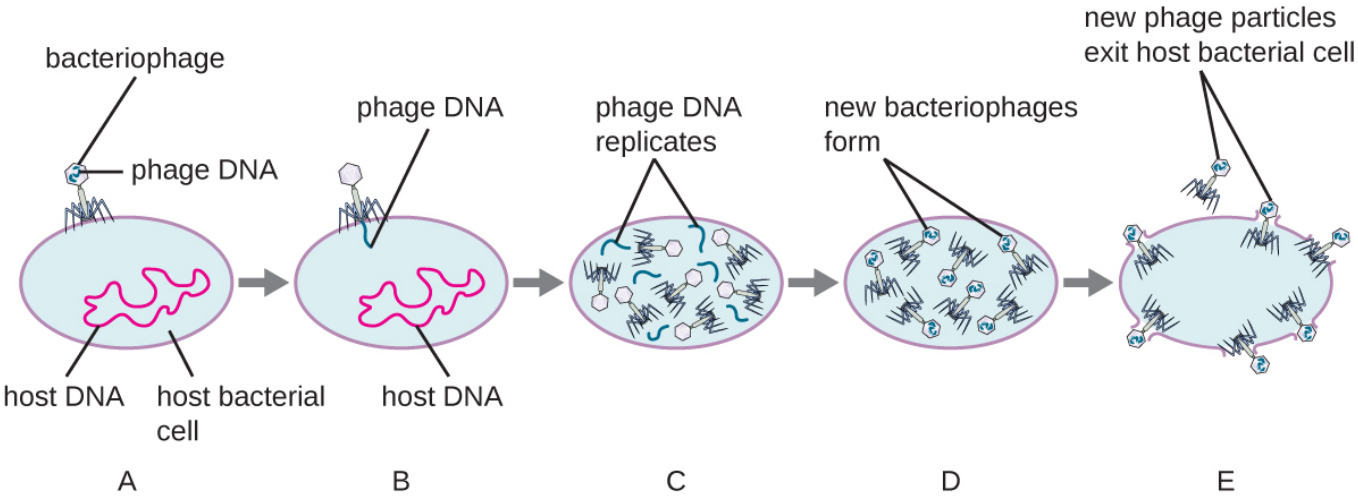
Briefly explain the difference between the mechanism of entry of a T-even bacteriophage and an animal virus.

Discuss the difference between generalized and specialized transduction.

Differentiate between lytic and lysogenic cycles.

Critical Thinking

Label the five stages of a bacteriophage infection in the figure:



Bacteriophages have lytic and lysogenic cycles. Discuss the advantages and disadvantages for the phage.

How does reverse transcriptase aid a retrovirus in establishing a chronic infection?

Discuss some methods by which plant viruses are transmitted from a diseased plant to a healthy one.

7.3: Isolation, Culture, and Identification of Viruses

Viral cultivation requires the presence of some form of host cell (whole organism, embryo, or cell culture). Viruses can be isolated from samples by filtration. Viral filtrate is a rich source of released virions. Bacteriophages are detected by presence of clear plaques on bacterial lawn. Animal and plant viruses are detected by cytopathic effects, molecular techniques (PCR, RT-PCR), enzyme immunoassays, and serological assays (hemagglutination assay, hemagglutination inhibition assay).

Multiple Choice

Which of the followings cannot be used to culture viruses?

A. tissue culture
B. liquid medium only
C. embryo
D. animal host

Answer

B

Which of the following tests can be used to detect the presence of a specific virus?

A. EIA
B. RT-PCR
C. PCR
D. all of the above

Answer

D

Which of the following is NOT a cytopathic effect?

A. transformation
B. cell fusion
C. mononucleated cell
D. inclusion bodies

Answer

C

Fill in the Blank

Viruses can be diagnosed and observed using a(n) _____ microscope.

Answer

Electron

Cell abnormalities resulting from a viral infection are called _____ _____.

Answer

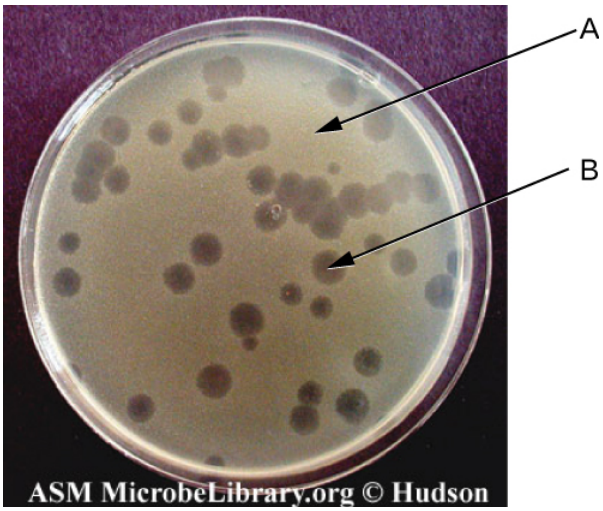
cytopathic effects

Short Answer

Briefly explain the various methods of culturing viruses.

Critical Thinking

Label the components indicated by arrows.



(credit: modification of work by American Society for Microbiology)

What are some characteristics of the viruses that are similar to a computer virus?

7.4: Viroids, Virusoids, and Prions

Other acellular agents such as viroids, virusoids, and prions also cause diseases. Viroids consist of small, naked ssRNAs that cause diseases in plants. Virusoids are ssRNAs that require other helper viruses to establish an infection. Prions are proteinaceous infectious particles that cause transmissible spongiform encephalopathies. Prions are extremely resistant to chemicals, heat, and radiation.

Multiple Choice

Which of these infectious agents do not have nucleic acid?

A. viroids
B. viruses
C. bacteria
D. prions

Answer

D

Which of the following is true of prions?

A. They can be inactivated by boiling at 100 °C.
B. They contain a capsid.
C. They are a rogue form of protein, PrP.
D. They can be reliably inactivated by an autoclave.

Answer

C

Fill in the Blank

Both viroids and virusoids have a(n) _____ genome, but virusoids require a(n) _____ to reproduce.

Answer

RNA, helper virus

Short Answer

Describe the disease symptoms observed in animals infected with prions.

Critical Thinking

Does a prion replicate? Explain.

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CHAPTER OVERVIEW

8: Acellular Pathogens

Public health measures in the developed world have dramatically reduced mortality from viral epidemics. But when epidemics do occur, they can spread quickly with global air travel. In 2009, an outbreak of H1N1 influenza spread across various continents. In early 2014, cases of Ebola in Guinea led to a massive epidemic in western Africa. This included the case of an infected man who traveled to the United States, sparking fears the epidemic might spread beyond Africa.

Until the late 1930s and the advent of the electron microscope, no one had seen a virus. Yet treatments for preventing or curing viral infections were used and developed long before that. Historical records suggest that by the 17th century, and perhaps earlier, inoculation (also known as variolation) was being used to prevent the viral disease smallpox in various parts of the world. By the late 18th century, Englishman Edward Jenner was inoculating patients with cowpox to prevent smallpox, a technique he coined *vaccination*.[1]

Today, the structure and genetics of viruses are well defined, yet new discoveries continue to reveal their complexities. In this chapter, we will learn about the structure, classification, and cultivation of viruses, and how they impact their hosts. In addition, we will learn about other infective particles such as viroids and prions.

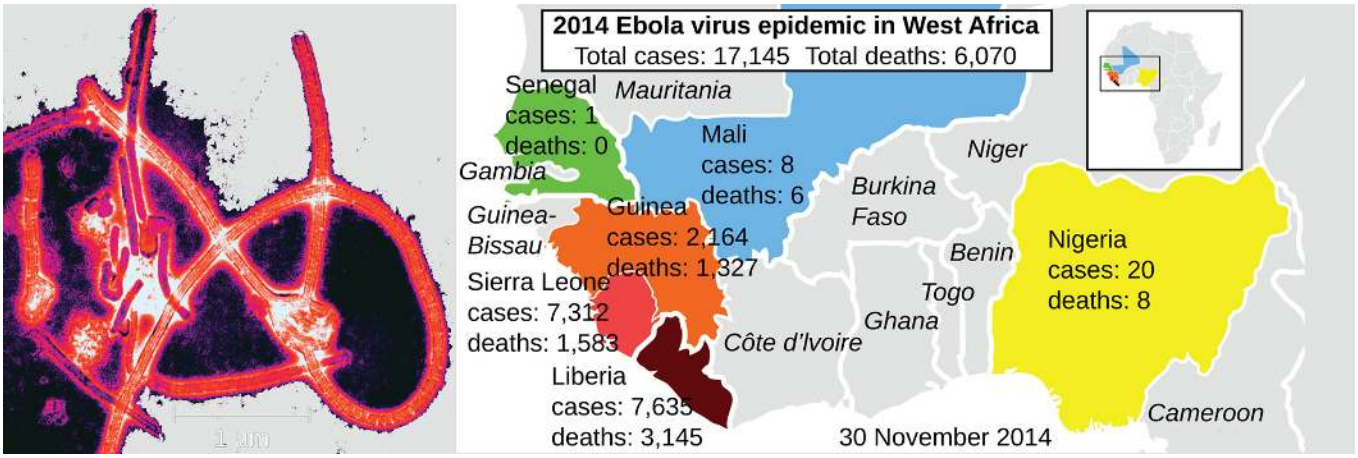


Figure 8.1: The year 2014 saw the first large-scale outbreak of Ebola virus (electron micrograph, left) in human populations in West Africa (right). Such epidemics are now widely reported and documented, but viral epidemics are sure to have plagued human populations since the origin of our species. (credit left: modification of work by Thomas W. Geisbert)

8.1: Isolation, Culture, and Identification of Viruses
8.2: Viroids, Virusoids, and Prions
8.E: Acellular Pathogens (Exercises)

Footnotes

1. 1 S. Riedel "Edward Jenner and the History of Smallpox and Vaccination." *Baylor University Medical Center Proceedings* 18, no. 1 (January 2005): 21–25.

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8.1: Isolation, Culture, and Identification of Viruses

8.01: Isolation Culture and Identification of Viruses

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8.2: Viroids, Virusoids, and Prions

8.02: Viroids Virusoids and Prions

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8.E: Acellular Pathogens (Exercises)

7.1: Viruses

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Multiple Choice

The component(s) of a virus that is/are extended from the envelope for attachment is/are the:

A. capsomeres
B. spikes
C. nucleic acid
D. viral whiskers

Answer

B

Which of the following does a virus lack? Select all that apply.

A. ribosomes
B. metabolic processes
C. nucleic acid
D. glycoprotein

Answer

A and B

The envelope of a virus is derived from the host's

A. nucleic acids
B. membrane structures
C. cytoplasm
D. genome

Answer

B

In naming viruses, the family name ends with _____ and genus name ends with _____.

A. *−virus; −viridae*
B. *−viridae; −virus*
C. *−virion; virus*
D. *−virus; virion*

Answer

B

What is another name for a nonenveloped virus?

A. enveloped virus
B. provirus
C. naked virus
D. latent virus

Answer

C

True/False

True or False: Scientists have identified viruses that are able to infect fungal cells.

Answer

True

Fill in the Blank

A virus that infects a bacterium is called a/an _____.

Answer

bacteriophage

A/an _____ virus possesses characteristics of both a polyhedral and helical virus.

Answer

complex

A virus containing only nucleic acid and a capsid is called a/an _____ virus or _____ virus.

Answer

naked or nonenveloped

The _____ _____ on the bacteriophage allow for binding to the bacterial cell.

Answer

tail fibers

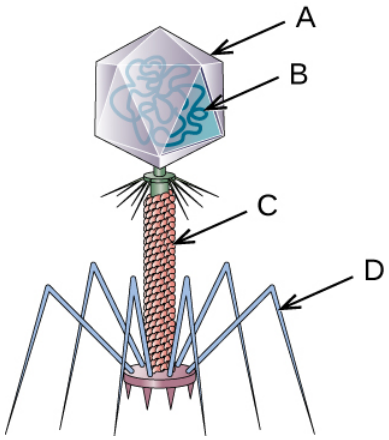
Short Answer

Discuss the geometric differences among helical, polyhedral, and complex viruses.

What was the meaning of the word “virus” in the 1880s and why was it used to describe the cause of tobacco mosaic disease?

Critical Thinking

Name each labeled part of the illustrated bacteriophage.



In terms of evolution, which do you think arises first? The virus or the host? Explain your answer.

Do you think it is possible to create a virus in the lab? Imagine that you are a mad scientist. Describe how you would go about creating a new virus.

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Multiple Choice

Which of the following leads to the destruction of the host cells?

A. lysogenic cycle
B. lytic cycle
C. prophage
D. temperate phage

Answer

B

A virus obtains its envelope during which of the following phases?

A. attachment
B. penetration
C. assembly
D. release

Answer

D

Which of the following components is brought into a cell by HIV?

A. a DNA-dependent DNA polymerase
B. RNA polymerase
C. ribosome
D. reverse transcriptase

Answer

D

A positive-strand RNA virus:

A. must first be converted to a mRNA before it can be translated.
B. can be used directly to translate viral proteins.
C. will be degraded by host enzymes.
D. is not recognized by host ribosomes.

Answer

B

What is the name for the transfer of genetic information from one bacterium to another bacterium by a phage?

A. transduction
B. penetration

C. excision
D. translation

Answer

A

Fill in the Blank

An enzyme from HIV that can make a copy of DNA from RNA is called _____.

Answer

reverse transcriptase

For lytic viruses, _____ is a phase during a viral growth curve when the virus is not detected.

Answer

eclipse

Short Answer

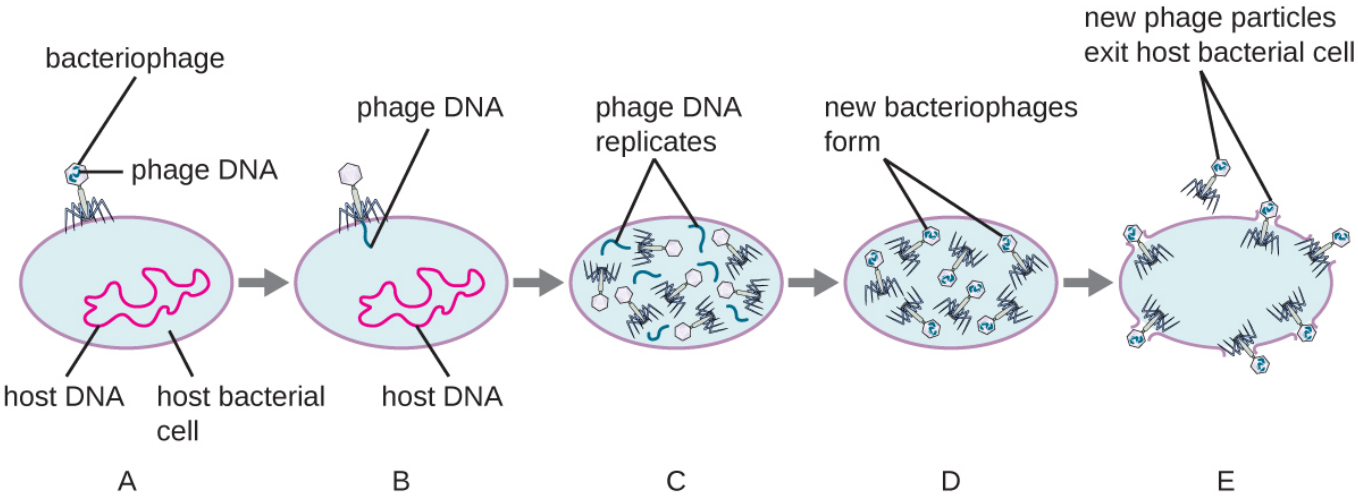
Briefly explain the difference between the mechanism of entry of a T-even bacteriophage and an animal virus.

Discuss the difference between generalized and specialized transduction.

Differentiate between lytic and lysogenic cycles.

Critical Thinking

Label the five stages of a bacteriophage infection in the figure:



Bacteriophages have lytic and lysogenic cycles. Discuss the advantages and disadvantages for the phage.

How does reverse transcriptase aid a retrovirus in establishing a chronic infection?

Discuss some methods by which plant viruses are transmitted from a diseased plant to a healthy one.

7.3: Isolation, Culture, and Identification of Viruses

Viral cultivation requires the presence of some form of host cell (whole organism, embryo, or cell culture). Viruses can be isolated from samples by filtration. Viral filtrate is a rich source of released virions. Bacteriophages are detected by presence of clear plaques on bacterial lawn. Animal and plant viruses are detected by cytopathic effects, molecular techniques (PCR, RT-PCR), enzyme immunoassays, and serological assays (hemagglutination assay, hemagglutination inhibition assay).

Multiple Choice

Which of the followings cannot be used to culture viruses?

A. tissue culture
B. liquid medium only
C. embryo
D. animal host

Answer

B

Which of the following tests can be used to detect the presence of a specific virus?

A. EIA
B. RT-PCR
C. PCR
D. all of the above

Answer

D

Which of the following is NOT a cytopathic effect?

A. transformation
B. cell fusion
C. mononucleated cell
D. inclusion bodies

Answer

C

Fill in the Blank

Viruses can be diagnosed and observed using a(n) _____ microscope.

Answer

Electron

Cell abnormalities resulting from a viral infection are called _____ _____.

Answer

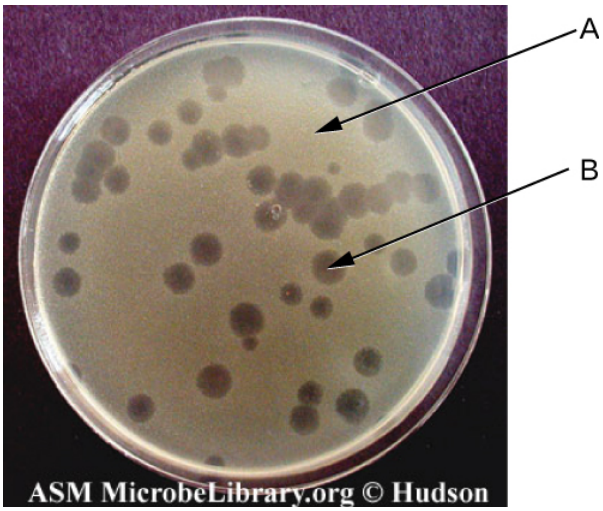
cytopathic effects

Short Answer

Briefly explain the various methods of culturing viruses.

Critical Thinking

Label the components indicated by arrows.



(credit: modification of work by American Society for Microbiology)

What are some characteristics of the viruses that are similar to a computer virus?

7.4: Viroids, Virusoids, and Prions

Other acellular agents such as viroids, virusoids, and prions also cause diseases. Viroids consist of small, naked ssRNAs that cause diseases in plants. Virusoids are ssRNAs that require other helper viruses to establish an infection. Prions are proteinaceous infectious particles that cause transmissible spongiform encephalopathies. Prions are extremely resistant to chemicals, heat, and radiation.

Multiple Choice

Which of these infectious agents do not have nucleic acid?

A. viroids
B. viruses
C. bacteria
D. prions

Answer

D

Which of the following is true of prions?

A. They can be inactivated by boiling at 100 °C.
B. They contain a capsid.
C. They are a rogue form of protein, PrP.
D. They can be reliably inactivated by an autoclave.

Answer

C

Fill in the Blank

Both viroids and virusoids have a(n) _____ genome, but virusoids require a(n) _____ to reproduce.

Answer

RNA, helper virus

Short Answer

Describe the disease symptoms observed in animals infected with prions.

Critical Thinking

Does a prion replicate? Explain.

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CHAPTER OVERVIEW

9: Microbial Metabolism I

Throughout earth's history, microbial metabolism has been a driving force behind the development and maintenance of the planet's biosphere. Eukaryotic organisms such as plants and animals typically depend on organic molecules for energy, growth, and reproduction. Prokaryotes, on the other hand, can metabolize a wide range of organic as well as inorganic matter, from complex organic molecules like cellulose to inorganic molecules and ions such as atmospheric nitrogen (N_2), molecular hydrogen (H_2), sulfide (S^{2-}), manganese (II) ions (Mn^{2+}), ferrous iron (Fe^{2+}), and ferric iron (Fe^{3+}), to name a few. By metabolizing such substances, microbes chemically convert them to other forms. In some cases, microbial metabolism produces chemicals that can be harmful to other organisms; in others, it produces substances that are essential to the metabolism and survival of other life forms (Figure **9.1**).



Figure **9.1**: Prokaryotes have great metabolic diversity with important consequences to other forms of life. Acidic mine drainage (left) is a serious environmental problem resulting from the introduction of water and oxygen to sulfide-oxidizing bacteria during mining processes. These bacteria produce large amounts of sulfuric acid as a byproduct of their metabolism, resulting in a low-pH environment that can kill many aquatic plants and animals. On the other hand, some prokaryotes are essential to other life forms. Root nodules of many plants (right) house nitrogen-fixing bacteria that convert atmospheric nitrogen into ammonia, providing a usable nitrogen source for these plants. (credit left: modification of work by D. Hardesty, USGS Columbia Environment Research Center; credit right: modification of work by Celmow SR, Clairmont L, Madsen LH, and Guinel FC)

9.1: Energy, Matter, and Enzymes

9.2: Catabolism of Carbohydrates

9.3: Cellular Respiration

9.4: Fermentation

9.5: Catabolism of Lipids and Proteins

9.6: Photosynthesis and the Importance of Light

9.7: Biogeochemical Cycles

9.E: Microbial Metabolism (Exercises)

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9.1: Energy, Matter, and Enzymes

Learning Objectives

- Define and describe metabolism
- Compare and contrast autotrophs and heterotrophs
- Describe the importance of oxidation-reduction reactions in metabolism
- Describe why ATP, FAD, NAD^+, and $NADP^+$ are important in a cell
- Identify the structure and structural components of an enzyme
- Describe the differences between competitive and noncompetitive enzyme inhibitors

The term used to describe all of the chemical reactions inside a cell is **metabolism** (Figure **9.1.1**). Cellular processes such as the building or breaking down of complex molecules occur through series of stepwise, interconnected chemical reactions called metabolic pathways. Reactions that are spontaneous and release energy are **exergonic reactions**, whereas **endergonic reactions** require energy to proceed. The term **anabolism** refers to those endergonic metabolic pathways involved in biosynthesis, converting simple molecular building blocks into more complex molecules, and fueled by the use of cellular energy. Conversely, the term **catabolism** refers to exergonic pathways that break down complex molecules into simpler ones. Molecular energy stored in the bonds of complex molecules is released in catabolic pathways and harvested in such a way that it can be used to produce high-energy molecules, which are used to drive anabolic pathways. Thus, in terms of energy and molecules, cells are continually balancing catabolism with anabolism.

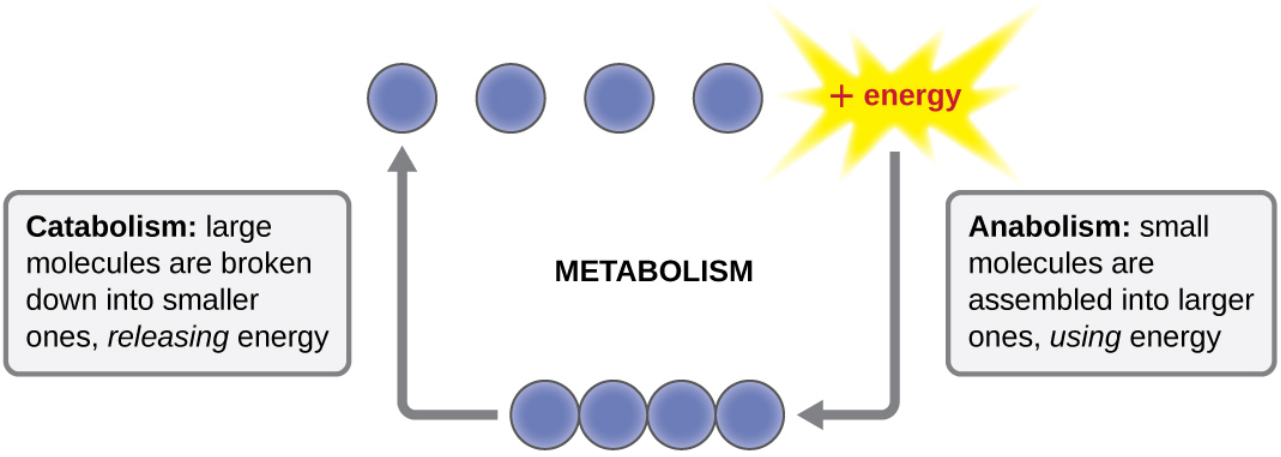


Figure **9.1.1**: Metabolism includes catabolism and anabolism. Anabolic pathways require energy to synthesize larger molecules. Catabolic pathways generate energy by breaking down larger molecules. Both types of pathways are required for maintaining the cell's energy balance.

Classification by Carbon and Energy Source

Organisms can be identified according to the source of carbon they use for metabolism as well as their energy source. The prefixes auto- ("self") and hetero- ("other") refer to the origins of the carbon sources various organisms can use. Organisms that convert inorganic carbon dioxide (CO_2) into organic carbon compounds are autotrophs. Plants and cyanobacteria are well-known examples of **autotrophs**. Conversely, **heterotrophs** rely on more complex organic carbon compounds as nutrients; these are provided to them initially by autotrophs. Many organisms, ranging from humans to many prokaryotes, including the well-studied *Escherichia coli*, are heterotrophic.

Organisms can also be identified by the energy source they use. All energy is derived from the transfer of electrons, but the source of electrons differs between various types of organisms. The prefixes photo- ("light") and chemo- ("chemical") refer to the energy sources that various organisms use. Those that get their energy for electron transfer from light are **phototrophs**, whereas **chemotrophs** obtain energy for electron transfer by breaking chemical bonds. There are two types of chemotrophs: **organotrophs**

and **lithotrophs**. Organotrophs, including humans, fungi, and many prokaryotes, are chemotrophs that obtain energy from organic compounds. Lithotrophs ("litho" means "rock") are chemotrophs that get energy from inorganic compounds, including hydrogen sulfide (H_2S) and reduced iron. Lithotrophy is unique to the microbial world.

The strategies used to obtain both carbon and energy can be combined for the classification of organisms according to nutritional type. Most organisms are chemoheterotrophs because they use organic molecules as both their electron and carbon sources. Table **9.1.1** summarizes this and the other classifications.

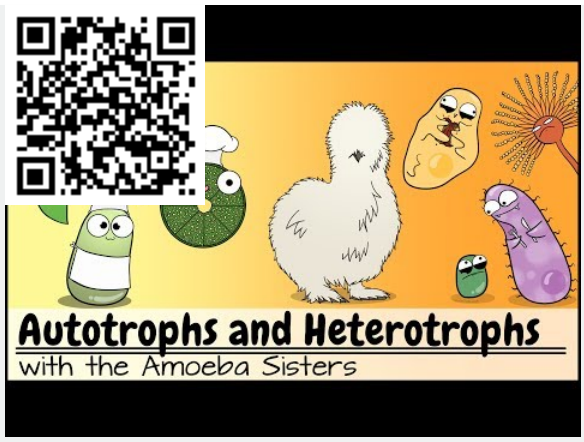
Table **9.1.1**: Classifications of Organisms by Energy and Carbon Source

Classifications		Energy Source	Carbon Source	Examples
Chemotrophs	Chemoautotrophs	Chemical	Inorganic	Hydrogen-, sulfur-, iron-, nitrogen-, and carbon monoxide-oxidizing bacteria
	Chemoheterotrophs	Chemical	Organic compounds	All animals, most fungi, protozoa, and bacteria
Phototrophs	Photoautotrophs	Light	Inorganic	All plants, algae, cyanobacteria, and green and purple sulfur bacteria
	Photoheterotrophs	Light	Organic compounds	Green and purple nonsulfur bacteria, heliobacteria

Video: Autotrophs and Heterotrophs

Curious about modes of nutrition? Join the Amoeba Sisters in learning about autotrophs and heterotrophs. Video explains these terms as well as how their carbon source differs. Photoautotrophs, photoheterotrophs, chemoautotrophs, and chemoheterotrophs, and their energy sources, are also discussed!





Oxidation and Reduction in Metabolism

The transfer of electrons between molecules is important because most of the energy stored in atoms and used to fuel cell functions is in the form of high-energy electrons. The transfer of energy in the form of electrons allows the cell to transfer and use energy incrementally; that is, in small packages rather than a single, destructive burst. Reactions that remove electrons from donor molecules, leaving them oxidized, are **oxidation reactions**; those that add electrons to acceptor molecules, leaving them reduced, are **reduction reactions**. Because electrons can move from one molecule to another, oxidation and reduction occur in tandem. These pairs of reactions are called oxidation-reduction reactions, or **redox reactions**.

Energy Carriers: NAD^+, $NADP^+$, FAD, and ATP

The energy released from the breakdown of the chemical bonds within nutrients can be stored either through the reduction of electron carriers or in the bonds of adenosine triphosphate (ATP). In living systems, a small class of compounds functions as mobile **electron carriers**, molecules that bind to and shuttle high-energy electrons between compounds in pathways. The principal electron carriers we will consider originate from the B vitamin group and are derivatives of nucleotides; they are **nicotinamide adenine dinucleotide**, **nicotine adenine dinucleotide phosphate**, and **flavin adenine dinucleotide**. These compounds can be easily reduced or oxidized. Nicotinamide adenine dinucleotide **(NAD^+/NADH)** is the most common mobile electron carrier used in catabolism. NAD^+ is the oxidized form of the molecule; NADH is the reduced form of the molecule. Nicotine adenine dinucleotide phosphate ($NADP^+$), the oxidized form of an NAD^+ variant that contains an extra phosphate group, is another important electron carrier; it forms NADPH when reduced. The oxidized form of flavin adenine dinucleotide is FAD, and its reduced form is $FADH_2$. Both NAD^+/NADH and **$FAD/FADH_2$** are extensively used in energy extraction from sugars during catabolism in chemoheterotrophs, whereas **$NADP^+$/NADPH** plays an important role in anabolic reactions and photosynthesis. Collectively, $FADH_2$, NADH, and NADPH are often referred to as having reducing power due to their ability to donate electrons to various chemical reactions.

A living cell must be able to handle the energy released during catabolism in a way that enables the cell to store energy safely and release it for use only as needed. Living cells accomplish this by using the compound **adenosine triphosphate (ATP)**. ATP is often called the “energy currency” of the cell, and, like currency, this versatile compound can be used to fill any energy need of the cell. At the heart of ATP is a molecule of **adenosine monophosphate (AMP)**, which is composed of an adenine molecule bonded to a ribose molecule and a single phosphate group. Ribose is a five-carbon sugar found in RNA, and AMP is one of the nucleotides in RNA. The addition of a second phosphate group to this core molecule results in the formation of **adenosine diphosphate (ADP)**; the addition of a third phosphate group forms ATP (Figure **9.1.2**). Adding a phosphate group to a molecule, a process called phosphorylation, requires energy. Phosphate groups are negatively charged and thus repel one another when they are arranged in series, as they are in ADP and ATP. This repulsion makes the ADP and ATP molecules inherently unstable. Thus, the bonds between phosphate groups (one in ADP and two in ATP) are called **high-energy phosphate bonds**. When these high-energy bonds are

broken to release one phosphate (called **inorganic phosphate [P_i]**) or two connected phosphate groups (called **pyrophosphate [PP_i]**) from ATP through a process called dephosphorylation, energy is released to drive endergonic reactions (Figure **9.1.3**).

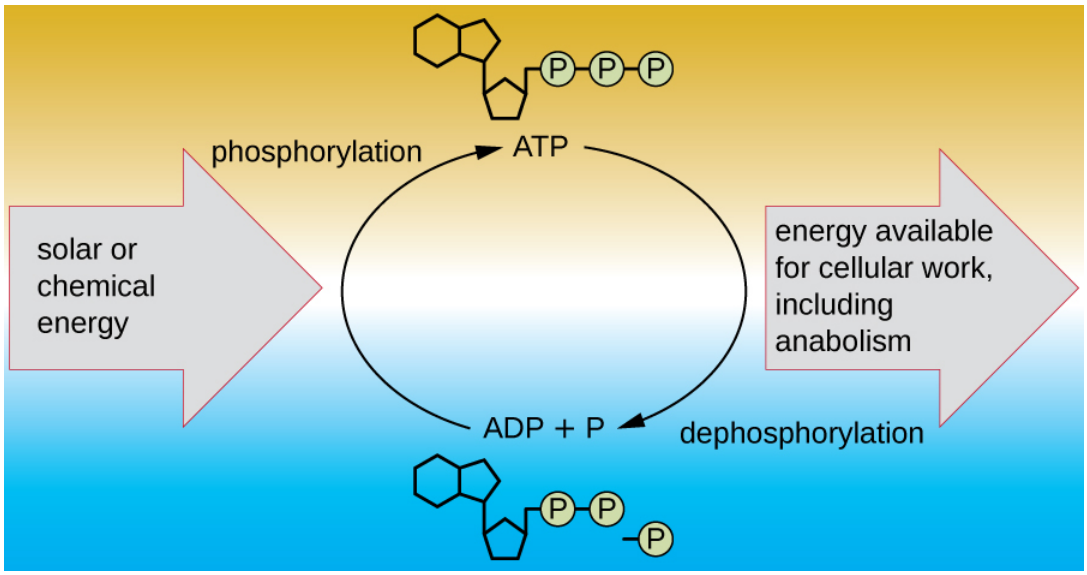


Figure **9.1.2**: The energy released from dephosphorylation of ATP is used to drive cellular work, including anabolic pathways. ATP is regenerated through phosphorylation, harnessing the energy found in chemicals or from sunlight. (credit: modification of work by Robert Bear, David Rintoul)

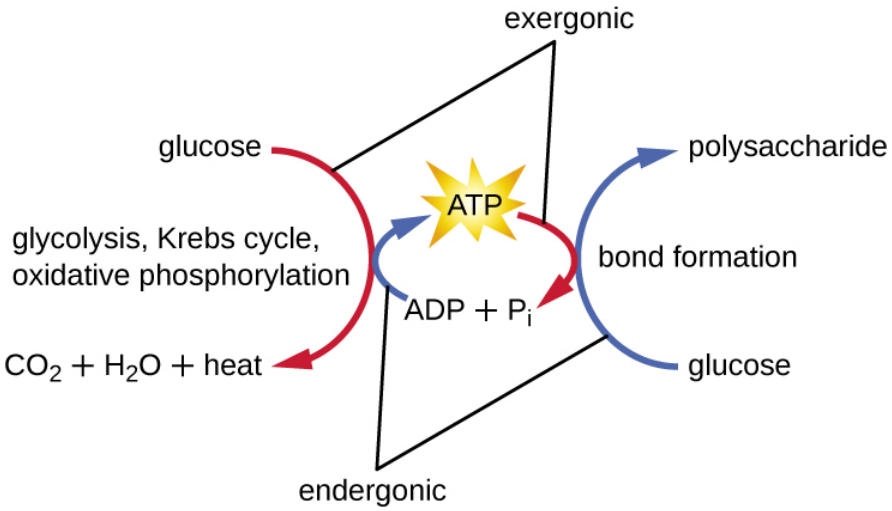


Figure **9.1.3**: Exergonic reactions are coupled to endergonic ones, making the combination favorable. Here, the endergonic reaction of ATP phosphorylation is coupled to the exergonic reactions of catabolism. Similarly, the exergonic reaction of ATP dephosphorylation is coupled to the endergonic reaction of polypeptide formation, an example of anabolism.

Key Concepts and Summary

- **Metabolism** includes chemical reactions that break down complex molecules (**catabolism**) and those that build complex molecules (**anabolism**).
- Organisms may be classified according to their source of carbon. **Autotrophs** convert inorganic carbon dioxide into organic carbon; **heterotrophs** use fixed organic carbon compounds.
- Organisms may also be classified according to their energy source. **Phototrophs** obtain their energy from light. **Chemotrophs** get their energy from chemical compounds. **Organotrophs** use organic molecules, and **lithotrophs** use inorganic chemicals.
- Cellular **electron carriers** accept high-energy electrons from foods and later serve as electron donors in subsequent **redox reactions**. **FAD/$FADH_2$, NAD^+/NADH, and $NADP^+$/NADPH** are important electron carriers.

- **Adenosine triphosphate (ATP)** serves as the energy currency of the cell, safely storing chemical energy in its two **high-energy phosphate bonds** for later use to drive processes requiring energy.

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9.2: Catabolism of Carbohydrates

Learning Objectives

- Describe why glycolysis is not oxygen dependent
- Define and describe the net yield of three-carbon molecules, ATP, and NADH from glycolysis
- Explain how three-carbon pyruvate molecules are converted into two-carbon acetyl groups that can be funneled into the Krebs cycle.
- Define and describe the net yield of CO_2, GTP/ATP, $FADH_2$, and NADH from the Krebs cycle
- Explain how intermediate carbon molecules of the Krebs cycle can be used in a cell

Extensive enzyme pathways exist for breaking down carbohydrates to capture energy in ATP bonds. In addition, many catabolic pathways produce intermediate molecules that are also used as building blocks for anabolism. Understanding these processes is important for several reasons. First, because the main metabolic processes involved are common to a wide range of chemoheterotrophic organisms, we can learn a great deal about human metabolism by studying metabolism in more easily manipulated bacteria like *E. coli*. Second, because animal and human pathogens are also chemoheterotrophs, learning about the details of metabolism in these bacteria, including possible differences between bacterial and human pathways, is useful for the diagnosis of pathogens as well as for the discovery of antimicrobial therapies targeting specific pathogens. Last, learning specifically about the pathways involved in chemoheterotrophic metabolism also serves as a basis for comparing other more unusual metabolic strategies used by microbes. Although the chemical source of electrons initiating electron transfer is different between chemoheterorophs and chemoautotrophs, many similar processes are used in both types of organisms.

The typical example used to introduce concepts of metabolism to students is carbohydrate catabolism. For chemoheterotrophs, our examples of metabolism start with the catabolism of polysaccharides such as glycogen, starch, or cellulose. Enzymes such as amylase, which breaks down glycogen or starch, and cellulases, which break down cellulose, can cause the hydrolysis of glycosidic bonds between the glucose monomers in these polymers, releasing glucose for further catabolism.

Glycolysis

For bacteria, eukaryotes, and most archaea, **glycolysis** is the most common pathway for the catabolism of glucose; it produces energy, reduced electron carriers, and precursor molecules for cellular metabolism. Every living organism carries out some form of glycolysis, suggesting this mechanism is an ancient universal metabolic process. The process itself does not use oxygen; however, glycolysis can be coupled with additional metabolic processes that are either aerobic or anaerobic. Glycolysis takes place in the cytoplasm of prokaryotic and eukaryotic cells. It begins with a single six-carbon glucose molecule and ends with two molecules of a three-carbon sugar called pyruvate. Pyruvate may be broken down further after glycolysis to harness more energy through aerobic or anaerobic respiration, but many organisms, including many microbes, may be unable to respire; for these organisms, glycolysis may be their only source of generating ATP.

The type of glycolysis found in animals and that is most common in microbes is the **Embden-Meyerhof-Parnas (EMP) pathway**, named after Gustav Embden (1874–1933), Otto Meyerhof (1884–1951), and Jakub Parnas (1884–1949). Glycolysis using the EMP pathway consists of two distinct phases (Figure **9.2.1**). The first part of the pathway, called the energy investment phase, uses energy from two ATP molecules to modify a glucose molecule so that the six-carbon sugar molecule can be split evenly into two phosphorylated three-carbon molecules called glyceraldehyde 3-phosphate (G3P). The second part of the pathway, called the energy payoff phase, extracts energy by oxidizing G3P to pyruvate, producing four ATP molecules and reducing two molecules of NAD^+ to two molecules of NADH, using electrons that originated from glucose. (A discussion and illustration of the full EMP pathway with chemical structures and enzyme names appear in Appendix C.)

The ATP molecules produced during the energy payoff phase of glycolysis are formed by **substrate-level phosphorylation** (Figure **9.2.1**), one of two mechanisms for producing ATP. In substrate-level phosphorylation, a phosphate group is removed from an organic molecule and is directly transferred to an available ADP molecule, producing ATP. During glycolysis, high-energy phosphate groups from the intermediate molecules are added to ADP to make ATP.

Overall, in this process of glycolysis, the net gain from the breakdown of a single glucose molecule is:

- two ATP molecules
- two NADH molecule, and
- two pyruvate molecules.

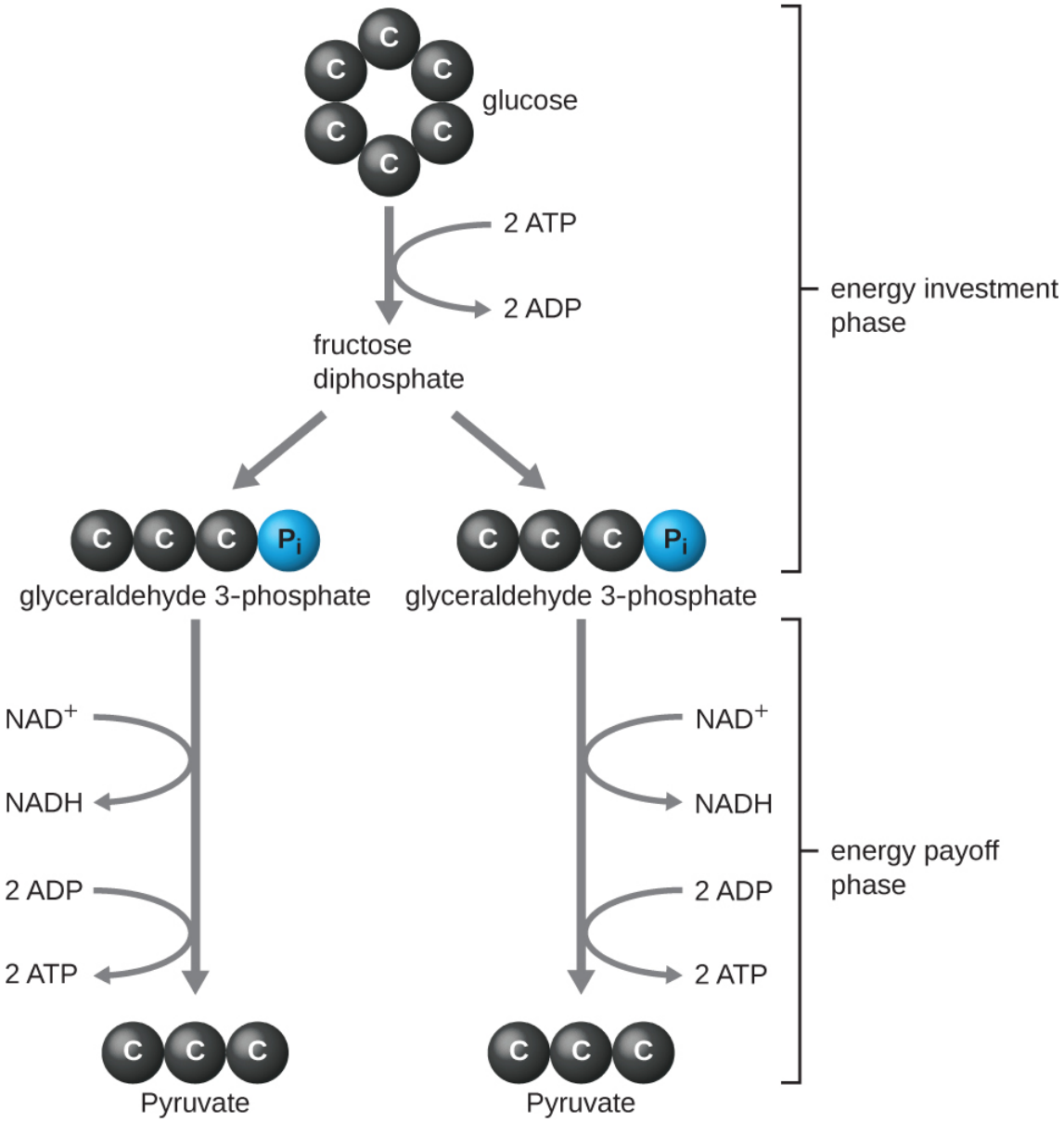


Figure 9.2.1: The energy investment phase of the Embden-Meyerhof-Parnas glycolysis pathway uses two ATP molecules to phosphorylate glucose, forming two glyceraldehyde 3-phosphate (G3P) molecules. The energy payoff phase harnesses the energy in the G3P molecules, producing four ATP molecules, two NADH molecules, and two pyruvates.

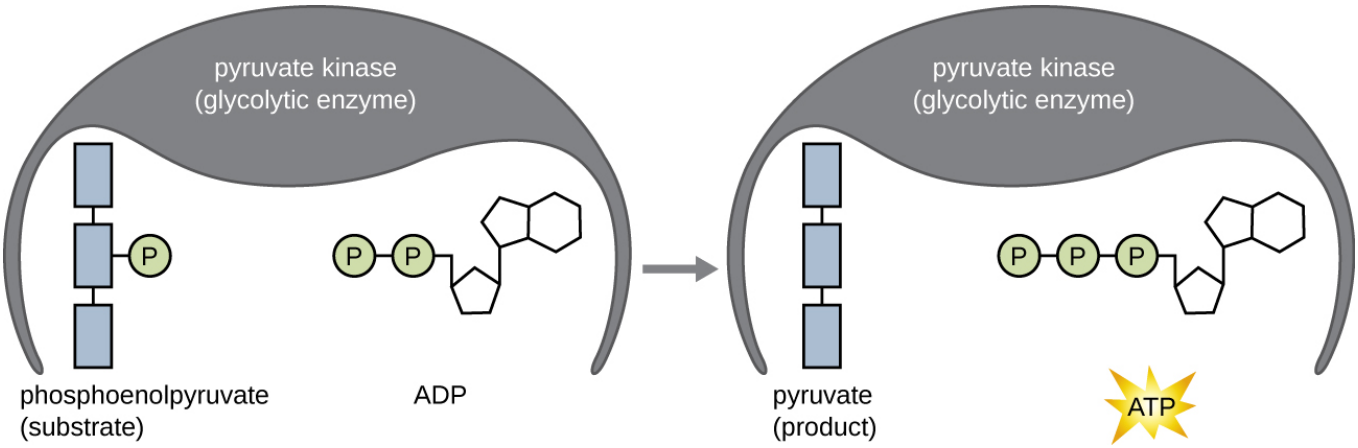


Figure **9.2.2**: The ATP made during glycolysis is a result of substrate-level phosphorylation. One of the two enzymatic reactions in the energy payoff phase of Embden Meyerhof-Parnas glycolysis that produce ATP in this way is shown here.

Other Glycolytic Pathways

When we refer to glycolysis, unless otherwise indicated, we are referring to the EMP pathway used by animals and many bacteria. However, some prokaryotes use alternative glycolytic pathways. One important alternative is the **Entner-Doudoroff (ED) pathway**, named after its discoverers Nathan Entner and Michael Doudoroff (1911–1975). Although some bacteria, including the opportunistic gram-negative pathogen *Pseudomonas aeruginosa*, contain only the ED pathway for glycolysis, other bacteria, like *E. coli*, have the ability to use either the ED pathway or the EMP pathway.

A third type of glycolytic pathway that occurs in all cells, which is quite different from the previous two pathways, is the **pentose phosphate pathway (PPP)** also called the phosphogluconate pathway or the **hexose monophosphate shunt**. Evidence suggests that the PPP may be the most ancient universal glycolytic pathway. The intermediates from the PPP are used for the biosynthesis of nucleotides and amino acids. Therefore, this glycolytic pathway may be favored when the cell has need for nucleic acid and/or protein synthesis, respectively. A discussion and illustration of the complete ED pathway and PPP with chemical structures and enzyme names appear in Appendix C.

Key Concepts and Summary

- **Glycolysis** is the first step in the breakdown of glucose, resulting in the formation of ATP, which is produced by **substrate-level phosphorylation**; NADH; and two pyruvate molecules. Glycolysis does not use oxygen and is not oxygen dependent.
- After glycolysis, a three-carbon pyruvate is decarboxylated to form a two-carbon acetyl group, coupled with the formation of NADH. The acetyl group is attached to a large carrier compound called coenzyme A.

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9.3: Cellular Respiration

9.03: Cellular Respiration

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9.4: Fermentation

9.04: Fermentation

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9.5: Catabolism of Lipids and Proteins

9.05: Catabolism of Lipids and Proteins

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9.6: Photosynthesis and the Importance of Light

9.06: Photosynthesis and the Importance of Light

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9.7: Biogeochemical Cycles

9.07: Biogeochemical Cycles

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9.E: Microbial Metabolism (Exercises)

8.1: Energy, Matter, and Enzymes

Cellular processes such as the building or breaking down of complex molecules occur through series of stepwise, interconnected chemical reactions called metabolic pathways. The term anabolism refers to those endergonic metabolic pathways involved in biosynthesis, converting simple molecular building blocks into more complex molecules, and fueled by the use of cellular energy.

Multiple Choice

Which of the following is an organism that obtains its energy from the transfer of electrons originating from chemical compounds and its carbon from an inorganic source?

A. chemoautotroph
B. chemoheterotroph
C. photoheterotroph
D. photoautotroph

Answer

A

Which of the following molecules is reduced?

A. NAD^+
B. FAD
C. O_2
D. NADPH

Answer

D

Enzymes work by which of the following?

A. increasing the activation energy
B. reducing the activation energy
C. making exergonic reactions endergonic
D. making endergonic reactions exergonic

Answer

B

To which of the following does a competitive inhibitor most structurally resemble?

A. the active site
B. the allosteric site
C. the substrate
D. a coenzyme

Answer

C

Which of the following are organic molecules that help enzymes work correctly?

A. cofactors
B. coenzymes
C. holoenzymes

D. apoenzymes

Answer

B

Fill in the Blank

Processes in which cellular energy is used to make complex molecules from simpler ones are described as _____.

Answer

anabolic

The loss of an electron from a molecule is called _____.

Answer

oxidation

The part of an enzyme to which a substrate binds is called the _____.

Answer

active site

True/False

Competitive inhibitors bind to allosteric sites.

Answer

False

Short Answer

In cells, can an oxidation reaction happen in the absence of a reduction reaction? Explain.

What is the function of molecules like NAD^+/NADH and FAD/$FADH_2$ in cells?

8.2: Catabolism of Carbohydrates

Glycolysis is the first step in the breakdown of glucose, resulting in the formation of ATP, which is produced by substrate-level phosphorylation; NADH; and two pyruvate molecules. Glycolysis does not use oxygen and is not oxygen dependent. After glycolysis, a three-carbon pyruvate is decarboxylated to form a two-carbon acetyl group, coupled with the formation of NADH. The acetyl group is attached to a large carrier compound called coenzyme A.

Multiple Choice

During which of the following is ATP not made by substrate-level phosphorylation?

A. Embden-Meyerhof pathway
B. Transition reaction
C. Krebs cycle
D. Entner-Doudoroff pathway

Answer

B

Which of the following products is made during Embden-Meyerhof glycolysis?

A. NAD^+
B. pyruvate

C. CO_2
D. two-carbon acetyl

Answer

B

During the catabolism of glucose, which of the following is produced only in the Krebs cycle?

A. ATP
B. NADH
C. NADPH
D. $FADH_2$

Answer

D

Which of the following is not a name for the cycle resulting in the conversion of a two-carbon acetyl to one ATP, two CO_2, one $FADH_2$, and three NADH molecules?

A. Krebs cycle
B. tricarboxylic acid cycle
C. Calvin cycle
D. citric acid cycle

Answer

C

True/False

Glycolysis requires oxygen or another inorganic final electron acceptor to proceed.

Answer

False

Fill in the Blank

Per turn of the Krebs cycle, one acetyl is oxidized, forming ____ CO_2, ____ ATP, ____ NADH, and ____ $FADH_2$molecules.

Answer

2; 1; 3; 1

Most commonly, glycolysis occurs by the _____ pathway.

Answer

Embden-Meyerhof

Short Answer

What is substrate-level phosphorylation? When does it occur during the breakdown of glucose to CO_2?

Why is the Krebs cycle important in both catabolism and anabolism?

Critical Thinking

What would be the consequences to a cell of having a mutation that knocks out coenzyme A synthesis?

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CHAPTER OVERVIEW

10: Microbial Metabolism II

Throughout earth's history, microbial metabolism has been a driving force behind the development and maintenance of the planet's biosphere. Eukaryotic organisms such as plants and animals typically depend on organic molecules for energy, growth, and reproduction. Prokaryotes, on the other hand, can metabolize a wide range of organic as well as inorganic matter, from complex organic molecules like cellulose to inorganic molecules and ions such as atmospheric nitrogen (N_2), molecular hydrogen (H_2), sulfide (S^{2-}), manganese (II) ions (Mn^{2+}), ferrous iron (Fe^{2+}), and ferric iron (Fe^{3+}), to name a few. By metabolizing such substances, microbes chemically convert them to other forms. In some cases, microbial metabolism produces chemicals that can be harmful to other organisms; in others, it produces substances that are essential to the metabolism and survival of other life forms (Figure **10.1**).



Figure **10.1**: Prokaryotes have great metabolic diversity with important consequences to other forms of life. Acidic mine drainage (left) is a serious environmental problem resulting from the introduction of water and oxygen to sulfide-oxidizing bacteria during mining processes. These bacteria produce large amounts of sulfuric acid as a byproduct of their metabolism, resulting in a low-pH environment that can kill many aquatic plants and animals. On the other hand, some prokaryotes are essential to other life forms. Root nodules of many plants (right) house nitrogen-fixing bacteria that convert atmospheric nitrogen into ammonia, providing a usable nitrogen source for these plants. (credit left: modification of work by D. Hardesty, USGS Columbia Environment Research Center; credit right: modification of work by Celmow SR, Clairmont L, Madsen LH, and Guinel FC)

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10.1: Cellular Respiration

Learning Objectives

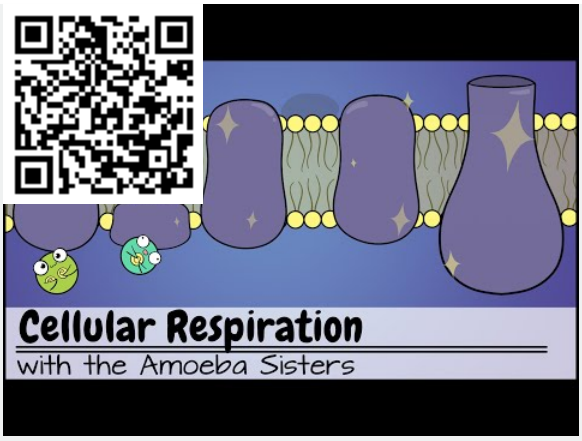
- Compare and contrast the electron transport system location and function in a prokaryotic cell and a eukaryotic cell
- Compare and contrast the differences between substrate-level and oxidative phosphorylation
- Explain the relationship between chemiosmosis and proton motive force
- Describe the function and location of ATP synthase in a prokaryotic versus eukaryotic cell
- Compare and contrast aerobic and anaerobic respiration

We have just discussed two pathways in glucose catabolism—glycolysis and the Krebs cycle—that generate ATP by substrate-level phosphorylation. Most ATP, however, is generated during a separate process called oxidative phosphorylation, which occurs during cellular respiration. Cellular respiration begins when electrons are transferred from NADH and $FADH_2$—made in glycolysis, the transition reaction, and the Krebs cycle—through a series of chemical reactions to a final inorganic electron acceptor (either oxygen in aerobic respiration or non-oxygen inorganic molecules in anaerobic respiration). These electron transfers take place on the inner part of the cell membrane of prokaryotic cells or in specialized protein complexes in the inner membrane of the mitochondria of eukaryotic cells. The energy of the electrons is harvested to generate an electrochemical gradient across the membrane, which is used to make ATP by oxidative phosphorylation.

Video: Cellular Respiration

Explore the process of aerobic cellular respiration and why ATP production is so important in this updated cellular respiration video by The Amoeba Sisters! This video compares the cellular respiration equation to photosynthesis before exploring the basics of glycolysis, Krebs Cycle (Citric Acid Cycle), and the Electron Transport Chain (and chemiosmosis).





Electron Transport System

The **electron transport system (ETS)** is the last component involved in the process of cellular respiration; it comprises a series of membrane-associated protein complexes and associated mobile accessory electron carriers. Electron transport is a series of chemical reactions that resembles a bucket brigade in that electrons from NADH and $FADH_2$ are passed rapidly from one ETS electron carrier to the next. These carriers can pass electrons along in the ETS because of their **redox potential**. For a protein or chemical to accept electrons, it must have a more positive redox potential than the electron donor. Therefore, electrons move from electron carriers with more negative redox potential to those with more positive redox potential. The four major classes of electron carriers involved in both eukaryotic and prokaryotic electron transport systems are the cytochromes, flavoproteins, iron-sulfur proteins, and the quinones.

In **aerobic respiration**, the final electron acceptor (i.e., the one having the most positive redox potential) at the end of the ETS is an oxygen molecule (O_2) that becomes reduced to water (H_2O) by the final ETS carrier. This electron carrier, **cytochrome oxidase**, differs between bacterial types and can be used to differentiate closely related bacteria for diagnoses. For example, the gram-negative opportunist *Pseudomonas aeruginosa* and the gram-negative cholera-causing *Vibrio cholerae* use cytochrome c oxidase, which can be detected by the oxidase test, whereas other gram-negative Enterobacteriaceae, like *E. coli*, are negative for this test because they produce different cytochrome oxidase types.

There are many circumstances under which aerobic respiration is not possible, including any one or more of the following:

- The cell lacks genes encoding an appropriate cytochrome oxidase for transferring electrons to oxygen at the end of the electron transport system.
- The cell lacks genes encoding enzymes to minimize the severely damaging effects of dangerous oxygen radicals produced during aerobic respiration, such as hydrogen peroxide (H_2O_2) or superoxide ($\mathbf{O_2^-}$).
- The cell lacks a sufficient amount of oxygen to carry out aerobic respiration.

One possible alternative to aerobic respiration is **anaerobic respiration**, using an inorganic molecule other than oxygen as a final electron acceptor. There are many types of anaerobic respiration found in bacteria and archaea. Denitrifiers are important soil bacteria that use nitrate ($\mathbf{NO_3^-}$) and nitrite ($\mathbf{NO_2^-}$) as final electron acceptors, producing nitrogen gas (N_2). Many aerobically respiring bacteria, including *E. coli*, switch to using nitrate as a final electron acceptor and producing nitrite when oxygen levels have been depleted.

Microbes using anaerobic respiration commonly have an intact Krebs cycle, so these organisms can access the energy of the NADH and $FADH_2$ molecules formed. However, anaerobic respirers use altered ETS carriers encoded by their genomes, including distinct complexes for electron transfer to their final electron acceptors. Smaller electrochemical gradients are generated from these electron transfer systems, so less ATP is formed through anaerobic respiration.

Chemiosmosis, Proton Motive Force, and Oxidative Phosphorylation

In each transfer of an electron through the ETS, the electron loses energy, but with some transfers, the energy is stored as potential energy by using it to pump hydrogen ions (H^+) across a membrane. In prokaryotic cells, H^+ is pumped to the outside of the cytoplasmic membrane (called the periplasmic space in gram-negative and gram-positive bacteria), and in eukaryotic cells, they are pumped from the mitochondrial matrix across the inner mitochondrial membrane into the intermembrane space. There is an uneven distribution of H^+ across the membrane that establishes an electrochemical gradient because H^+ ions are positively charged (electrical) and there is a higher concentration (chemical) on one side of the membrane. This electrochemical gradient formed by the accumulation of H^+ (also known as a proton) on one side of the membrane compared with the other is referred to as the **proton motive force (PMF)**. Because the ions involved are H^+, a pH gradient is also established, with the side of the membrane having the higher concentration of H^+ being more acidic. Beyond the use of the PMF to make ATP, as discussed in this chapter, the PMF can also be used to drive other energetically unfavorable processes, including nutrient transport and flagella rotation for motility.

The potential energy of this electrochemical gradient generated by the ETS causes the H^+ to diffuse across a membrane (the plasma membrane in prokaryotic cells and the inner membrane in mitochondria in eukaryotic cells). This flow of hydrogen ions across the membrane, called **chemiosmosis**, must occur through a channel in the membrane via a membrane-bound enzyme complex called **ATP synthase** (Figure **10.1.1**). The tendency for movement in this way is much like water accumulated on one side of a dam, moving through the dam when opened. ATP synthase (like a combination of the intake and generator of a hydroelectric dam) is a complex protein that acts as a tiny generator, turning by the force of the H^+ diffusing through the enzyme, down their electrochemical gradient from where there are many mutually repelling H^+ to where there are fewer H^+. In prokaryotic cells, H^+ flows from the outside of the cytoplasmic membrane into the cytoplasm, whereas in eukaryotic mitochondria, H^+ flows from the intermembrane space to the mitochondrial matrix. The turning of the parts of this molecular machine regenerates ATP from ADP and inorganic phosphate (P_i) by oxidative phosphorylation, a second mechanism for making ATP that harvests the potential energy stored within an electrochemical gradient.

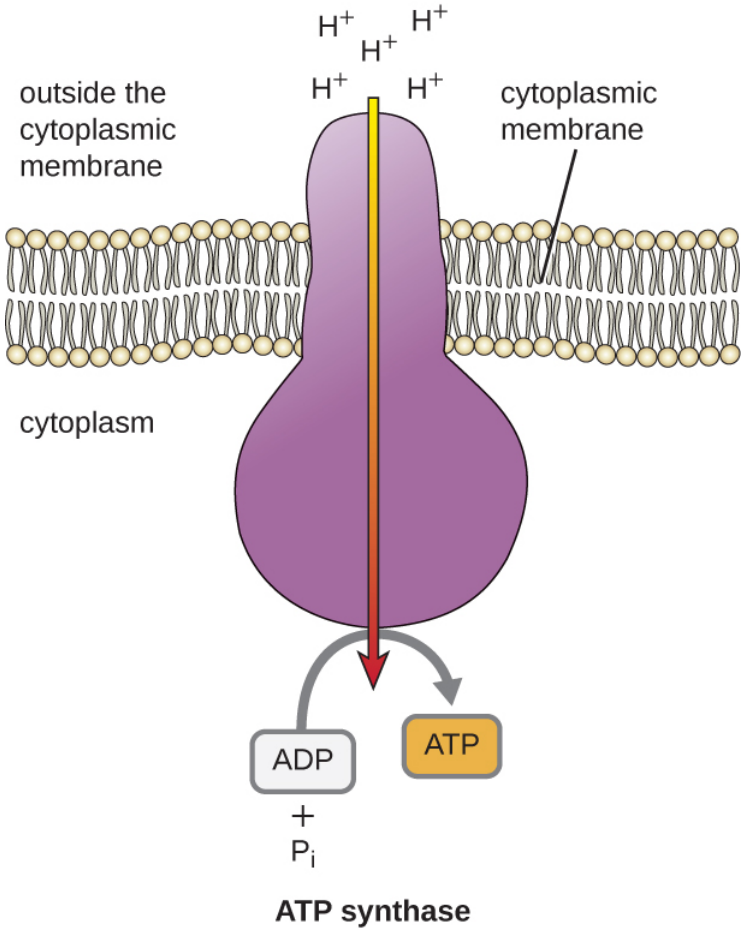


Figure **10.1.1**: ATP synthase is a complex integral membrane protein through which H^+ flows down an electrochemical gradient, providing the energy for ATP production by oxidative phosphorylation. (credit: modification of work by Klaus Hoffmeier)

The number of ATP molecules generated from the catabolism of glucose varies. For example, the number of hydrogen ions that the electron transport system complexes can pump through the membrane varies between different species of organisms. In aerobic respiration in mitochondria, the passage of electrons from one molecule of NADH generates enough proton motive force to make three ATP molecules by oxidative phosphorylation, whereas the passage of electrons from one molecule of $FADH_2$ generates enough proton motive force to make only two ATP molecules. Thus, the 10 NADH molecules made per glucose during glycolysis, the transition reaction, and the Krebs cycle carry enough energy to make 30 ATP molecules, whereas the two $FADH_2$ molecules made per glucose during these processes provide enough energy to make four ATP molecules. Overall, the theoretical maximum yield of ATP made during the complete aerobic respiration of glucose is 38 molecules, with four being made by substrate-level phosphorylation and 34 being made by oxidative phosphorylation (Figure **10.1.2**). In reality, the total ATP yield is usually less, ranging from one to 34 ATP molecules, depending on whether the cell is using aerobic respiration or anaerobic respiration; in eukaryotic cells, some energy is expended to transport intermediates from the cytoplasm into the mitochondria, affecting ATP yield.

Figure **10.1.2** summarizes the theoretical maximum yields of ATP from various processes during the complete aerobic respiration of one glucose molecule.

Source	Carbon Flow	Molecules of Reduced Coenzymes Produced	Net ATP Molecules Made by Substrate-Level Phosphory-lation	Net ATP Molecules Made by Oxidative Phosphory-lation	Theoretical Maximum Yield of ATP Molecules
Glycolysis (EMP)	Glucose (6C) ⟶ 2 pyruvates (2C)	2 NADH	2 ATP	6 ATP from 2 NADH	8
Transition reaction	2 pyruvates (3C) ⟶ 2 acetyl (2C) + 2 CO_2	2 NADH		6 ATP from 2 NADH	6
Krebs cycle	2 acetyl (2C) ⟶ 4 CO_2	6 NADH 2 $FADH_2$	2 ATP	18 ATP from 6 NADH 4 ATP from 2 $FADH_2$	24
Total:	glucose (6C) ⟶ 6 CO_2	10 NADH 2 $FADH_2$	4 ATP	34 ATP	38 ATP

Figure **10.1.2**: Theoretical maximum yields of ATP from various processes during the complete aerobic respiration of one glucose molecule.

Key Concepts and Summary

- Most ATP generated during the cellular respiration of glucose is made by **oxidative phosphorylation**.
- An **electron transport system (ETS)** is composed of a series of membrane-associated protein complexes and associated mobile accessory electron carriers. The ETS is embedded in the cytoplasmic membrane of prokaryotes and the inner mitochondrial membrane of eukaryotes.
- Each ETS complex has a different redox potential, and electrons move from electron carriers with more negative redox potential to those with more positive redox potential.
- To carry out **aerobic respiration**, a cell requires oxygen as the final electron acceptor. A cell also needs a complete Krebs cycle, an appropriate cytochrome oxidase, and oxygen detoxification enzymes to prevent the harmful effects of oxygen radicals produced during aerobic respiration.
- Organisms performing **anaerobic respiration** use alternative electron transport system carriers for the ultimate transfer of electrons to the final non-oxygen electron acceptors.
- Microbes show great variation in the composition of their electron transport systems, which can be used for diagnostic purposes to help identify certain pathogens.
- As electrons are passed from NADH and $FADH_2$ through an ETS, the electron loses energy. This energy is stored through the pumping of H^+ across the membrane, generating a **proton motive force**.
- The energy of this proton motive force can be harnessed by allowing hydrogen ions to diffuse back through the membrane by **chemiosmosis** using **ATP synthase**. As hydrogen ions diffuse through down their electrochemical gradient, components of ATP synthase spin, making ATP from ADP and P_i by oxidative phosphorylation.
- Aerobic respiration forms more ATP (a maximum of 34 ATP molecules) during oxidative phosphorylation than does anaerobic respiration (between one and 32 ATP molecules).

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10.2: Fermentation

Learning Objectives

- Define fermentation and explain why it does not require oxygen
- Describe the fermentation pathways and their end products and give examples of microorganisms that use these pathways
- Compare and contrast fermentation and anaerobic respiration

Many cells are unable to carry out respiration because of one or more of the following circumstances:

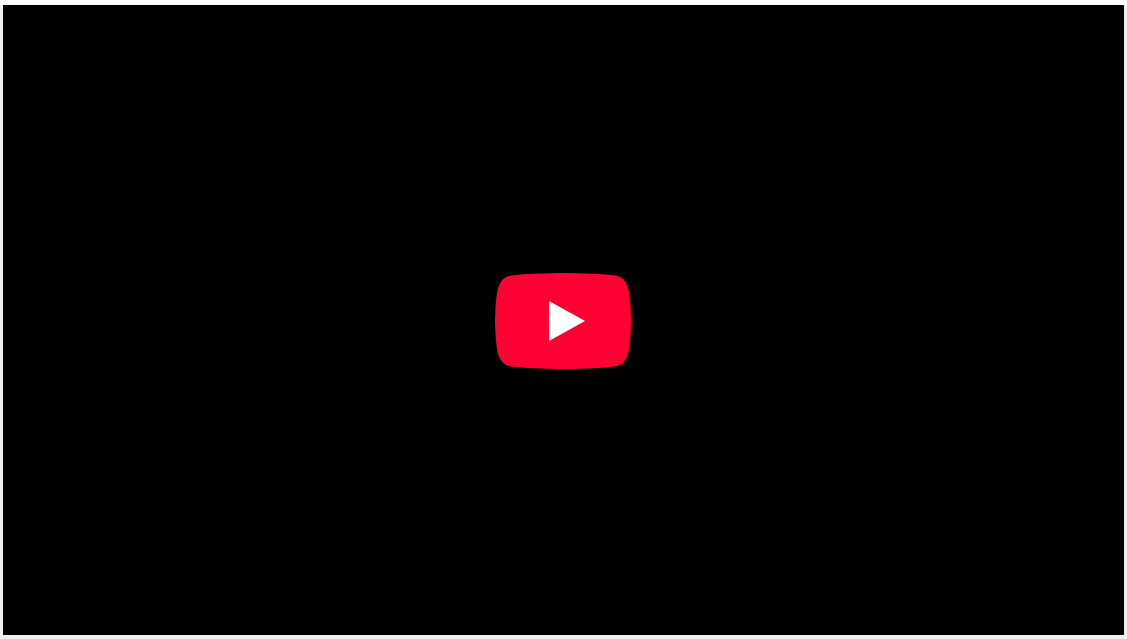
1. The cell lacks a sufficient amount of any appropriate, inorganic, final electron acceptor to carry out cellular respiration.
2. The cell lacks genes to make appropriate complexes and electron carriers in the electron transport system.
3. The cell lacks genes to make one or more enzymes in the Krebs cycle.

Whereas lack of an appropriate inorganic final electron acceptor is environmentally dependent, the other two conditions are genetically determined. Thus, many prokaryotes, including members of the clinically important genus *Streptococcus*, are permanently incapable of respiration, even in the presence of oxygen. Conversely, many prokaryotes are facultative, meaning that, should the environmental conditions change to provide an appropriate inorganic final electron acceptor for respiration, organisms containing all the genes required to do so will switch to cellular respiration for glucose metabolism because respiration allows for much greater ATP production per glucose molecule.

If respiration does not occur, NADH must be reoxidized to NAD^+ for reuse as an electron carrier for glycolysis, the cell's only mechanism for producing any ATP, to continue. Some living systems use an organic molecule (commonly pyruvate) as a final electron acceptor through a process called fermentation. Fermentation does not involve an electron transport system and does not directly produce any additional ATP beyond that produced during glycolysis by substrate-level phosphorylation. Organisms carrying out fermentation, called fermenters, produce a maximum of two ATP molecules per glucose during glycolysis. Table **10.2.1** compares the final electron acceptors and methods of ATP synthesis in aerobic respiration, anaerobic respiration, and fermentation. Note that the number of ATP molecules shown for glycolysis assumes the Embden-Meyerhof-Parnas pathway. The number of ATP molecules made by substrate-level phosphorylation (SLP) versus oxidative phosphorylation (OP) are indicated.

Video: Fermentation

What happens when you can't do aerobic cellular respiration because oxygen isn't available? Explore fermentation with The Amoeba Sisters! This video focuses on alcoholic fermentation and lactic acid fermentation, and it also mentions how this pathway is different from anaerobic cellular respiration.



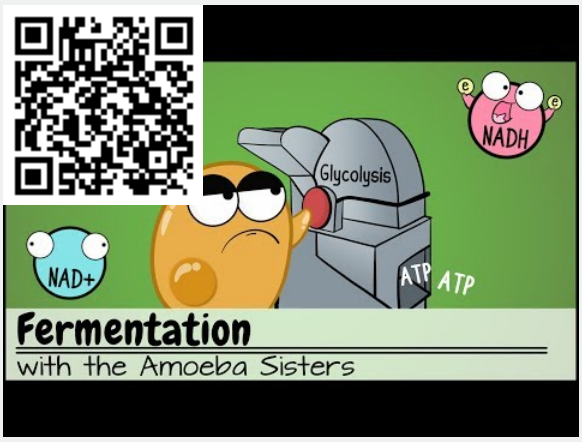


Table **10.2.1**: Comparison of Respiration Versus Fermentation

Type of Metabolism	Example	Final Electron Acceptor	Pathways Involved in ATP Synthesis (Type of Phosphorylation)	Maximum Yield of ATP Molecules
Aerobic respiration	*Pseudomonas aeruginosa*	O_2	EMP glycolysis (SLP) Krebs cycle (SLP) Electron transport and chemiosmosis (OP):	2 2 34
			Total	**38**
Anaerobic respiration	*Paracoccus denitrificans*	NO_3^-, SO_4^{-2}, Fe^{+3}, CO_2, other inorganics	EMP glycolysis (SLP) Krebs cycle (SLP) Electron transport and chemiosmosis (OP):	2 2 1–32
			Total	**5–36**
Fermentation	*Candida albicans*	Organics (usually pyruvate)	EMP glycolysis (SLP) Fermentation	2 0
			Total	**2**

Microbial fermentation processes have been manipulated by humans and are used extensively in the production of various foods and other commercial products, including pharmaceuticals. Microbial fermentation can also be useful for identifying microbes for diagnostic purposes.

Lactic Acid Fermentation

Fermentation by some bacteria, like those in yogurt and other soured food products, and by animals in muscles during oxygen depletion, is **lactic acid fermentation**. The chemical reaction of lactic acid fermentation is as follows:

$$\text{Pyruvate} + \text{NADH} \leftrightarrow \text{lactic acid} + \text{NAD}^+ \quad (10.2.1)$$

Bacteria of several gram-positive genera, including *Lactobacillus*, *Leuconostoc*, and *Streptococcus*, are collectively known as the **lactic acid bacteria (LAB)**, and various strains are important in food production. During yogurt and cheese production, the highly acidic environment generated by lactic acid fermentation denatures proteins contained in milk, causing it to solidify. When lactic acid is the only fermentation product, the process is said to be homolactic fermentation; such is the case for *Lactobacillus delbrueckii* and *S. thermophiles* used in yogurt production. However, many bacteria perform heterolactic fermentation, producing a mixture of lactic acid, ethanol and/or acetic acid, and CO_2 as a result, because of their use of the branched pentose phosphate pathway instead of the EMP pathway for glycolysis. One important heterolactic fermenter is *Leuconostoc mesenteroides*, which is used for souring vegetables like cucumbers and cabbage, producing pickles and sauerkraut, respectively.

Lactic acid bacteria are also important medically. The production of low pH environments within the body inhibits the establishment and growth of pathogens in these areas. For example, the vaginal microbiota is composed largely of lactic acid bacteria, but when these bacteria are reduced, yeast can proliferate, causing a yeast infection. Additionally, lactic acid bacteria are important in maintaining the health of the gastrointestinal tract and, as such, are the primary component of probiotics.

Alcoholic Fermentation

Another familiar fermentation process is **alcohol fermentation**, which produces ethanol. The ethanol fermentation reaction is shown in Figure **10.2.1**. In the first reaction, the enzyme pyruvate decarboxylase removes a carboxyl group from pyruvate, releasing CO_2 gas while producing the two-carbon molecule acetaldehyde. The second reaction, catalyzed by the enzyme alcohol dehydrogenase, transfers an electron from NADH to acetaldehyde, producing ethanol and NAD^+. The ethanol fermentation of pyruvate by the yeast *Saccharomyces cerevisiae* is used in the production of alcoholic beverages and also makes bread products rise due to CO_2 production. Outside of the food industry, ethanol fermentation of plant products is important in biofuel production.

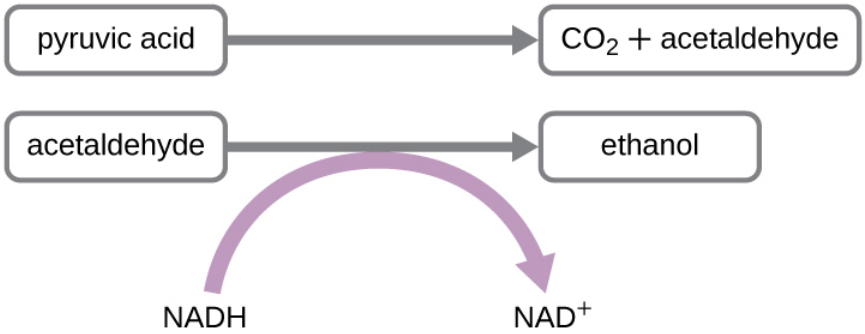


Figure **10.2.1**: The chemical reactions of alcohol fermentation are shown here. Ethanol fermentation is important in the production of alcoholic beverages and bread.

Key Concepts and Summary

- Fermentation uses an organic molecule as a final electron acceptor to regenerate NAD^+ from NADH so that glycolysis can continue.
- Fermentation does not involve an electron transport system, and no ATP is made by the fermentation process directly. Fermenters make very little ATP—only two ATP molecules per glucose molecule during glycolysis.
- Microbial fermentation processes have been used for the production of foods and pharmaceuticals, and for the identification of microbes.
- During lactic acid fermentation, pyruvate accepts electrons from NADH and is reduced to lactic acid. Microbes performing **homolactic fermentation** produce only lactic acid as the fermentation product; microbes performing **heterolactic fermentation** produce a mixture of lactic acid, ethanol and/or acetic acid, and CO_2.
- Lactic acid production by the normal microbiota prevents growth of pathogens in certain body regions and is important for the health of the gastrointestinal tract.
- During ethanol fermentation, pyruvate is first decarboxylated (releasing CO_2) to acetaldehyde, which then accepts electrons from NADH, reducing acetaldehyde to ethanol. Ethanol fermentation is used for the production of alcoholic beverages, for making bread products rise, and for biofuel production.

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10.3: Catabolism of Lipids and Proteins

Learning Objectives

- Describe how lipids are catabolized
- Describe how lipid catabolism can be used to identify microbes
- Describe how proteins are catabolized
- Describe how protein catabolism can be used to identify bacteria

Previous sections have discussed the catabolism of glucose, which provides energy to living cells, as well as how polysaccharides like glycogen, starch, and cellulose are degraded to glucose monomers. But microbes consume more than just carbohydrates for food. In fact, the microbial world is known for its ability to degrade a wide range of molecules, both naturally occurring and those made by human processes, for use as carbon sources. In this section, we will see that the pathways for both lipid and protein catabolism connect to those used for carbohydrate catabolism, eventually leading into glycolysis, the transition reaction, and the Krebs cycle pathways. Metabolic pathways should be considered to be porous—that is, substances enter from other pathways, and intermediates leave for other pathways. These pathways are not closed systems. Many of the substrates, intermediates, and products in a particular pathway are reactants in other pathways.

Lipid Catabolism

Triglycerides are a form of long-term energy storage in animals. They are made of glycerol and three fatty acids (see Figure 7.3.1). Phospholipids compose the cell and organelle membranes of all organisms except the archaea. Phospholipid structure is similar to triglycerides except that one of the fatty acids is replaced by a phosphorylated head group (see Figure 7.3.2). Triglycerides and phospholipids are broken down first by releasing fatty acid chains (and/or the phosphorylated head group, in the case of phospholipids) from the three-carbon glycerol backbone. The reactions breaking down triglycerides are catalyzed by **lipases** and those involving phospholipids are catalyzed by **phospholipases**. These enzymes contribute to the virulence of certain microbes, such as the bacterium *Staphylococcus aureus* and the fungus *Cryptococcus neoformans*. These microbes use phospholipases to destroy lipids and phospholipids in host cells and then use the catabolic products for energy (see Virulence Factors of Bacterial and Viral Pathogens).

The resulting products of lipid catabolism, glycerol and fatty acids, can be further degraded. Glycerol can be phosphorylated to glycerol-3-phosphate and easily converted to glyceraldehyde 3-phosphate, which continues through glycolysis. The released fatty acids are catabolized in a process called **β-oxidation**, which sequentially removes two-carbon acetyl groups from the ends of fatty acid chains, reducing NAD^+ and FAD to produce NADH and $FADH_2$, respectively, whose electrons can be used to make ATP by oxidative phosphorylation. The acetyl groups produced during β-oxidation are carried by coenzyme A to the Krebs cycle, and their movement through this cycle results in their degradation to CO_2, producing ATP by substrate-level phosphorylation and additional NADH and $FADH_2$ molecules (see Appendix C for a detailed illustration of β-oxidation).

Other types of lipids can also be degraded by certain microbes. For example, the ability of certain pathogens, like *Mycobacterium tuberculosis*, to degrade cholesterol contributes to their virulence. The side chains of cholesterol can be easily removed enzymatically, but degradation of the remaining fused rings is more problematic. The four fused rings are sequentially broken in a multistep process facilitated by specific enzymes, and the resulting products, including pyruvate, can be further catabolized in the Krebs cycle.

Protein Catabolism

Proteins are degraded through the concerted action of a variety of microbial **protease** enzymes. Extracellular proteases cut proteins internally at specific amino acid sequences, breaking them down into smaller peptides that can then be taken up by cells. Some clinically important pathogens can be identified by their ability to produce a specific type of extracellular protease. For example, the production of the extracellular protease **gelatinase** by members of the genera *Proteus* and *Serratia* can be used to distinguish them from other gram-negative enteric bacteria. Following inoculation and growth of microbes in gelatin broth, degradation of the gelatin protein due to gelatinase production prevents solidification of gelatin when refrigerated. Other pathogens can be distinguished by their ability to degrade casein, the main protein found in milk. When grown on skim milk agar, production of the

extracellular protease **caseinase** causes degradation of casein, which appears as a zone of clearing around the microbial growth. Caseinase production by the opportunist pathogen *Pseudomonas aeruginosa* can be used to distinguish it from other related gram-negative bacteria.

After extracellular protease degradation and uptake of peptides in the cell, the peptides can then be broken down further into individual amino acids by additional intracellular proteases, and each amino acid can be enzymatically deaminated to remove the amino group. The remaining molecules can then enter the transition reaction or the Krebs cycle.

Key Concepts and Summary

- Collectively, microbes have the ability to degrade a wide variety of carbon sources besides carbohydrates, including lipids and proteins. The catabolic pathways for all of these molecules eventually connect into glycolysis and the Krebs cycle.
- Several types of lipids can be microbially degraded. Triglycerides are degraded by extracellular **lipases**, releasing fatty acids from the glycerol backbone. Phospholipids are degraded by **phospholipases**, releasing fatty acids and the phosphorylated head group from the glycerol backbone. Lipases and phospholipases act as virulence factors for certain pathogenic microbes.
- Fatty acids can be further degraded inside the cell through **β-oxidation**, which sequentially removes two-carbon acetyl groups from the ends of fatty acid chains.
- Protein degradation involves extracellular **proteases** that degrade large proteins into smaller peptides. Detection of the extracellular proteases gelatinase and caseinase can be used to differentiate clinically relevant bacteria.

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10.4: Photosynthesis and the Importance of Light

10.04: Photosynthesis and the Importance of Light

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10.5: Biogeochemical Cycles

10.05: Biogeochemical Cycles

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10.E: Microbial Metabolism (Exercises)

9.1: Cellular Respiration

Cellular respiration begins when electrons are transferred from NADH and $FADH_2$—through a series of chemical reactions to a final inorganic electron acceptor (either oxygen in aerobic respiration or non-oxygen inorganic molecules in anaerobic respiration). These electron transfers take place on the inner part of the cell membrane of prokaryotic cells or in specialized protein complexes in the inner membrane of the mitochondria of eukaryotic cells.

Multiple Choice

Which is the location of electron transports systems in prokaryotes?

A. the outer mitochondrial membrane
B. the cytoplasm
C. the inner mitochondrial membrane
D. the cytoplasmic membrane

Answer

D

Which is the source of the energy used to make ATP by oxidative phosphorylation?

A. oxygen
B. high-energy phosphate bonds
C. the proton motive force
D. P_i

Answer

C

A cell might perform anaerobic respiration for which of the following reasons?

A. It lacks glucose for degradation.
B. It lacks the transition reaction to convert pyruvate to acetyl-CoA.
C. It lacks Krebs cycle enzymes for processing acetyl-CoA to CO_2.
D. It lacks a cytochrome oxidase for passing electrons to oxygen.

Answer

D

In prokaryotes, which of the following is true?

A. As electrons are transferred through an ETS, H^+ is pumped out of the cell.
B. As electrons are transferred through an ETS, H^+ is pumped into the cell.
C. As protons are transferred through an ETS, electrons are pumped out of the cell.
D. As protons are transferred through an ETS, electrons are pumped into the cell.

Answer

A

Which of the following is not an electron carrier within an electron transport system?

A. flavoprotein
B. ATP synthase
C. ubiquinone

D. cytochrome oxidase

Answer

B

Fill in the Blank

The final ETS complex used in aerobic respiration that transfers energy-depleted electrons to oxygen to form H_2O is called _____.

Answer

cytochrome oxidase

The passage of hydrogen ions through _____ down their electrochemical gradient harnesses the energy needed for ATP synthesis by oxidative phosphorylation.

Answer

ATP synthase

True/False

All organisms that use aerobic cellular respiration have cytochrome oxidase.

Answer

True

Short Answer

What is the relationship between chemiosmosis and the proton motive force?

How does oxidative phosphorylation differ from substrate-level phosphorylation?

How does the location of ATP synthase differ between prokaryotes and eukaryotes? Where do protons accumulate as a result of the ETS in each cell type?

9.2: Fermentation

Fermentation uses an organic molecule as a final electron acceptor to regenerate NAD^+ from NADH so that glycolysis can continue. Fermentation does not involve an electron transport system, and no ATP is made by the fermentation process directly. Fermenters make very little ATP—only two ATP molecules per glucose molecule during glycolysis. Microbial fermentation processes have been used for the production of foods and pharmaceuticals, and for the identification of microbes.

Multiple Choice

Which of the following is the purpose of fermentation?

A. to make ATP
B. to make carbon molecule intermediates for anabolism
C. to make NADH
D. to make NAD^+

Answer

D

Which molecule typically serves as the final electron acceptor during fermentation?

A. oxygen
B. NAD^+
C. pyruvate

D. CO_2

Answer

C

Which fermentation product is important for making bread rise?

A. ethanol
B. CO_2
C. lactic acid
D. hydrogen gas

Answer

B

Which of the following is not a commercially important fermentation product?

A. ethanol
B. pyruvate
C. butanol
D. penicillin

Answer

B

Fill in the Blank

The microbe responsible for ethanol fermentation for the purpose of producing alcoholic beverages is _____.

Answer

yeast (*Saccharomyces cerevisiae*)

_____ results in the production of a mixture of fermentation products, including lactic acid, ethanol and/or acetic acid, and CO_2.

Answer

Heterolactic fermentation

Fermenting organisms make ATP through the process of _____.

Answer

glycolysis

Matching

Match the fermentation pathway with the correct commercial product it is used to produce:

___acetone-butanol-ethanol fermentation	a. bread
___alcohol fermentation	b. pharmaceuticals
___lactic acid fermentation	c. Swiss cheese
___mixed acid fermentation	d. yogurt
___propionic acid fermentation	e. industrial solvents

Answer

e; 2. a; 3. d; 4. b; 5. c

Short Answer

Why are some microbes, including *Streptococcus* spp., unable to perform aerobic respiration, even in the presence of oxygen?

How can fermentation be used to differentiate various types of microbes?

Critical Thinking

The bacterium *E. coli* is capable of performing aerobic respiration, anaerobic respiration, and fermentation. When would it perform each process and why? How is ATP made in each case?

9.3: Catabolism of Lipids and Proteins

Collectively, microbes have the ability to degrade a wide variety of carbon sources besides carbohydrates, including lipids and proteins. The catabolic pathways for all of these molecules eventually connect into glycolysis and the Krebs cycle. Several types of lipids can be microbially degraded. Triglycerides are degraded by extracellular lipases, releasing fatty acids from the glycerol backbone. Phospholipids are degraded by phospholipases, releasing fatty acids and phosphorylated head groups.

Multiple Choice

Which of the following molecules is not produced during the breakdown of phospholipids?

A. glucose
B. glycerol
C. acetyl groups
D. fatty acids

Answer

A

Caseinase is which type of enzyme?

A. phospholipase
B. lipase
C. extracellular protease
D. intracellular protease

Answer

C

Which of the following is the first step in triglyceride degradation?

A. removal of fatty acids
B. β-oxidation
C. breakage of fused rings
D. formation of smaller peptides

Answer

A

Fill in the Blank

The process by which two-carbon units are sequentially removed from fatty acids, producing acetyl-CoA, $FADH_2$, and NADH is called _____.

Answer

β-oxidation

The NADH and $FADH_2$ produced during β-oxidation are used to make _____.

Answer

ATP by oxidative phosphorylation

_____ is a type of medium used to detect the production of an extracellular protease called caseinase.

Answer

Skim milk agar

Short Answer

How are the products of lipid and protein degradation connected to glucose metabolism pathways?

What is the general strategy used by microbes for the degradation of macromolecules?

Critical Thinking

Do you think that β-oxidation can occur in an organism incapable of cellular respiration? Why or why not?

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CHAPTER OVERVIEW

11: Microbial Growth

We are all familiar with the slimy layer on a pond surface or that makes rocks slippery. These are examples of biofilms—microorganisms embedded in thin layers of matrix material (Figure **11.1**). Biofilms were long considered random assemblages of cells and had little attention from researchers. Recently, progress in visualization and biochemical methods has revealed that biofilms are an organized ecosystem within which many cells, usually of different species of bacteria, fungi, and algae, interact through cell signaling and coordinated responses. The biofilm provides a protected environment in harsh conditions and aids colonization by microorganisms. Biofilms also have clinical importance. They form on medical devices, resist routine cleaning and sterilization, and cause health-acquired infections. Within the body, biofilms form on the teeth as plaque, in the lungs of patients with cystic fibrosis, and on the cardiac tissue of patients with endocarditis. The slime layer helps protect the cells from host immune defenses and antibiotic treatments.

Studying biofilms requires new approaches. Because of the cells' adhesion properties, many of the methods for culturing and counting cells that are explored in this chapter are not easily applied to biofilms. This is the beginning of a new era of challenges and rewarding insight into the ways that microorganisms grow and thrive in nature.

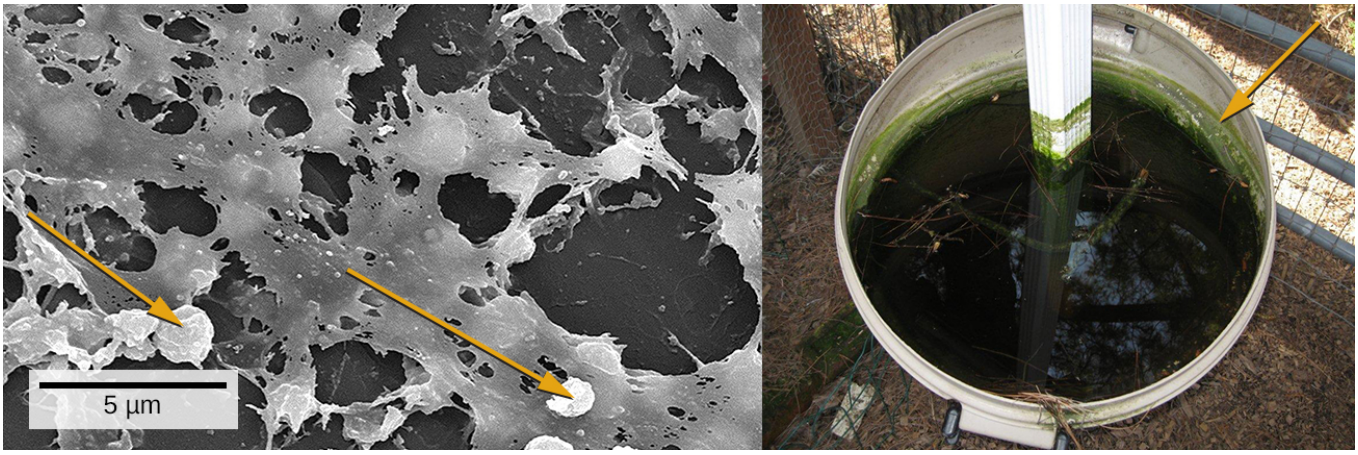


Figure **11.1**: Medical devices that are inserted into a patient's body often become contaminated with a thin biofilm of microorganisms enmeshed in the sticky material they secrete. The electron micrograph (left) shows the inside walls of an indwelling catheter. Arrows point to the round cells of *Staphylococcus aureus* bacteria attached to the layers of extracellular substrate. The garbage can (right) served as a rain collector. The arrow points to a green biofilm on the sides of the container. (credit left: modification of work by Centers for Disease Control and Prevention; credit right: modification of work by NASA)

11.1: How Microbes Grow

11.2: Oxygen Requirements for Microbial Growth

11.3: The Effects of pH on Microbial Growth

11.4: Temperature and Microbial Growth

11.5: Other Environmental Conditions that Affect Growth

11.6: Media Used for Bacterial Growth

11.E: Microbial Growth (Exercises)

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11.1: How Microbes Grow

Learning Objectives

- Describe the process of binary fission and identify the key steps
- Define generation time and explain the significance in microbial growth
- Calculate the number of bacterial cells after a given number of generations using generation time

The bacterial cell cycle involves the formation of new cells through the replication of DNA and partitioning of cellular components into two daughter cells. In prokaryotes, reproduction is always asexual, although extensive genetic recombination in the form of horizontal gene transfer takes place, as will be explored in a different chapter. Most bacteria have a single circular chromosome; however, some exceptions exist. For example, *Borrelia burgdorferi*, the causative agent of Lyme disease, has a linear chromosome.

Binary Fission

The most common mechanism of cell replication in bacteria is a process called **binary fission**, which is depicted in Figure 11.1.1. Before dividing, the cell grows and increases its number of cellular components. Next, the replication of DNA starts at a location on the circular chromosome called the origin of replication, where the chromosome is attached to the inner cell membrane. Replication continues in opposite directions along the chromosome until the terminus is reached.

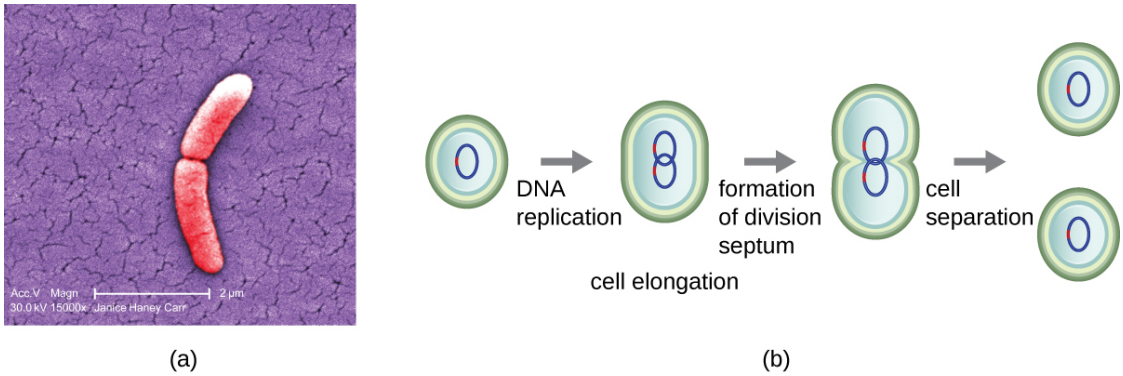


Figure 11.1.1: (a) The electron micrograph depicts two cells of *Salmonella typhimurium* after a binary fission event. (b) Binary fission in bacteria starts with the replication of DNA as the cell elongates. A division septum forms in the center of the cell. Two daughter cells of similar size form and separate, each receiving a copy of the original chromosome. (credit a: modification of work by Centers for Disease Control and Prevention).

The center of the enlarged cell constricts until two daughter cells are formed, each offspring receiving a complete copy of the parental genome and a division of the cytoplasm (cytokinesis). This process of cytokinesis and cell division is directed by a protein called **FtsZ**. FtsZ assembles into a Z ring on the cytoplasmic membrane (Figure 11.1.2). The Z ring is anchored by FtsZ-binding proteins and defines the division plane between the two daughter cells. Additional proteins required for cell division are added to the Z ring to form a structure called the **divisome**. The divisome activates to produce a peptidoglycan cell wall and build a **septum** that divides the two daughter cells. The daughter cells are separated by the division septum, where all of the cells' outer layers (the cell wall and outer membranes, if present) must be remodeled to complete division. For example, we know that specific enzymes break bonds between the monomers in peptidoglycans and allow addition of new subunits along the division septum.

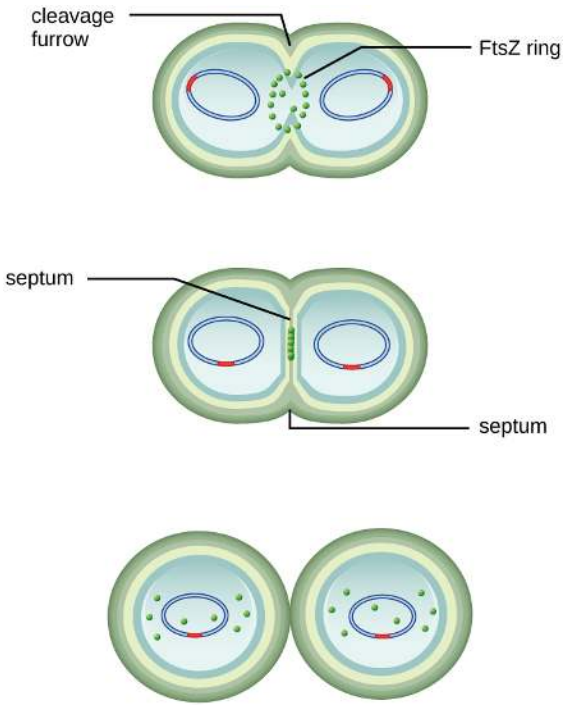


Figure **11.1.2**: FtsZ proteins assemble to form a Z ring that is anchored to the plasma membrane. The Z ring pinches the cell envelope to separate the cytoplasm of the new cells.

Generation Time

In eukaryotic organisms, the generation time is the time between the same points of the life cycle in two successive generations. For example, the typical generation time for the human population is 25 years. This definition is not practical for bacteria, which may reproduce rapidly or remain dormant for thousands of years. In prokaryotes (Bacteria and Archaea), the **generation time** is also called the **doubling time** and is defined as the time it takes for the population to double through one round of binary fission. Bacterial doubling times vary enormously. Whereas *Escherichia coli* can double in as little as 20 minutes under optimal growth conditions in the laboratory, bacteria of the same species may need several days to double in especially harsh environments. Most pathogens grow rapidly, like *E. coli*, but there are exceptions. For example, *Mycobacterium tuberculosis*, the causative agent of tuberculosis, has a generation time of between 15 and 20 hours. On the other hand, *M. leprae*, which causes Hansen's disease (leprosy), grows much more slowly, with a doubling time of 14 days.

Calculating Number of Cells

It is possible to predict the number of cells in a population when they divide by binary fission at a constant rate. As an example, consider what happens if a single cell divides every 30 minutes for 24 hours. The diagram in Figure **11.1.3** shows the increase in cell numbers for the first three generations.

The number of cells increases exponentially and can be expressed as 2^n, where n is the number of generations. If cells divide every 30 minutes, after 24 hours, 48 divisions would have taken place. If we apply the formula 2^n, where n is equal to 48, the single cell would give rise to 2^{48} or 281,474,976,710,656 cells at 48 generations (24 hours). When dealing with such huge numbers, it is more practical to use scientific notation. Therefore, we express the number of cells as 2.8×10^{14} cells.

In our example, we used one cell as the initial number of cells. For any number of starting cells, the formula is adapted as follows:

$$N_n = N_0 2^n \tag{11.1.1}$$

N_n is the number of cells at any generation n, N_0 is the initial number of cells, and n is the number of generations.

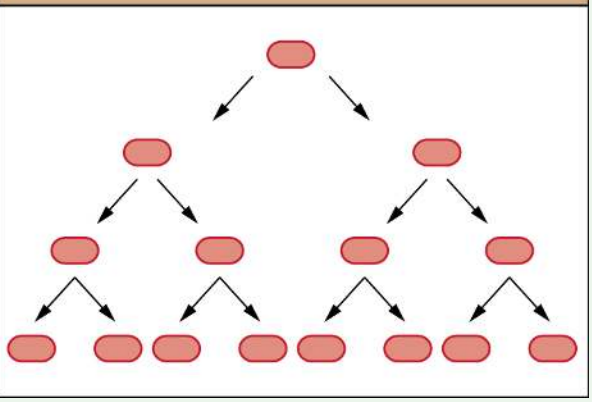
Number of generations (*n*)	Number of cells	Each division adds two new cells
0	1	
1	2	
2	4	
3	8	

Figure **11.1.3**: The parental cell divides and gives rise to two daughter cells. Each of the daughter cells, in turn, divides, giving a total of four cells in the second generation and eight cells in the third generation. Each division doubles the number of cells.

Key Concepts and Summary

- Most bacterial cells divide by **binary fission**. **Generation time** in bacterial growth is defined as the **doubling time**of the population.

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11.2: Oxygen Requirements for Microbial Growth

Learning Objectives

- Interpret visual data demonstrating minimum, optimum, and maximum oxygen or carbon dioxide requirements for growth
- Identify and describe different categories of microbes with requirements for growth with or without oxygen: obligate aerobe, obligate anaerobe, facultative anaerobe, aerotolerant anaerobe, microaerophile, and capnophile
- Give examples of microorganisms for each category of growth requirements

Ask most people "What are the major requirements for life?" and the answers are likely to include water and oxygen. Few would argue about the need for water, but what about oxygen? Can there be life without oxygen?

The answer is that molecular oxygen (O_2) is not always needed. The earliest signs of life are dated to a period when conditions on earth were highly reducing and free oxygen gas was essentially nonexistent. Only after cyanobacteria started releasing oxygen as a byproduct of photosynthesis and the capacity of iron in the oceans for taking up oxygen was exhausted did oxygen levels increase in the atmosphere. This event, often referred to as the Great Oxygenation Event or the Oxygen Revolution, caused a massive extinction. Most organisms could not survive the powerful oxidative properties of **reactive oxygen species (ROS)**, highly unstable ions and molecules derived from partial reduction of oxygen that can damage virtually any macromolecule or structure with which they come in contact. Singlet oxygen (O_2•), superoxide (O_2^-), peroxides (H_2O_2), hydroxyl radical (OH•), and hypochlorite ion (OCl^-), the active ingredient of household bleach, are all examples of ROS. The organisms that were able to detoxify reactive oxygen species harnessed the high electronegativity of oxygen to produce free energy for their metabolism and thrived in the new environment.

Oxygen Requirements of Microorganisms

Many ecosystems are still free of molecular oxygen. Some are found in extreme locations, such as deep in the ocean or in earth's crust; others are part of our everyday landscape, such as marshes, bogs, and sewers. Within the bodies of humans and other animals, regions with little or no oxygen provide an anaerobic environment for microorganisms. (Figure 11.2.1).



(a) (b)

Figure 11.2.1: Anaerobic environments are still common on earth. They include environments like (a) a bog where undisturbed dense sediments are virtually devoid of oxygen, and (b) the rumen (the first compartment of a cow's stomach), which provides an oxygen-free incubator for methanogens and other obligate anaerobic bacteria. (credit a: modification of work by National Park Service; credit b: modification of work by US Department of Agriculture)

We can easily observe different requirements for molecular oxygen by growing bacteria in **thioglycolate tube cultures**. A test-tube culture starts with autoclaved **thioglycolate medium** containing a low percentage of agar to allow motile bacteria to move throughout the medium. Thioglycolate has strong reducing properties and autoclaving flushes out most of the oxygen. The tubes are inoculated with the bacterial cultures to be tested and incubated at an appropriate temperature. Over time, oxygen slowly diffuses throughout the thioglycolate tube culture from the top. Bacterial density increases in the area where oxygen concentration is best suited for the growth of that particular organism.

The growth of bacteria with varying oxygen requirements in thioglycolate tubes is illustrated in Figure 11.2.2. In tube A, all the growth is seen at the top of the tube. The bacteria are **obligate (strict) aerobes** that cannot grow without an abundant supply of

oxygen. Tube B looks like the opposite of tube A. Bacteria grow at the bottom of tube B. Those are **obligate anaerobes**, which are killed by oxygen. Tube C shows heavy growth at the top of the tube and growth throughout the tube, a typical result with **facultative anaerobes**. Facultative anaerobes are organisms that thrive in the presence of oxygen but also grow in its absence by relying on fermentation or anaerobic respiration, if there is a suitable electron acceptor other than oxygen and the organism is able to perform anaerobic respiration. The **aerotolerant anaerobes** in tube D are indifferent to the presence of oxygen. They do not use oxygen because they usually have a fermentative metabolism, but they are not harmed by the presence of oxygen as obligate anaerobes are. Tube E on the right shows a "Goldilocks" culture. The oxygen level has to be just right for growth, not too much and not too little. These **microaerophiles** are bacteria that require a minimum level of oxygen for growth, about 1%–10%, well below the 21% found in the atmosphere.

Examples of Oxygen Categories

Examples of **obligate aerobes** are *Mycobacterium tuberculosis*, the causative agent of tuberculosis and *Micrococcus luteus*, a gram-positive bacterium that colonizes the skin. *Neisseria meningitidis*, the causative agent of severe bacterial meningitis, and *N. gonorrheae*, the causative agent of sexually transmitted gonorrhea, are also obligate aerobes.

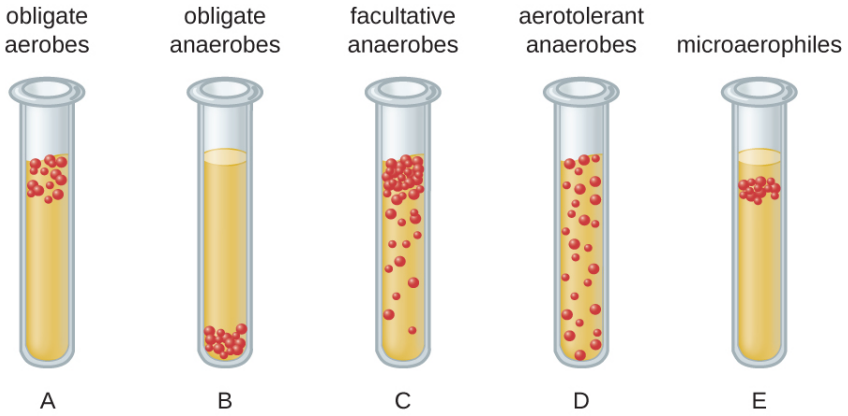
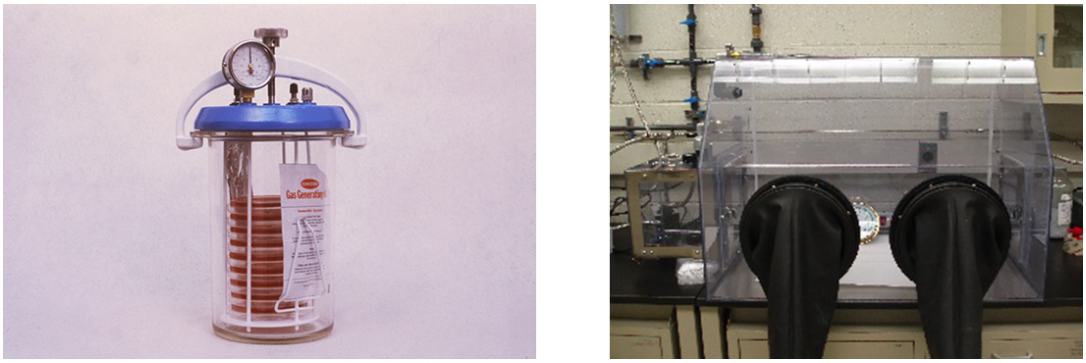


Figure **11.2.2**: Diagram of bacterial cell distribution in thioglycolate tubes.

Many **obligate anaerobes** are found in the environment where anaerobic conditions exist, such as in deep sediments of soil, still waters, and at the bottom of the deep ocean where there is no photosynthetic life. Anaerobic conditions also exist naturally in the intestinal tract of animals. Obligate anaerobes, mainly *Bacteroidetes*, represent a large fraction of the microbes in the human gut. Transient anaerobic conditions exist when tissues are not supplied with blood circulation; they die and become an ideal breeding ground for obligate anaerobes. Another type of obligate anaerobe encountered in the human body is the gram-positive, rod-shaped *Clostridium* spp. Their ability to form endospores allows them to survive in the presence of oxygen. One of the major causes of health-acquired infections is *C. difficile*, known as C. diff. Prolonged use of antibiotics for other infections increases the probability of a patient developing a secondary *C. difficile* infection. Antibiotic treatment disrupts the balance of microorganisms in the intestine and allows the colonization of the gut by *C. difficile*, causing a significant inflammation of the colon.

Other clostridia responsible for serious infections include *C. tetani*, the agent of tetanus, and *C. perfringens*, which causes gas gangrene. In both cases, the infection starts in necrotic tissue (dead tissue that is not supplied with oxygen by blood circulation). This is the reason that deep puncture wounds are associated with tetanus. When tissue death is accompanied by lack of circulation, gangrene is always a danger.

The study of obligate anaerobes requires special equipment. Obligate anaerobic bacteria must be grown under conditions devoid of oxygen. The most common approach is culture in an anaerobic jar (Figure **11.2.3**). Anaerobic jars include chemical packs that remove oxygen and release carbon dioxide (CO_2). An anaerobic chamber is an enclosed box from which all oxygen is removed. Gloves sealed to openings in the box allow handling of the cultures without exposing the culture to air (Figure **11.2.3**).



(a) (b)

Figure **11.2.3**: (a) An anaerobic jar is pictured that is holding nine Petri plates supporting cultures. (b) Openings in the side of an anaerobic box are sealed by glove-like sleeves that allow for the handling of cultures inside the box. (credit a: modification of work by Centers for Disease Control and Prevention; credit b: modification of work by NIST)

Staphylococci and Enterobacteriaceae are examples of **facultative anaerobes**. Staphylococci are found on the skin and upper respiratory tract. Enterobacteriaceae are found primarily in the gut and upper respiratory tract but can sometimes spread to the urinary tract, where they are capable of causing infections. It is not unusual to see mixed bacterial infections in which the facultative anaerobes use up the oxygen, creating an environment for the obligate anaerobes to flourish.

Examples of **aerotolerant anaerobes** include lactobacilli and streptococci, both found in the oral microbiota. *Campylobacter jejuni*, which causes gastrointestinal infections, is an example of a **microaerophile** and is grown under low-oxygen conditions.

The **optimum oxygen concentration**, as the name implies, is the ideal concentration of oxygen for a particular microorganism. The lowest concentration of oxygen that allows growth is called the **minimum permissive oxygen concentration**. The highest tolerated concentration of oxygen is the **maximum permissive oxygen concentration**. The organism will not grow outside the range of oxygen levels found between the minimum and maximum permissive oxygen concentrations.

An Unwelcome Anaerobe

Charles is a retired bus driver who developed type 2 diabetes over 10 years ago. Since his retirement, his lifestyle has become very sedentary and he has put on a substantial amount of weight. Although he has felt tingling and numbness in his left foot for a while, he has not been worried because he thought his foot was simply “falling asleep.” Recently, a scratch on his foot does not seem to be healing and is becoming increasingly ugly. Because the sore did not bother him much, Charles figured it could not be serious until his daughter noticed a purplish discoloration spreading on the skin and oozing (Figure **11.2.4**). When he was finally seen by his physician, Charles was rushed to the operating room. His open sore, or ulcer, is the result of a diabetic foot.

The concern here is that gas gangrene may have taken hold in the dead tissue. The most likely agent of gas gangrene is *Clostridium perfringens*, an endospore-forming, gram-positive bacterium. It is an obligate anaerobe that grows in tissue devoid of oxygen. Since dead tissue is no longer supplied with oxygen by the circulatory system, the dead tissue provides pockets of ideal environment for the growth of *C. perfringens*.

A surgeon examines the ulcer and radiographs of Charles’s foot and determines that the bone is not yet infected. The wound will have to be surgically debrided (debridement refers to the removal of dead and infected tissue) and a sample sent for microbiological lab analysis, but Charles will not have to have his foot amputated. Many diabetic patients are not so lucky. In 2008, nearly 70,000 diabetic patients in the United States lost a foot or limb to amputation, according to statistics from the Centers for Disease Control and Prevention.



Figure 11.2.4: This clinical photo depicts ulcers on the foot of a diabetic patient. Dead tissue accumulating in ulcers can provide an ideal growth environment for the anaerobe *C. perfringens*, a causative agent of gas gangrene. (credit: Shigeo Kono, Reiko Nakagawachi, Jun Arata, Benjamin A Lipsky)

Exercise 11.2.2

Which growth conditions would you recommend for the detection of *C. perfringens*?

Detoxification of Reactive Oxygen Species

Aerobic respiration constantly generates reactive oxygen species (ROS), byproducts that must be detoxified. Even organisms that do not use aerobic respiration need some way to break down some of the ROS that may form from atmospheric oxygen. Three main enzymes break down those toxic byproducts: superoxide dismutase, peroxidase, and catalase. Each one catalyzes a different reaction. Reactions of type seen in Reaction 1 are catalyzed by **peroxidases**.

$$\mathrm{X-(2H^+)+H_2O_2 \rightarrow oxidized\text{-}X+2H_2O} \tag{11.2.1}$$

In these reactions, an electron donor (reduced compound; e.g., reduced nicotinamide adenine dinucleotide [NADH]) oxidizes hydrogen peroxide, or other peroxides, to water. The enzymes play an important role by limiting the damage caused by peroxidation of membrane lipids. Reaction 2 is mediated by the enzyme **superoxide dismutase (SOD)** and breaks down the powerful superoxide anions generated by aerobic metabolism:

$$\mathrm{2O_2^- + 2H^+ \rightarrow H_2O_2 + O_2} \tag{11.2.2}$$

The enzyme **catalase** converts hydrogen peroxide to water and oxygen as shown in Reaction 3.

$$\mathrm{2H_2O_2 \rightarrow 2H_2O + O_2} \tag{11.2.3}$$

Obligate anaerobes usually lack all three enzymes. Aerotolerant anaerobes do have SOD but no catalase. Reaction 3, shown occurring in Figure 11.2.5, is the basis of a useful and rapid test to distinguish streptococci, which are aerotolerant and do not possess catalase, from staphylococci, which are facultative anaerobes. A sample of culture rapidly mixed in a drop of 3% hydrogen peroxide will release bubbles if the culture is catalase positive.

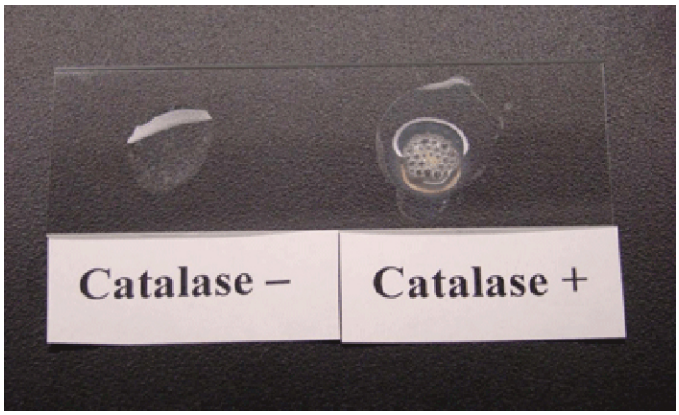


Figure **11.2.5**: The catalase test detects the presence of the enzyme catalase by noting whether bubbles are released when hydrogen peroxide is added to a culture sample. Compare the positive result (right) with the negative result (left). (credit: Centers for Disease Control and Prevention)

Bacteria that grow best in a higher concentration of CO_2 and a lower concentration of oxygen than present in the atmosphere are called capnophiles. One common approach to grow **capnophiles** is to use a candle jar. A **candle jar** consists of a jar with a tight-fitting lid that can accommodate the cultures and a candle. After the cultures are added to the jar, the candle is lit and the lid closed. As the candle burns, it consumes most of the oxygen present and releases CO_2.

Key Concepts and Summary

- Aerobic and anaerobic environments can be found in diverse niches throughout nature, including different sites within and on the human body.
- Microorganisms vary in their requirements for molecular oxygen. **Obligate aerobes** depend on aerobic respiration and use oxygen as a terminal electron acceptor. They cannot grow without oxygen.
- **Obligate anaerobes** cannot grow in the presence of oxygen. They depend on fermentation and anaerobic respiration using a final electron acceptor other than oxygen.
- **Facultative anaerobes** show better growth in the presence of oxygen but will also grow without it.
- Although **aerotolerant anaerobes** do not perform aerobic respiration, they can grow in the presence of oxygen. Most aerotolerant anaerobes test negative for the enzyme **catalase**.
- **Microaerophiles** need oxygen to grow, albeit at a lower concentration than 21% oxygen in air.
- **Optimum oxygen concentration** for an organism is the oxygen level that promotes the fastest growth rate. The **minimum permissive oxygen concentration** and the **maximum permissive oxygen concentration** are, respectively, the lowest and the highest oxygen levels that the organism will tolerate.
- **Peroxidase**, **superoxide dismutase**, and **catalase** are the main enzymes involved in the detoxification of the **reactive oxygen species**. Superoxide dismutase is usually present in a cell that can tolerate oxygen. All three enzymes are usually detectable in cells that perform aerobic respiration and produce more ROS.
- A **capnophile** is an organism that requires a higher than atmospheric concentration of CO_2 to grow.

Footnotes

1. 1 Centers for Disease Control and Prevention. “Living With Diabetes: Keep Your Feet Healthy.” http://www.cdc.gov/Features/DiabetesFootHealth/

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11.3: The Effects of pH on Microbial Growth

Learning Objectives

- Illustrate and briefly describe minimum, optimum, and maximum pH requirements for growth
- Identify and describe the different categories of microbes with pH requirements for growth: acidophiles, neutrophiles, and alkaliphiles
- Give examples of microorganisms for each category of pH requirement

Yogurt, pickles, sauerkraut, and lime-seasoned dishes all owe their tangy taste to a high acid content (Figure **11.3.1**). Recall that acidity is a function of the concentration of hydrogen ions [H^+] and is measured as pH. Environments with pH values below 7.0 are considered acidic, whereas those with pH values above 7.0 are considered basic. Extreme pH affects the structure of all macromolecules. The hydrogen bonds holding together strands of DNA break up at high pH. Lipids are hydrolyzed by an extremely basic pH. The proton motive force responsible for production of ATP in cellular respiration depends on the concentration gradient of H^+ across the plasma membrane (see Cellular Respiration). If H^+ ions are neutralized by hydroxide ions, the concentration gradient collapses and impairs energy production. But the component most sensitive to pH in the cell is its workhorse, the protein. Moderate changes in pH modify the ionization of amino-acid functional groups and disrupt hydrogen bonding, which, in turn, promotes changes in the folding of the molecule, promoting denaturation and destroying activity.



Figure **11.3.1**: Lactic acid bacteria that ferment milk into yogurt or transform vegetables in pickles thrive at a pH close to 4.0. Sauerkraut and dishes such as pico de gallo owe their tangy flavor to their acidity. Acidic foods have been a mainstay of the human diet for centuries, partly because most microbes that cause food spoilage grow best at a near neutral pH and do not tolerate acidity well. (credit “yogurt”: modification of work by “nina.jsc”/Flickr; credit “pickles”: modification of work by Noah Sussman; credit “sauerkraut”: modification of work by Jesse LaBuff; credit “pico de gallo”: modification of work by “regan76”/Flickr)

The **optimum growth pH** is the most favorable pH for the growth of an organism. The lowest pH value that an organism can tolerate is called the **minimum growth pH** and the highest pH is the **maximum growth pH**. These values can cover a wide range, which is important for the preservation of food and to microorganisms’ survival in the stomach. For example, the optimum growth pH of *Salmonella* spp. is 7.0–7.5, but the minimum growth pH is closer to 4.2.

Most bacteria are **neutrophiles**, meaning they grow optimally at a pH within one or two pH units of the neutral pH of 7 (see Figure **11.3.2**). Most familiar bacteria, like *Escherichia coli*, staphylococci, and *Salmonella* spp. are neutrophiles and do not fare well in the acidic pH of the stomach. However, there are pathogenic strains of *E. coli*, *S. typhi*, and other species of intestinal pathogens that are much more resistant to stomach acid. In comparison, fungi thrive at slightly acidic pH values of 5.0–6.0.

Microorganisms that grow optimally at pH less than 5.55 are called **acidophiles**. For example, the sulfur-oxidizing *Sulfolobus* spp. isolated from sulfur mud fields and hot springs in Yellowstone National Park are extreme acidophiles. These archaea survive at pH values of 2.5–3.5. Species of the archaean genus *Ferroplasma* live in acid mine drainage at pH values of 0–2.9. *Lactobacillus* bacteria, which are an important part of the normal microbiota of the vagina, can tolerate acidic environments at pH values 3.5–6.8 and also contribute to the acidity of the vagina (pH of 4, except at the onset of menstruation) through their metabolic production of lactic acid. The vagina’s acidity plays an important role in inhibiting other microbes that are less tolerant of acidity. Acidophilic microorganisms display a number of adaptations to survive in strong acidic environments. For example, proteins show increased negative surface charge that stabilizes them at low pH. Pumps actively eject H^+ ions out of the cells. The changes in the composition of membrane phospholipids probably reflect the need to maintain membrane fluidity at low pH.

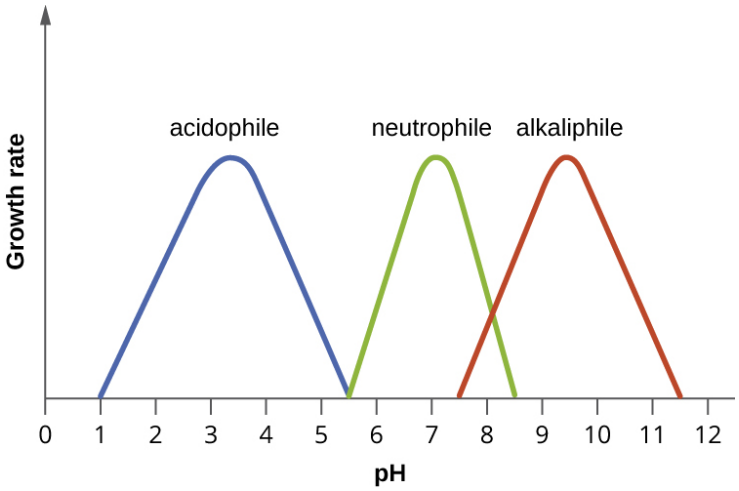


Figure 11.3.2: The curves show the approximate pH ranges for the growth of the different classes of pH-specific prokaryotes. Each curve has an optimal pH and extreme pH values at which growth is much reduced. Most bacteria are neutrophiles and grow best at near-neutral pH (center curve). Acidophiles have optimal growth at pH values near 3 and alkaliphiles have optimal growth at pH values above 9.

At the other end of the spectrum are **alkaliphiles**, microorganisms that grow best at pH between 8.0 and 10.5. *Vibrio cholerae*, the pathogenic agent of cholera, grows best at the slightly basic pH of 8.0; it can survive pH values of 11.0 but is inactivated by the acid of the stomach. When it comes to survival at high pH, the bright pink archaean *Natronobacterium*, found in the soda lakes of the African Rift Valley, may hold the record at a pH of 10.5 (Figure 11.3.3). Extreme alkaliphiles have adapted to their harsh environment through evolutionary modification of lipid and protein structure and compensatory mechanisms to maintain the proton motive force in an alkaline environment. For example, the alkaliphile *Bacillus firmus* derives the energy for transport reactions and motility from a Na^+ ion gradient rather than a proton motive force. Many enzymes from alkaliphiles have a higher isoelectric point, due to an increase in the number of basic amino acids, than homologous enzymes from neutrophiles.



Figure 11.3.3: View from space of Lake Natron in Tanzania. The pink color is due to the pigmentation of the extreme alkaliphilic and halophilic microbes that colonize the lake. (credit: NASA)

Survival at the Low pH of the Stomach

Peptic ulcers (or stomach ulcers) are painful sores on the stomach lining. Until the 1980s, they were believed to be caused by spicy foods, stress, or a combination of both. Patients were typically advised to eat bland foods, take anti-acid medications, and avoid stress. These remedies were not particularly effective, and the condition often recurred. This all changed dramatically when the real cause of most peptic ulcers was discovered to be a slim, corkscrew-shaped bacterium, *Helicobacter pylori*. This organism was identified and isolated by Barry Marshall and Robin Warren, whose discovery earned them the Nobel Prize in Medicine in 2005.

The ability of *H. pylori* to survive the low pH of the stomach would seem to suggest that it is an extreme acidophile. As it turns out, this is not the case. In fact, *H. pylori* is a neutrophile. So, how does it survive in the stomach? Remarkably, *H. pylori* creates a microenvironment in which the pH is nearly neutral. It achieves this by producing large amounts of the enzyme urease, which breaks down urea to form NH_4^+ and CO_2. The ammonium ion raises the pH of the immediate environment.

This metabolic capability of *H. pylori* is the basis of an accurate, noninvasive test for infection. The patient is given a solution of urea containing radioactively labeled carbon atoms. If *H. pylori* is present in the stomach, it will rapidly break down the urea, producing radioactive CO_2 that can be detected in the patient's breath. Because peptic ulcers may lead to gastric cancer, patients who are determined to have *H. pylori* infections are treated with antibiotics.

Key Concepts and Summary

- Bacteria are generally **neutrophiles**. They grow best at neutral pH close to 7.0.
- **Acidophiles** grow optimally at a pH near 3.0. **Alkaliphiles** are organisms that grow optimally between a pH of 8 and 10.5. Extreme acidophiles and alkaliphiles grow slowly or not at all near neutral pH.
- Microorganisms grow best at their **optimum growth pH**. Growth occurs slowly or not at all below the **minimum growth pH** and above the **maximum growth pH**.

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11.4: Temperature and Microbial Growth

Learning Objectives

- Illustrate and briefly describe minimum, optimum, and maximum temperature requirements for growth
- Identify and describe different categories of microbes with temperature requirements for growth: psychrophile, psychrotrophs, mesophile, thermophile, hyperthermophile
- Give examples of microorganisms in each category of temperature tolerance

When the exploration of Lake Whillans started in Antarctica, researchers did not expect to find much life. Constant subzero temperatures and lack of obvious sources of nutrients did not seem to be conditions that would support a thriving ecosystem. To their surprise, the samples retrieved from the lake showed abundant microbial life. In a different but equally harsh setting, bacteria grow at the bottom of the ocean in sea vents (Figure **11.4.1**), where temperatures can reach 340 °C (700 °F).

Microbes can be roughly classified according to the range of temperature at which they can grow. The growth rates are the highest at the **optimum growth temperature** for the organism. The lowest temperature at which the organism can survive and replicate is its **minimum growth temperature**. The highest temperature at which growth can occur is its **maximum growth temperature**. The following ranges of permissive growth temperatures are approximate only and can vary according to other environmental factors.

Organisms categorized as **mesophiles** (“middle loving”) are adapted to moderate temperatures, with optimal growth temperatures ranging from room temperature (about 20 °C) to about 45 °C. As would be expected from the core temperature of the human body, 37 °C (98.6 °F), normal human microbiota and pathogens (e.g., *E. coli*, *Salmonella* spp., and *Lactobacillus* spp.) are mesophiles.

Organisms called **psychrotrophs**, also known as psychrotolerant, prefer cooler environments, from a high temperature of 25 °C to refrigeration temperature about 4 °C. They are found in many natural environments in temperate climates. They are also responsible for the spoilage of refrigerated food.

The organisms retrieved from arctic lakes such as Lake Whillans are considered extreme **psychrophiles** (cold loving). Psychrophiles are microorganisms that can grow at 0 °C and below, have an optimum growth temperature close to 15 °C, and usually do not survive at temperatures above 20 °C. They are found in permanently cold environments such as the deep waters of the oceans. Because they are active at low temperature, psychrophiles and psychrotrophs are important decomposers in cold climates.

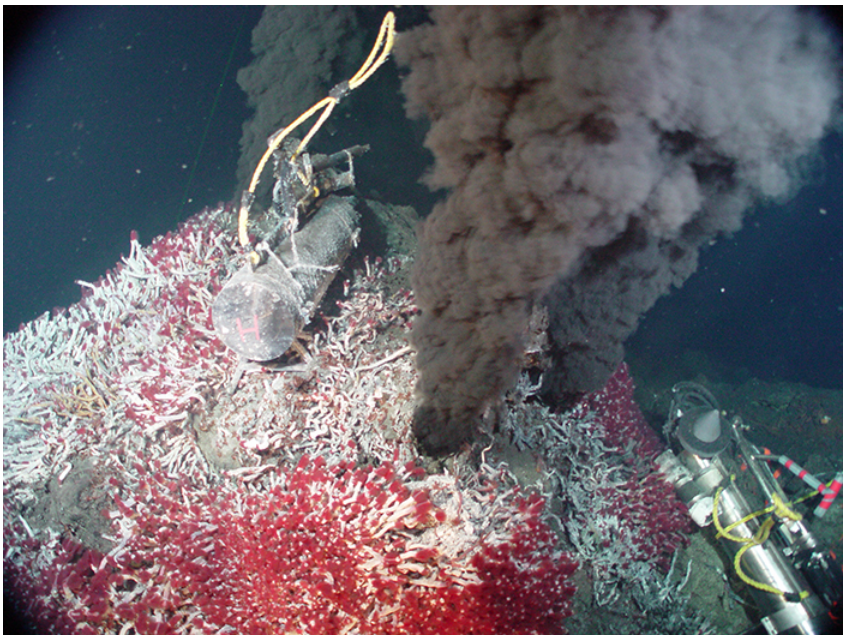


Figure 11.4.1: A black smoker at the bottom of the ocean belches hot, chemical-rich water, and heats the surrounding waters. Sea vents provide an extreme environment that is nonetheless teeming with macroscopic life (the red tubeworms) supported by an abundant microbial ecosystem. (credit: NOAA)

Organisms that grow at optimum temperatures of 50 °C to a maximum of 80 °C are called **thermophiles** (“heat loving”). They do not multiply at room temperature. Thermophiles are widely distributed in hot springs, geothermal soils, and manmade environments such as garden compost piles where the microbes break down kitchen scraps and vegetal material. Examples of thermophiles include *Thermus aquaticus* and *Geobacillus* spp. Higher up on the extreme temperature scale we find the **hyperthermophiles**, which are characterized by growth ranges from 80 °C to a maximum of 110 °C, with some extreme examples that survive temperatures above 121 °C, the average temperature of an autoclave. The hydrothermal vents at the bottom of the ocean are a prime example of extreme environments, with temperatures reaching an estimated 340 °C (Figure 11.4.1). Microbes isolated from the vents achieve optimal growth at temperatures higher than 100 °C. Noteworthy examples are *Pyrobolus* and *Pyrodictium*, archaea that grow at 105 °C and survive autoclaving. Figure 11.4.2 shows the typical skewed curves of temperature-dependent growth for the categories of microorganisms we have discussed.

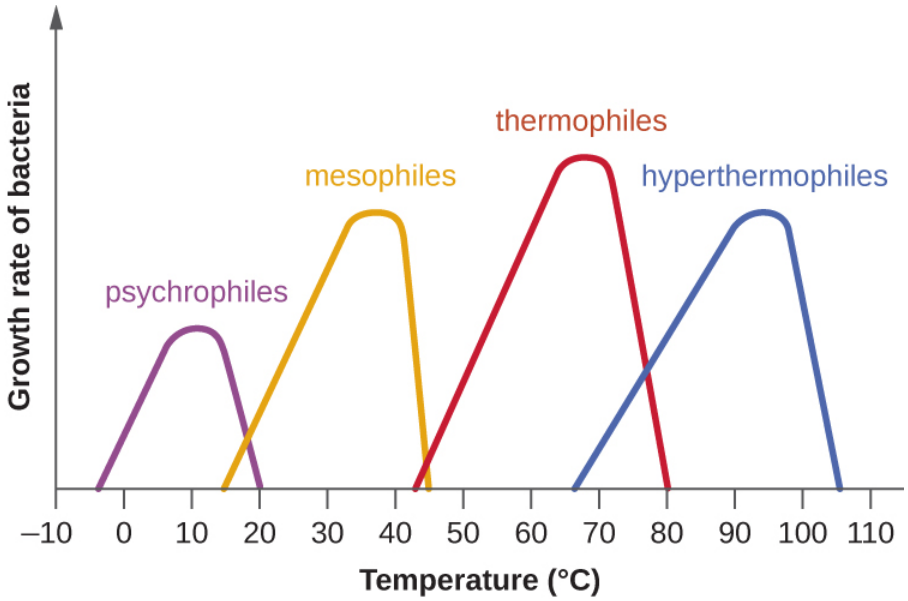


Figure 11.4.2: The graph shows growth rate of bacteria as a function of temperature. Notice that the curves are skewed toward the optimum temperature. The skewing of the growth curve is thought to reflect the rapid denaturation of proteins as the temperature rises past the optimum for growth of the microorganism.

Life in extreme environments raises fascinating questions about the adaptation of macromolecules and metabolic processes. Very low temperatures affect cells in many ways. Membranes lose their fluidity and are damaged by ice crystal formation. Chemical reactions and diffusion slow considerably. Proteins become too rigid to catalyze reactions and may undergo denaturation. At the opposite end of the temperature spectrum, heat denatures proteins and nucleic acids. Increased fluidity impairs metabolic processes in membranes. Some of the practical applications of the destructive effects of heat on microbes are sterilization by steam, pasteurization, and incineration of inoculating loops. Proteins in psychrophiles are, in general, rich in hydrophobic residues, display an increase in flexibility, and have a lower number of secondary stabilizing bonds when compared with homologous proteins from mesophiles. Antifreeze proteins and solutes that decrease the freezing temperature of the cytoplasm are common. The lipids in the membranes tend to be unsaturated to increase fluidity. Growth rates are much slower than those encountered at moderate temperatures. Under appropriate conditions, mesophiles and even thermophiles can survive freezing. Liquid cultures of bacteria are mixed with sterile glycerol solutions and frozen to −80 °C for long-term storage as stocks. Cultures can withstand freeze drying (lyophilization) and then be stored as powders in sealed ampules to be reconstituted with broth when needed.

Macromolecules in thermophiles and hyperthermophiles show some notable structural differences from what is observed in the mesophiles. The ratio of saturated to polyunsaturated lipids increases to limit the fluidity of the cell membranes. Their DNA sequences show a higher proportion of guanine–cytosine nitrogenous bases, which are held together by three hydrogen bonds in contrast to adenine and thymine, which are connected in the double helix by two hydrogen bonds. Additional secondary ionic and covalent bonds, as well as the replacement of key amino acids to stabilize folding, contribute to the resistance of proteins to denaturation. The so-called thermoenzymes purified from thermophiles have important practical applications. For example, amplification of nucleic acids in the polymerase chain reaction (PCR) depends on the thermal stability of *Taq* polymerase, an enzyme isolated from *T. aquaticus*. Degradation enzymes from thermophiles are added as ingredients in hot-water detergents, increasing their effectiveness.

Feeding the World…and the World's Algae

Artificial fertilizers have become an important tool in food production around the world. They are responsible for many of the gains of the so-called green revolution of the 20th century, which has allowed the planet to feed many of its more than 7 billion people. Artificial fertilizers provide nitrogen and phosphorus, key limiting nutrients, to crop plants, removing the normal barriers that would otherwise limit the rate of growth. Thus, fertilized crops grow much faster, and farms that use fertilizer produce higher crop yields.

However, careless use and overuse of artificial fertilizers have been demonstrated to have significant negative impacts on aquatic ecosystems, both freshwater and marine. Fertilizers that are applied at inappropriate times or in too-large quantities allow nitrogen and phosphorus compounds to escape use by crop plants and enter drainage systems. Inappropriate use of fertilizers in residential settings can also contribute to nutrient loads, which find their way to lakes and coastal marine ecosystems. As water warms and nutrients are plentiful, microscopic algae bloom, often changing the color of the water because of the high cell density.

Most algal blooms are not directly harmful to humans or wildlife; however, they can cause harm indirectly. As the algal population expands and then dies, it provides a large increase in organic matter to the bacteria that live in deep water. With this large supply of nutrients, the population of nonphotosynthetic microorganisms explodes, consuming available oxygen and creating "dead zones" where animal life has virtually disappeared.

Depletion of oxygen in the water is not the only damaging consequence of some algal blooms. The algae that produce red tides in the Gulf of Mexico, *Karenia brevis*, secrete potent toxins that can kill fish and other organisms and also accumulate in shellfish. Consumption of contaminated shellfish can cause severe neurological and gastrointestinal symptoms in humans. Shellfish beds must be regularly monitored for the presence of the toxins, and harvests are often shut down when it is present, incurring economic costs to the fishery. Cyanobacteria, which can form blooms in marine and freshwater ecosystems, produce toxins called microcystins, which can cause allergic reactions and liver damage when ingested in drinking water or during swimming. Recurring cyanobacterial algal blooms in Lake Erie (Figure **11.4.3**) have forced municipalities to issue drinking water bans for days at a time because of unacceptable toxin levels.

This is just a small sampling of the negative consequences of algal blooms, red tides, and dead zones. Yet the benefits of crop fertilizer—the main cause of such blooms—are difficult to dispute. There is no easy solution to this dilemma, as a ban on fertilizers is not politically or economically feasible. In lieu of this, we must advocate for responsible use and regulation in agricultural and residential contexts, as well as the restoration of wetlands, which can absorb excess fertilizers before they reach lakes and oceans.



Figure **11.4.3**: Heavy rains cause runoff of fertilizers into Lake Erie, triggering extensive algal blooms, which can be observed along the shoreline. Notice the brown unplanted and green planted agricultural land on the shore. (credit: NASA)

Link to Learning

This video discusses algal blooms and dead zones in more depth.

Key Concepts and Summary

- Microorganisms thrive at a wide range of temperatures; they have colonized different natural environments and have adapted to extreme temperatures. Both extreme cold and hot temperatures require evolutionary adjustments to macromolecules and biological processes.
- **Psychrophiles** grow best in the temperature range of 0–15 °C whereas **psychrotrophs** thrive between 4 °C and 25 °C.
- **Mesophiles** grow best at moderate temperatures in the range of 20 °C to about 45 °C. Pathogens are usually mesophiles.
- **Thermophiles** and **hyperthemophiles** are adapted to life at temperatures above 50 °C.
- Adaptations to cold and hot temperatures require changes in the composition of membrane lipids and proteins.

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11.5: Other Environmental Conditions that Affect Growth

Learning Objectives

- Identify and describe different categories of microbes with specific growth requirements other than oxygen, pH, and temperature, such as altered barometric pressure, osmotic pressure, humidity, and light
- Give at least one example microorganism for each category of growth requirement

Microorganisms interact with their environment along more dimensions than pH, temperature, and free oxygen levels, although these factors require significant adaptations. We also find microorganisms adapted to varying levels of salinity, barometric pressure, humidity, and light.

Osmotic and Barometric Pressure

Most natural environments tend to have lower solute concentrations than the cytoplasm of most microorganisms. Rigid cell walls protect the cells from bursting in a dilute environment. Not much protection is available against high osmotic pressure. In this case, water, following its concentration gradient, flows out of the cell. This results in plasmolysis (the shrinking of the protoplasm away from the intact cell wall) and cell death. This fact explains why brines and layering meat and fish in salt are time-honored methods of preserving food. Microorganisms called **halophiles** ("salt loving") actually require high salt concentrations for growth. These organisms are found in marine environments where salt concentrations hover at 3.5%. Extreme halophilic microorganisms, such as the red alga *Dunaliella salina* and the archaeal species *Halobacterium* in Figure **11.5.1**, grow in hypersaline lakes such as the Great Salt Lake, which is 3.5–8 times saltier than the ocean, and the Dead Sea, which is 10 times saltier than the ocean.

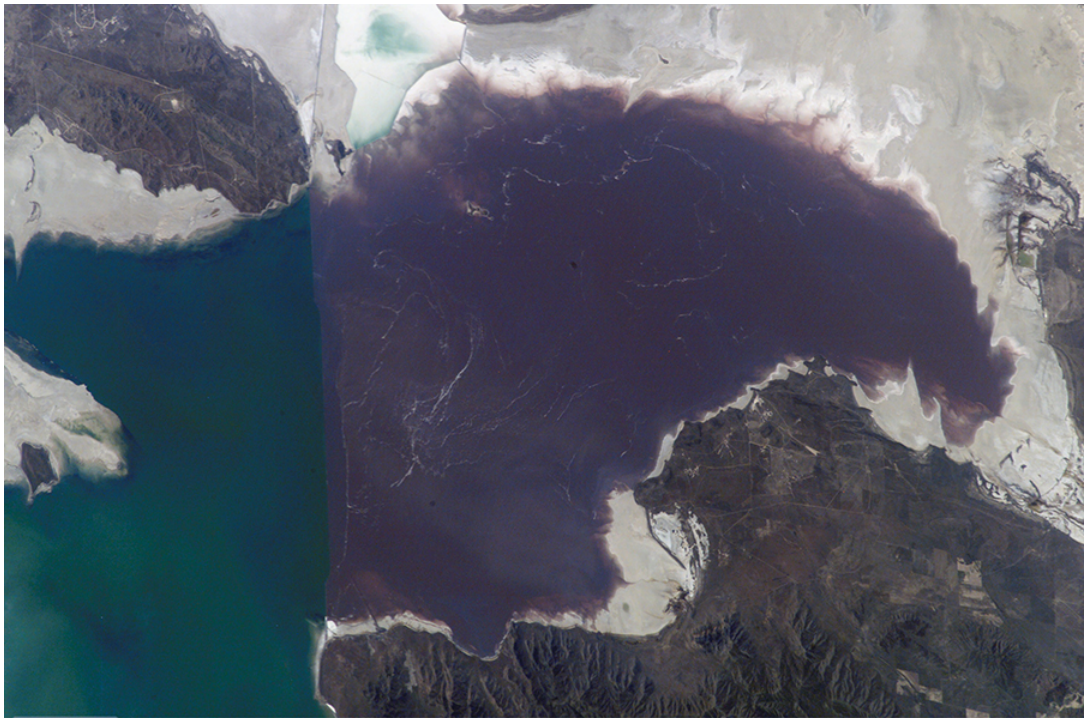


Figure **11.5.1**: Photograph taken from space of the Great Salt Lake in Utah. The purple color is caused by high density of the alga *Dunaliella* and the archaean *Halobacterium* spp. (credit: NASA)

Dunaliella spp. counters the tremendous osmotic pressure of the environment with a high cytoplasmic concentration of glycerol and by actively pumping out salt ions. *Halobacterium* spp. accumulates large concentrations of K^+ and other ions in its cytoplasm. Its proteins are designed for high salt concentrations and lose activity at salt concentrations below 1–2 M. Although most **halotolerant** organisms, for example *Halomonas* spp. in salt marshes, do not need high concentrations of salt for growth, they will survive and divide in the presence of high salt. Not surprisingly, the staphylococci, micrococci, and corynebacteria that colonize our skin tolerate salt in their environment. Halotolerant pathogens are an important cause of food-borne illnesses because they

survive and multiply in salty food. For example, the halotolerant bacteria *S. aureus*, *Bacillus cereus*, and *V. cholerae* produce dangerous enterotoxins and are major causes of food poisoning.

Microorganisms depend on available water to grow. Available moisture is measured as water activity (a_w), which is the ratio of the vapor pressure of the medium of interest to the vapor pressure of pure distilled water; therefore, the a_w of water is equal to 1.0. Bacteria require high a_w (0.97–0.99), whereas fungi can tolerate drier environments; for example, the range of a_w for growth of *Aspergillus* spp. is 0.8–0.75. Decreasing the water content of foods by drying, as in jerky, or through freeze-drying or by increasing osmotic pressure, as in brine and jams, are common methods of preventing spoilage.

Microorganisms that require high atmospheric pressure for growth are called barophiles. The bacteria that live at the bottom of the ocean must be able to withstand great pressures. Because it is difficult to retrieve intact specimens and reproduce such growth conditions in the laboratory, the characteristics of these microorganisms are largely unknown.

Light

Photoautotrophs, such as cyanobacteria or green sulfur bacteria, and photoheterotrophs, such as purple nonsulfur bacteria, depend on sufficient light intensity at the wavelengths absorbed by their pigments to grow and multiply. Energy from light is captured by pigments and converted into chemical energy that drives carbon fixation and other metabolic processes. The portion of the electromagnetic spectrum that is absorbed by these organisms is defined as photosynthetically active radiation (PAR). It lies within the visible light spectrum ranging from 400 to 700 nanometers (nm) and extends in the near infrared for some photosynthetic bacteria. A number of accessory pigments, such as fucoxanthin in brown algae and phycobilins in cyanobacteria, widen the useful range of wavelengths for photosynthesis and compensate for the low light levels available at greater depths of water. Other microorganisms, such as the archaea of the class Halobacteria, use light energy to drive their proton and sodium pumps. The light is absorbed by a pigment protein complex called bacteriorhodopsin, which is similar to the eye pigment rhodopsin. Photosynthetic bacteria are present not only in aquatic environments but also in soil and in symbiosis with fungi in lichens. The peculiar watermelon snow is caused by a microalga *Chlamydomonas nivalis*, a green alga rich in a secondary red carotenoid pigment (astaxanthin) which gives the pink hue to the snow where the alga grows.

Key Concepts and Summary

- **Halophiles** require high salt concentration in the medium, whereas **halotolerant** organisms can grow and multiply in the presence of high salt but do not require it for growth.
- Halotolerant pathogens are an important source of foodborne illnesses because they contaminate foods preserved in salt.
- Photosynthetic bacteria depend on visible light for energy.
- Most bacteria, with few exceptions, require high moisture to grow.

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11.6: Media Used for Bacterial Growth

Learning Objectives

- Identify and describe culture media for the growth of bacteria, including examples of all-purpose media, enriched, selective, differential, defined, and enrichment media

The study of microorganisms is greatly facilitated if we are able to culture them, that is, to keep reproducing populations alive under laboratory conditions. Culturing many microorganisms is challenging because of highly specific nutritional and environmental requirements and the diversity of these requirements among different species.

Nutritional Requirements

The number of available media to grow bacteria is considerable. Some media are considered **general all-purpose media** and support growth of a large variety of organisms. A prime example of an all-purpose medium is tryptic soy broth (TSB). **Specialized media** are used in the identification of bacteria and are supplemented with dyes, pH indicators, or antibiotics. One type, **enriched media**, contains growth factors, vitamins, and other essential nutrients to promote the growth of **fastidious organisms**, organisms that cannot make certain nutrients and require them to be added to the medium. When the complete chemical composition of a medium is known, it is called a **chemically defined medium**. For example, in EZ medium, all individual chemical components are identified and the exact amounts of each is known. In **complex media**, which contain extracts and digests of yeasts, meat, or plants, the precise chemical composition of the medium is not known. Amounts of individual components are undetermined and variable. Nutrient broth, tryptic soy broth, and brain heart infusion, are all examples of complex media.

Media that inhibit the growth of unwanted microorganisms and support the growth of the organism of interest by supplying nutrients and reducing competition are called **selective media**. An example of a selective medium is MacConkey agar. It contains bile salts and crystal violet, which interfere with the growth of many gram-positive bacteria and favor the growth of gram-negative bacteria, particularly the Enterobacteriaceae. These species are commonly named enterics, reside in the intestine, and are adapted to the presence of bile salts. The **enrichment cultures** foster the preferential growth of a desired microorganism that represents a fraction of the organisms present in an inoculum. For example, if we want to isolate bacteria that break down crude oil, hydrocarbonoclastic bacteria, sequential subculturing in a medium that supplies carbon only in the form of crude oil will enrich the cultures with oil-eating bacteria. The **differential media** make it easy to distinguish colonies of different bacteria by a change in the color of the colonies or the color of the medium. Color changes are the result of end products created by interaction of bacterial enzymes with differential substrates in the medium or, in the case of hemolytic reactions, the lysis of red blood cells in the medium. In Figure **11.6.1**, the differential fermentation of lactose can be observed on MacConkey agar. The lactose fermenters produce acid, which turns the medium and the colonies of strong fermenters hot pink. The medium is supplemented with the pH indicator neutral red, which turns to hot pink at low pH. Selective and differential media can be combined and play an important role in the identification of bacteria by biochemical methods.



Figure **11.6.1**: On this MacConkey agar plate, the lactose-fermenter *E. coli* colonies are bright pink. *Serratia marcescens*, which does not ferment lactose, forms a cream-colored streak on the tan medium. (credit: American Society for Microbiology)

Link to Learning

Compare the compositions of EZ medium and sheep blood agar.

The End-of-Year Picnic

The microbiology department is celebrating the end of the school year in May by holding its traditional picnic on the green. The speeches drag on for a couple of hours, but finally all the faculty and students can dig into the food: chicken salad, tomatoes, onions, salad, and custard pie. By evening, the whole department, except for two vegetarian students who did not eat the chicken salad, is stricken with nausea, vomiting, retching, and abdominal cramping. Several individuals complain of diarrhea. One patient shows signs of shock (low blood pressure). Blood and stool samples are collected from patients, and an analysis of all foods served at the meal is conducted.

Bacteria can cause gastroenteritis (inflammation of the stomach and intestinal tract) either by colonizing and replicating in the host, which is considered an infection, or by secreting toxins, which is considered intoxication. Signs and symptoms of infections are typically delayed, whereas intoxication manifests within hours, as happened after the picnic.

Blood samples from the patients showed no signs of bacterial infection, which further suggests that this was a case of intoxication. Since intoxication is due to secreted toxins, bacteria are not usually detected in blood or stool samples. MacConkey agar and sorbitol-MacConkey agar plates and xylose-lysine-deoxycholate (XLD) plates were inoculated with stool samples and did not reveal any unusually colored colonies, and no black colonies or white colonies were observed on XLD. All lactose fermenters on MacConkey agar also ferment sorbitol. These results ruled out common agents of food-borne illnesses: *E. coli*, *Salmonella* spp., and *Shigella* spp.

Analysis of the chicken salad revealed an abnormal number of gram-positive cocci arranged in clusters (Figure **11.6.2**). A culture of the gram-positive cocci releases bubbles when mixed with hydrogen peroxide. The culture turned mannitol salt agar yellow after a 24-hour incubation.

All the tests point to *Staphylococcus aureus* as the organism that secreted the toxin. Samples from the salad showed the presence of gram-positive cocci bacteria in clusters. The colonies were positive for catalase. The bacteria grew on mannitol salt agar fermenting mannitol, as shown by the change to yellow of the medium. The pH indicator in mannitol salt agar is phenol red, which turns to yellow when the medium is acidified by the products of fermentation.

The toxin secreted by *S. aureus* is known to cause severe gastroenteritis. The organism was probably introduced into the salad during preparation by the food handler and multiplied while the salad was kept in the warm ambient temperature during the

speeches.

Exercise 11.6.2

1. What are some other factors that might have contributed to rapid growth of *S. aureus* in the chicken salad?
2. Why would *S. aureus* not be inhibited by the presence of salt in the chicken salad?

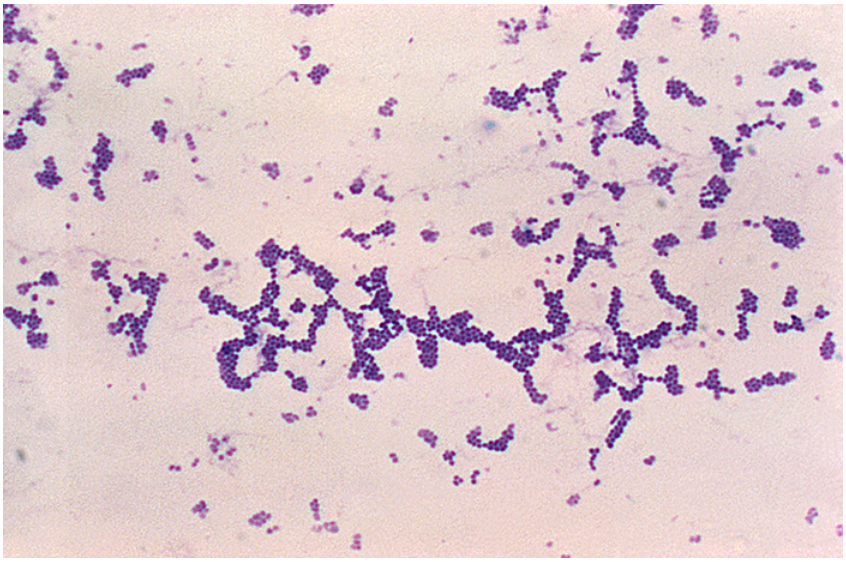


Figure **11.6.2**: Gram-positive cocci in clusters. (credit: Centers for Disease Control and Prevention)

Key Concepts and Summary

- **Chemically defined media** contain only chemically known components.
- **Selective media** favor the growth of some microorganisms while inhibiting others.
- **Enriched media** contain added essential nutrients a specific organism needs to grow
- **Differential media** help distinguish bacteria by the color of the colonies or the change in the medium.

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11.E: Microbial Growth (Exercises)

9.1: How Microbes Grow

The bacterial cell cycle involves the formation of new cells through the replication of DNA and partitioning of cellular components into two daughter cells. In prokaryotes, reproduction is always asexual, although extensive genetic recombination in the form of horizontal gene transfer takes place, as will be explored in a different chapter. Most bacteria have a single circular chromosome; however, some exceptions exist.

Multiple Choice

Which of the following methods would be used to measure the concentration of bacterial contamination in processed peanut butter?

A. turbidity measurement
B. total plate count
C. dry weight measurement
D. direct counting of bacteria on a calibrated slide under the microscope

Answer

B

In which phase would you expect to observe the most endospores in a *Bacillus* cell culture?

A. death phase
B. lag phase
C. log phase
D. log, lag, and death phases would all have roughly the same number of endospores.

Answer

A

During which phase would penicillin, an antibiotic that inhibits cell-wall synthesis, be most effective?

A. death phase
B. lag phase
C. log phase
D. stationary phase

Answer

C

Which of the following is the best definition of generation time in a bacterium?

A. the length of time it takes to reach the log phase
B. the length of time it takes for a population of cells to double
C. the time it takes to reach stationary phase
D. the length of time of the exponential phase

Answer

B

What is the function of the Z ring in binary fission?

A. It controls the replication of DNA.
B. It forms a contractile ring at the septum.
C. It separates the newly synthesized DNA molecules.

D. It mediates the addition of new peptidoglycan subunits.

Answer

B

If a culture starts with 50 cells, how many cells will be present after five generations with no cell death?

A. 200
B. 400
C. 1600
D. 3200

Answer

C

Filamentous cyanobacteria often divide by which of the following?

A. budding
B. mitosis
C. fragmentation
D. formation of endospores

Answer

C

Which is a reason for antimicrobial resistance being higher in a biofilm than in free-floating bacterial cells?

A. The EPS allows faster diffusion of chemicals in the biofilm.
B. Cells are more metabolically active at the base of a biofilm.
C. Cells are metabolically inactive at the base of a biofilm.
D. The structure of a biofilm favors the survival of antibiotic resistant cells.

Answer

C

Quorum sensing is used by bacterial cells to determine which of the following?

A. the size of the population
B. the availability of nutrients
C. the speed of water flow
D. the density of the population

Answer

D

Which of the following statements about autoinducers is incorrect?

A. They bind directly to DNA to activate transcription.
B. They can activate the cell that secreted them.
C. N-acylated homoserine lactones are autoinducers in gram-negative cells.
D. Autoinducers may stimulate the production of virulence factors.

Answer

A

Fill in the Blank

Direct count of total cells can be performed using a _____ or a _____.

Answer

hemocytometer, Petroff-Hausser counting chamber

The _____ method allows direct count of total cells growing on solid medium.

Answer

plate count

A statistical estimate of the number of live cells in a liquid is usually done by _____.

Answer

most probable number

For this indirect method of estimating the growth of a culture, you measure _____ using a spectrophotometer.

Answer

turbidity

Active growth of a culture may be estimated indirectly by measuring the following products of cell metabolism: _____ or _____.

Answer

ATP, acid from fermentation

Matching

Match the definition with the name of the growth phase in the growth curve.

___Number of dying cells is higher than the number of cells dividing	A. Lag phase
___Number of new cells equal to number of dying cells	B. Log phase
___New enzymes to use available nutrients are induced	C. Stationary phase
___Binary fission is occurring at maximum rate	D. Death phase

Answer

D, C, A, B

Short Answer

Why is it important to measure the transmission of light through a control tube with only broth in it when making turbidity measures of bacterial cultures?

In terms of counting cells, what does a plating method accomplish that an electronic cell counting method does not?

Order the following stages of the development of a biofilm from the earliest to the last step.

1. secretion of EPS
2. reversible attachment
3. dispersal
4. formation of water channels
5. irreversible attachment

Infections among hospitalized patients are often related to the presence of a medical device in the patient. Which conditions favor the formation of biofilms on in-dwelling catheters and prostheses?

Critical Thinking

A patient in the hospital has an intravenous catheter inserted to allow for the delivery of medications, fluids, and electrolytes. Four days after the catheter is inserted, the patient develops a fever and an infection in the skin around the catheter. Blood cultures reveal that the patient has a blood-borne infection. Tests in the clinical laboratory identify the blood-borne pathogen as *Staphylococcus epidermidis*, and antibiotic susceptibility tests are performed to provide doctors with essential information for selecting the best drug for treatment of the infection. Antibacterial chemotherapy is initiated and delivered through the intravenous catheter that was originally inserted into the patient. Within 7 days, the skin infection is gone, blood cultures are negative for *S. epidermidis*, and the antibacterial chemotherapy is discontinued. However, 2 days after discontinuing the antibacterial chemotherapy, the patient develops another fever and skin infection and the blood cultures are positive for the same strain of *S. epidermidis* that had been isolated the previous week. This time, doctors remove the intravenous catheter and administer oral antibiotics, which successfully treat both the skin and blood-borne infection caused by *S. epidermidis*. Furthermore, the infection does not return after discontinuing the oral antibacterial chemotherapy. What are some possible reasons why intravenous chemotherapy failed to completely cure the patient despite laboratory tests showing the bacterial strain was susceptible to the prescribed antibiotic? Why might the second round of antibiotic therapy have been more successful? Justify your answers.

Why are autoinducers small molecules?

Refer to Figure B.1 in Appendix B. If the results from a pond water sample were recorded as 3, 2, 1, what would be the MPN of bacteria in 100 mL of pond water?

Refer to Figure 9.1.14. Why does turbidity lose reliability at high cell concentrations when the culture reaches the stationary phase?

9.2: Oxygen Requirements for Microbial Growth

Ask most people “What are the major requirements for life?” and the answers are likely to include water and oxygen. Few would argue about the need for water, but what about oxygen? Can there be life without oxygen? The answer is that molecular oxygen is not always needed. The earliest signs of life are dated to a period when conditions on earth were highly reducing and free oxygen gas was essentially nonexistent.

Multiple Choice

An inoculated thioglycolate medium culture tube shows dense growth at the surface and turbidity throughout the rest of the tube. What is your conclusion?

A. The organisms die in the presence of oxygen
B. The organisms are facultative anaerobes.
C. The organisms should be grown in an anaerobic chamber.
D. The organisms are obligate aerobes.

Answer

B

An inoculated thioglycolate medium culture tube is clear throughout the tube except for dense growth at the bottom of the tube. What is your conclusion?

A. The organisms are obligate anaerobes.
B. The organisms are facultative anaerobes.
C. The organisms are aerotolerant.
D. The organisms are obligate aerobes.

Answer

A

Pseudomonas aeruginosa is a common pathogen that infects the airways of patients with cystic fibrosis. It does not grow in the absence of oxygen. The bacterium is probably which of the following?

A. an aerotolerant anaerobe
B. an obligate aerobe
C. an obligate anaerobe
D. a facultative anaerobe

Answer

B

Streptococcus mutans is a major cause of cavities. It resides in the gum pockets, does not have catalase activity, and can be grown outside of an anaerobic chamber. The bacterium is probably which of the following?

A. a facultative anaerobe
B. an obligate aerobe
C. an obligate anaerobe
D. an aerotolerant anaerobe

Answer

D

Why do the instructions for the growth of *Neisseria gonorrheae* recommend a CO_2-enriched atmosphere?

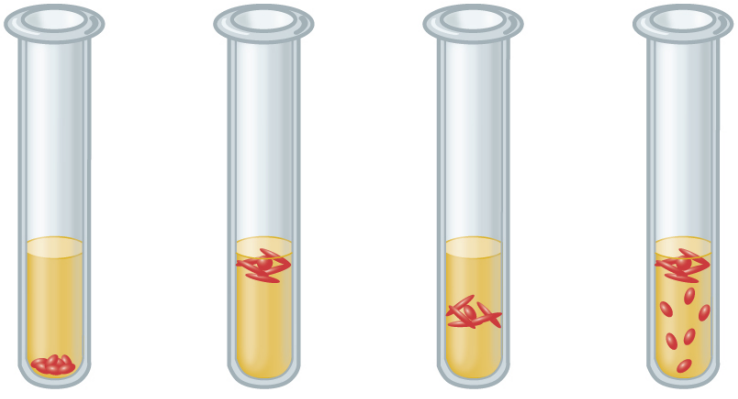
A. It uses CO_2 as a final electron acceptor in respiration.
B. It is an obligate anaerobe.
C. It is a capnophile.
D. It fixes CO_2 through photosynthesis.

Answer

C

Matching

Four tubes are illustrated with cultures grown in a medium that slows oxygen diffusion. Match the culture tube with the correct type of bacteria from the following list: facultative anaerobe, obligate anaerobe, microaerophile, aerotolerant anaerobe, obligate aerobe.



Answer

(a) obligate anaerobe, (b) obligate aerobe, (c) microaerophile, (d) facultative anaerobe

Short Answer

Why are some obligate anaerobes able to grow in tissues (e.g., gum pockets) that are not completely free of oxygen?

Why should *Haemophilus influenzae* be grown in a candle jar?

In terms of oxygen requirements, what type of organism would most likely be responsible for a foodborne illness associated with canned foods?

Critical Thinking

A microbiology instructor prepares cultures for a gram-staining practical laboratory by inoculating growth medium with a gram-positive coccus (nonmotile) and a gram-negative rod (motile). The goal is to demonstrate staining of a mixed culture. The flask is incubated at 35 °C for 24 hours without aeration. A sample is stained and reveals only gram-negative rods. Both cultures are known facultative anaerobes. Give a likely reason for success of the gram-negative rod. Assume that the cultures have comparable intrinsic growth rates.

9.3: The Effects of pH on Microbial Growth

Bacteria are generally neutrophiles. They grow best at neutral pH close to 7.0. Acidophiles grow optimally at a pH near 3.0. Alkaliphiles are organisms that grow optimally between a pH of 8 and 10.5. Extreme acidophiles and alkaliphiles grow slowly or not at all near neutral pH. Microorganisms grow best at their optimum growth pH. Growth occurs slowly or not at all below the minimum growth pH and above the maximum growth pH.

Multiple Choice

Bacteria that grow in mine drainage at pH 1–2 are probably which of the following?

A. alkaliphiles
B. acidophiles
C. neutrophiles
D. obligate anaerobes

Answer

B

Bacteria isolated from Lake Natron, where the water pH is close to 10, are which of the following?

A. alkaliphiles
B. facultative anaerobes
C. neutrophiles
D. obligate anaerobes

Answer

A

In which environment are you most likely to encounter an acidophile?

A. human blood at pH 7.2
B. a hot vent at pH 1.5
C. human intestine at pH 8.5
D. milk at pH 6.5

Answer

B

Fill in the Blank

A bacterium that thrives in a soda lake where the average pH is 10.5 can be classified as a(n) _____.

Answer

alkaliphile

Lactobacillus acidophilus grows best at pH 4.5. It is considered a(n) _____.

Answer

acidophile

Short Answer

Which macromolecule in the cell is most sensitive to changes in pH?

Which metabolic process in the bacterial cell is particularly challenging at high pH?

Critical Thinking

People who use proton pumps inhibitors or antacids are more prone to infections of the gastrointestinal tract. Can you explain the observation in light of what you have learned?

9.4: Temperature and Microbial Growth

Microorganisms thrive at a wide range of temperatures; they have colonized different natural environments and have adapted to extreme temperatures. Both extreme cold and hot temperatures require evolutionary adjustments to macromolecules and biological processes. Psychrophiles grow best in the temperature range of 0–15 °C whereas psychrotrophs thrive between 4 °C and 25 °C. Mesophiles grow best at moderate temperatures in the range of 20 °C to about 45 °C. Pathogens are usually mesophiles.

Multiple Choice

A soup container was forgotten in the refrigerator and shows contamination. The contaminants are probably which of the following?

A. thermophiles
B. acidophiles
C. mesophiles
D. psychrotrophs

Answer

D

Bacteria isolated from a hot tub at 39 °C are probably which of the following?

A. thermophiles
B. psychrotrophs
C. mesophiles
D. hyperthermophiles

Answer

C

In which environment are you most likely to encounter a hyperthermophile?

A. hot tub
B. warm ocean water in Florida
C. hydrothermal vent at the bottom of the ocean
D. human body

Answer

C

Which of the following environments would harbor psychrophiles?

A. mountain lake with a water temperature of 12 °C
B. contaminated plates left in a 35 °C incubator
C. yogurt cultured at room temperature
D. salt pond in the desert with a daytime temperature of 34 °C

Answer

A

Matching

Match the type of bacterium with its environment. Each choice may be used once, more than once, or not at all. Put the appropriate letter beside the environment.

___psychotroph	A. water heater set at 50 °C
___mesophile	B. hydrothermal vent
___thermophile	C. deep ocean waters
___hyperthermophile	D. human pathogen
___psychrophile	E. soil bacteria in temperate forest

Answer

C, D, E, B, A

Short Answer

How are hyperthermophile's proteins adapted to the high temperatures of their environment?

Why would NASA be funding microbiology research in Antarctica?

Critical Thinking

The bacterium that causes Hansen's disease (leprosy), *Mycobacterium leprae*, infects mostly the extremities of the body: hands, feet, and nose. Can you make an educated guess as to its optimum temperature of growth?

Refer to Figure 9.4.2. Some hyperthermophiles can survive autoclaving temperatures. Are they a concern in health care?

9.5: Other Environmental Conditions that Affect Growth

Microorganisms interact with their environment along more dimensions than pH, temperature, and free oxygen levels, although these factors require significant adaptations. We also find microorganisms adapted to varying levels of salinity, barometric pressure, humidity, and light.

Multiple Choice

Which of the following is the reason jams and dried meats often do not require refrigeration to prevent spoilage?

A. low pH
B. toxic alkaline chemicals
C. naturally occurring antibiotics
D. low water activity

Answer

D

Bacteria living in salt marshes are most likely which of the following?

A. acidophiles
B. barophiles
C. halotolerant
D. thermophiles

Answer

C

Fill in the Blank

A bacterium that thrives in the Great Salt Lake but not in fresh water is probably a _____.

Answer

halophile

Bacteria isolated from the bottom of the ocean need high atmospheric pressures to survive. They are _____.

Answer

barophiles

Staphylococcus aureus can be grown on multipurpose growth medium or on mannitol salt agar that contains 7.5% NaCl. The bacterium is _____.

Answer

halotolerant

Short Answer

Fish sauce is a salty condiment produced using fermentation. What type of organism is likely responsible for the fermentation of the fish sauce?

9.6: Media Used for Bacterial Growth

The study of microorganisms is greatly facilitated if we are able to culture them, that is, to keep reproducing populations alive under laboratory conditions. Culturing many microorganisms is challenging because of highly specific nutritional and environmental requirements and the diversity of these requirements among different species.

Multiple Choice

EMB agar is a medium used in the identification and isolation of pathogenic bacteria. It contains digested meat proteins as a source of organic nutrients. Two indicator dyes, eosin and methylene blue, inhibit the growth of gram-positive bacteria and distinguish between lactose fermenting and nonlactose fermenting organisms. Lactose fermenters form metallic green or deep purple colonies, whereas the nonlactose fermenters form completely colorless colonies. EMB agar is an example of which of the following?

A. a selective medium only
B. a differential medium only
C. a selective medium and a chemically defined medium
D. a selective medium, a differential medium, and a complex medium

Answer

D

Haemophilus influenzae must be grown on chocolate agar, which is blood agar treated with heat to release growth factors in the medium. *H. influenzae* is described as _____.

A. an acidophile
B. a thermophile
C. an obligate anaerobe
D. fastidious

Answer

D

Fill in the Blank

Blood agar contains many unspecified nutrients, supports the growth of a large number of bacteria, and allows differentiation of bacteria according to hemolysis (breakdown of blood). The medium is _____ and _____.

Answer

complex, differential

Rogosa agar contains yeast extract. The pH is adjusted to 5.2 and discourages the growth of many microorganisms; however, all the colonies look similar. The medium is _____ and _____.

Answer

complex, selective

Short Answer

What is the major difference between an enrichment culture and a selective culture?

Critical Thinking

Haemophilus, *influenzae* grows best at 35–37 °C with ~5% CO_2 (or in a candle-jar) and requires hemin (X factor) and nicotinamide-adenine-dinucleotide (NAD, also known as V factor) for growth.[1] Using the vocabulary learned in this chapter, describe *H. influenzae*.

Footnotes

1. 1 Centers for Disease Control and Prevention, World Health Organization. "*CDC Laboratory Methods for the Diagnosis of Meningitis Caused by* Neisseria meningitidis, Streptococcus pneumoniae, *and* Haemophilus influenza. WHO Manual, 2nd edition." 2011. http://www.cdc.gov/meningitis/lab-ma...ull-manual.pdf

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CHAPTER OVERVIEW

12: DNA and RNA

Children inherit some characteristics from each parent. Siblings typically look similar to each other, but not exactly the same—except in the case of identical twins. How can we explain these phenomena? The answers lie in heredity (the transmission of traits from one generation to the next) and genetics (the science of heredity). Because humans reproduce sexually, 50% of a child's genes come from the mother's egg cell and the remaining 50% from the father's sperm cell. Sperm and egg are formed through the process of meiosis, where DNA recombination occurs. Thus, there is no predictable pattern as to which 50% comes from which parent. Thus, siblings have only some genes, and their associated characteristics, in common. Identical twins are the exception, because they are genetically identical.

Genetic differences among related microbes also dictate many observed biochemical and virulence differences. For example, some strains of the bacterium *Escherichia coli* are harmless members of the normal microbiota in the human gastrointestinal tract. Other strains of the same species have genes that give them the ability to cause disease. In bacteria, such genes are not inherited via sexual reproduction, as in humans. Often, they are transferred via plasmids, small circular pieces of double-stranded DNA that can be exchanged between prokaryotes.



Figure **12.1**: Siblings within a family share some genes with each other and with each parent. Identical twins, however, are genetically identical. Bacteria like *Escherichia coli* may acquire genes encoding virulence factors, converting them into pathogenic strains, like this uropathogenic *E. coli*. (Credit left: Army twins, Myrtle Beach, S.C. / U.S. Army; Public Domain; credit right: modification of work by American Society for Microbiology)

12.1: Using Microbiology to Discover the Secrets of Life
12.2: Structure and Function of DNA
12.3: Structure and Function of RNA
12.4: The Structure and Function of Cellular Genomes

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12.1: Using Microbiology to Discover the Secrets of Life

Learning Objectives

- Describe the discovery of nucleic acid and nucleotides
- Explain the historical experiments that led to the characterization of DNA
- Describe how microbiology and microorganisms have been used to discover the biochemistry of genes

Through the early 20th century, DNA was not yet recognized as the genetic material responsible for heredity, the passage of traits from one generation to the next. In fact, much of the research was dismissed until the mid-20th century. The scientific community believed, incorrectly, that the process of inheritance involved a blending of parental traits that produced an intermediate physical appearance in offspring; this hypothetical process appeared to be correct because of what we know now as continuous variation, which results from the action of many genes to determine a particular characteristic, like human height. Offspring appear to be a "blend" of their parents' traits when we look at characteristics that exhibit continuous variation. The blending theory of inheritance asserted that the original parental traits were lost or absorbed by the blending in the offspring, but we now know that this is not the case.

Two separate lines of research, begun in the mid to late 1800s, ultimately led to the discovery and characterization of DNA and the foundations of genetics, the science of heredity. These lines of research began to converge in the 1920s, and research using microbial systems ultimately resulted in significant contributions to elucidating the molecular basis of genetics.

Discovery and Characterization of DNA

Modern understanding of DNA has evolved from the discovery of nucleic acid to the development of the double-helix model. In the 1860s, Friedrich Miescher (1844–1895), a physician by profession, was the first person to isolate phosphorus-rich chemicals from leukocytes (white blood cells) from the pus on used bandages from a local surgical clinic. He named these chemicals (which would eventually be known as RNA and DNA) "nuclein" because they were isolated from the nuclei of the cells. His student Richard Altmann (1852–1900) subsequently termed it "nucleic acid" 20 years later when he discovered the acidic nature of nuclein. In the last two decades of the 19th century, German biochemist Albrecht Kossel (1853–1927) isolated and characterized the five different nucleotide bases composing nucleic acid. These are adenine, guanine, cytosine, thymine (in DNA), and uracil (in RNA). Kossell received the Nobel Prize in Physiology or Medicine in 1910 for his work on nucleic acids and for his considerable work on proteins, including the discovery of histidine.

Foundations of Genetics

Despite the discovery of DNA in the late 1800s, scientists did not make the association with heredity for many more decades. To make this connection, scientists, including a number of microbiologists, performed many experiments on plants, animals, and bacteria.

Mendel's Pea Plants

While Miescher was isolating and discovering DNA in the 1860s, Austrian monk and botanist **Johann Gregor Mendel**(1822–1884) was experimenting with garden peas, demonstrating and documenting basic patterns of inheritance, now known as **Mendel's laws**.

In 1856, Mendel began his decade-long research into inheritance patterns. He used the diploid **garden pea**, *Pisum sativum*, as his primary model system because it naturally self-fertilizes and is highly inbred, producing "true-breeding" pea plant lines—plants that always produce offspring that look like the parent. By experimenting with true-breeding pea plants, Mendel avoided the appearance of unexpected traits in offspring that might occur if he used plants that were not true-breeding. Mendel performed **hybridizations**, which involve mating two true-breeding individuals (P generation) that have different traits, and examined the characteristics of their offspring (first filial generation, F_1) as well as the offspring of self-fertilization of the F_1 generation (second filial generation, F_2) (Figure **12.1.1**).

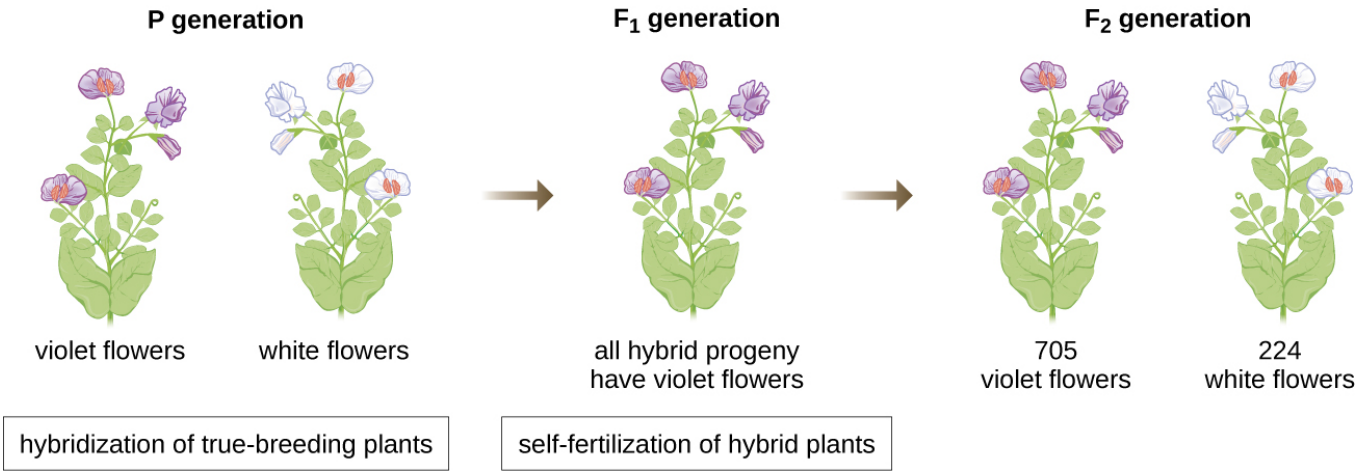


Figure **12.1.1**: In one of his experiments on inheritance patterns, Mendel crossed plants that were true- breeding for violet flower color with plants true-breeding for white flower color (the P generation). The resulting hybrids in the F_1 generation all had violet flowers. In the F_2 generation, approximately three-quarters of the plants had violet flowers, and one-quarter had white flowers.

In 1865, Mendel presented the results of his experiments with nearly 30,000 pea plants to the local natural history society. He demonstrated that traits are transmitted faithfully from parents to offspring independently of other traits. In 1866, he published his work, "Experiments in Plant Hybridization,"[1] in the *Proceedings of the Natural History Society of Brünn*. Mendel's work went virtually unnoticed by the scientific community, which believed, incorrectly, in the theory of blending of traits in continuous variation.

He was not recognized for his extraordinary scientific contributions during his lifetime. In fact, it was not until 1900 that his work was rediscovered, reproduced, and revitalized by scientists on the brink of discovering the chromosomal basis of heredity.

The Chromosomal Theory of Inheritance

Mendel carried out his experiments long before chromosomes were visualized under a microscope. However, with the improvement of microscopic techniques during the late 1800s, cell biologists could stain and visualize subcellular structures with dyes and observe their actions during meiosis. They were able to observe chromosomes replicating, condensing from an amorphous nuclear mass into distinct X-shaped bodies and migrating to separate cellular poles. The speculation that chromosomes might be the key to understanding heredity led several scientists to examine Mendel's publications and re-evaluate his model in terms of the behavior of chromosomes during mitosis and meiosis.

In 1902, **Theodor Boveri** (1862–1915) observed that in sea urchins, nuclear components (chromosomes) determined proper embryonic development. That same year, **Walter Sutton** (1877–1916) observed the separation of chromosomes into daughter cells during meiosis. Together, these observations led to the development of the Chromosomal Theory of Inheritance, which identified chromosomes as the genetic material responsible for Mendelian inheritance.

Despite compelling correlations between the behavior of chromosomes during meiosis and Mendel's observations, the Chromosomal Theory of Inheritance was proposed long before there was any direct evidence that traits were carried on chromosomes. **Thomas Hunt Morgan** (1866–1945) and his colleagues spent several years carrying out crosses with the fruit fly, *Drosophila melanogaster*. They performed meticulous microscopic observations of fly chromosomes and correlated these observations with resulting fly characteristics. Their work provided the first experimental evidence to support the Chromosomal Theory of Inheritance in the early 1900s. In 1915, Morgan and his "Fly Room" colleagues published *The Mechanism of Mendelian Heredity*, which identified chromosomes as the cellular structures responsible for heredity. For his many significant contributions to genetics, Morgan received the Nobel Prize in Physiology or Medicine in 1933.

In the late 1920s, **Barbara McClintock** (1902–1992) developed chromosomal staining techniques to visualize and differentiate between the different chromosomes of maize (corn). In the 1940s and 1950s, she identified a breakage event on chromosome 9, which she named the dissociation locus (*Ds*). *Ds* could change position within the chromosome. She also identified an activator locus (*Ac*). *Ds* chromosome breakage could be activated by an *Ac* element (transposase enzyme). At first, McClintock's finding of

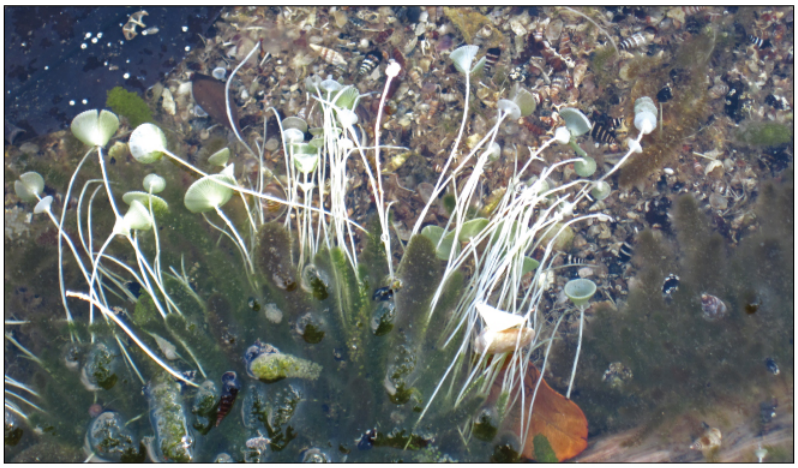
these jumping genes, which we now call transposons, was not accepted by the scientific community. It wasn't until the 1960s and later that transposons were discovered in bacteriophages, bacteria, and *Drosophila*. Today, we know that transposons are mobile segments of DNA that can move within the genome of an organism. They can regulate gene expression, protein expression, and virulence (ability to cause disease).

Microbes and Viruses in Genetic Research

Microbiologists have also played a crucial part in our understanding of genetics. Experimental organisms such as Mendel's garden peas, Morgan's fruit flies, and McClintock's corn had already been used successfully to pave the way for an understanding of genetics. However, microbes and viruses were (and still are) excellent model systems for the study of genetics because, unlike peas, fruit flies, and corn, they are propagated more easily in the laboratory, growing to high population densities in a small amount of space and in a short time. In addition, because of their structural simplicity, microbes and viruses are more readily manipulated genetically.

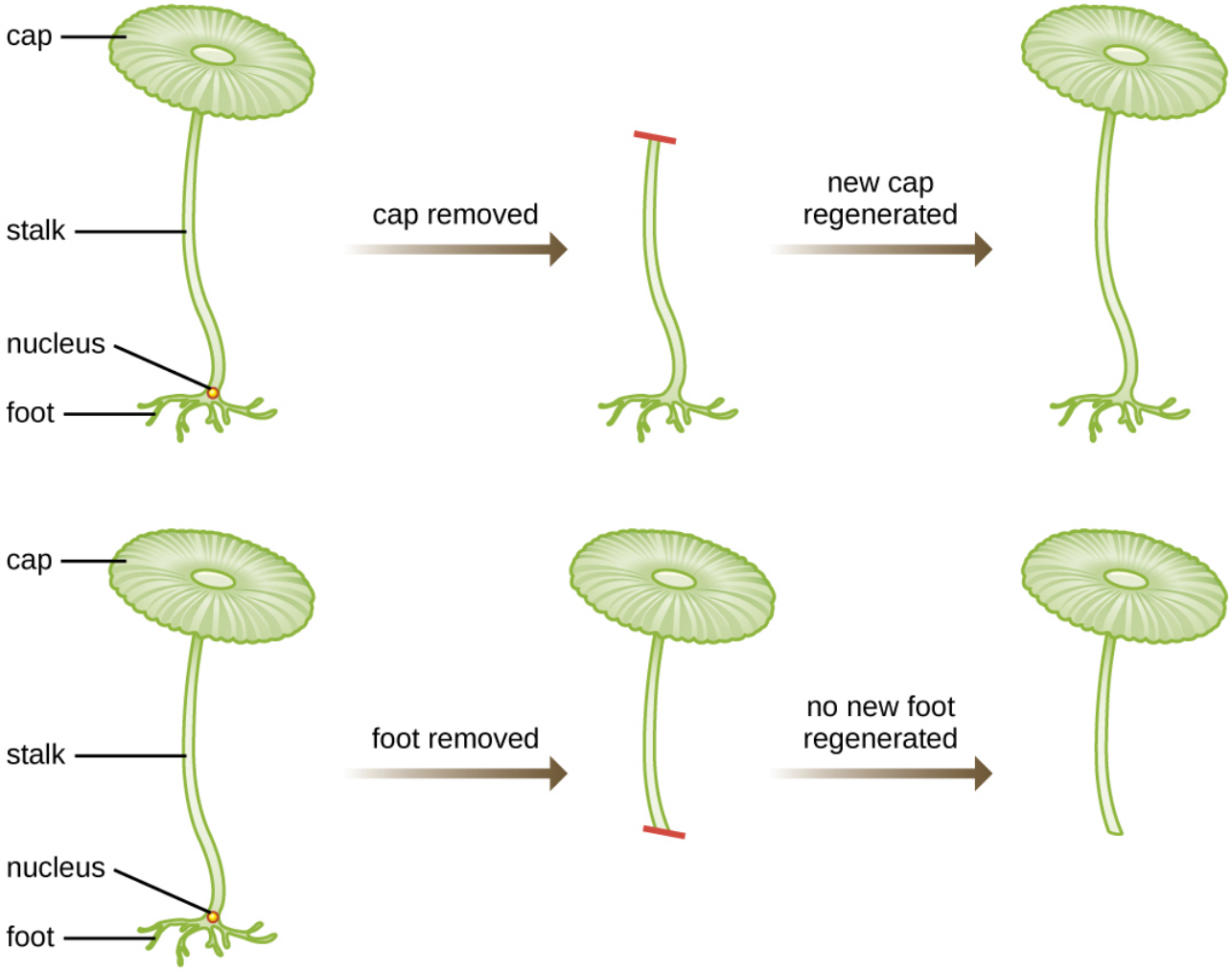
Fortunately, despite significant differences in size, structure, reproduction strategies, and other biological characteristics, there is biochemical unity among all organisms; they have in common the same underlying molecules responsible for heredity and the use of genetic material to give cells their varying characteristics. In the words of French scientist Jacques Monod, "What is true for *E. coli* is also true for the elephant," meaning that the biochemistry of life has been maintained throughout evolution and is shared in all forms of life, from simple unicellular organisms to large, complex organisms. This biochemical continuity makes microbes excellent models to use for genetic studies.

In a clever set of experiments in the 1930s and 1940s, German scientist **Joachim Hämmerling** (1901–1980), using the single-celled alga *Acetabularia* as a microbial model, established that the genetic information in a eukaryotic cell is housed within the nucleus. *Acetabularia* spp. are unusually large algal cells that grow asymmetrically, forming a "foot" containing the nucleus, which is used for substrate attachment; a stalk; and an umbrella-like cap—structures that can all be easily seen with the naked eye. In an early set of experiments, Hämmerling removed either the cap or the foot of the cells and observed whether new caps or feet were regenerated (Figure **12.1.2**). He found that when the foot of these cells was removed, new feet did not grow; however, when caps were removed from the cells, new caps were regenerated. This suggested that the hereditary information was located in the nucleus-containing foot of each cell.



Acetabularia

(a)



(b)

Figure **12.1.2**: (a) The cells of the single-celled alga *Acetabularia* measure 2–6 cm and have a cell morphology that can be observed with the naked eye. Each cell has a cap, a stalk, and a foot, which contains the nucleus. (b) Hämmerling found that if he removed the cap, a new cap would regenerate; but if he removed the foot, a new foot would not regenerate. He concluded that the genetic information needed for regeneration was found in the nucleus. (credit a: modification of work by James St. John)

In another set of experiments, Hämmerling used two species of *Acetabularia* that have different cap morphologies, *A. crenulata* and *A. mediterranea* (Figure **12.1.3**). He cut the caps from both types of cells and then grafted the stalk from an *A. crenulata* onto an *A. mediterranea* foot, and vice versa. Over time, he observed that the grafted cell with the *A. crenulata* foot and *A. mediterranea* stalk developed a cap with the *A. crenulata* morphology. Conversely, the grafted cell with the *A. mediterranea* foot and *A. crenulata* stalk developed a cap with the *A. mediterranea* morphology. He microscopically confirmed the presence of nuclei in the feet of these cells and attributed the development of these cap morphologies to the nucleus of each grafted cell. Thus, he showed experimentally that the nucleus was the location of genetic material that dictated a cell's properties.

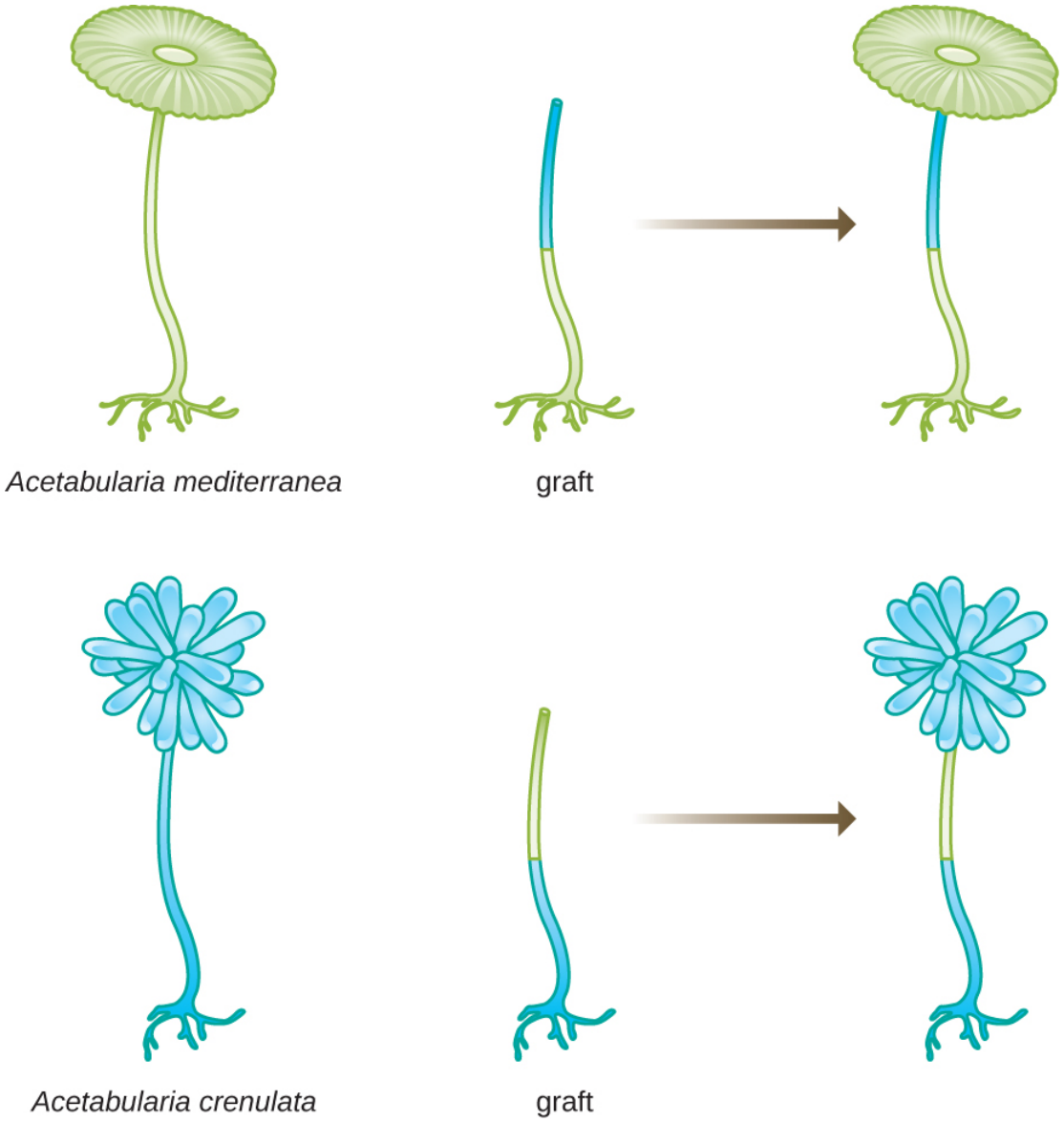


Figure **12.1.3**: In a second set of experiments, Hämmerling used two morphologically different species and grafted stalks from each species to the feet of the other. He found that the properties of the regenerated caps were dictated by the species of the nucleus-containing foot.

Another microbial model, the red bread mold *Neurospora crassa*, was used by **George Beadle** and **Edward Tatum** to demonstrate the relationship between genes and the proteins they encode. Beadle had worked with fruit flies in Morgan's laboratory but found them too complex to perform certain types of experiments. *N. crassa*, on the other hand, is a simpler organism and has the ability to

grow on a minimal medium because it contains enzymatic pathways that allow it to use the medium to produce its own vitamins and amino acids.

Beadle and Tatum irradiated the mold with X-rays to induce changes to a sequence of nucleic acids, called **mutations**. They mated the irradiated mold spores and attempted to grow them on both a complete medium and a minimal medium. They looked for mutants that grew on a complete medium, supplemented with vitamins and amino acids, but did not grow on the minimal medium lacking these supplements. Such molds theoretically contained mutations in the genes that encoded biosynthetic pathways. Upon finding such mutants, they systematically tested each to determine which vitamin or amino acid it was unable to produce (Figure 12.1.4) and published this work in 1941.

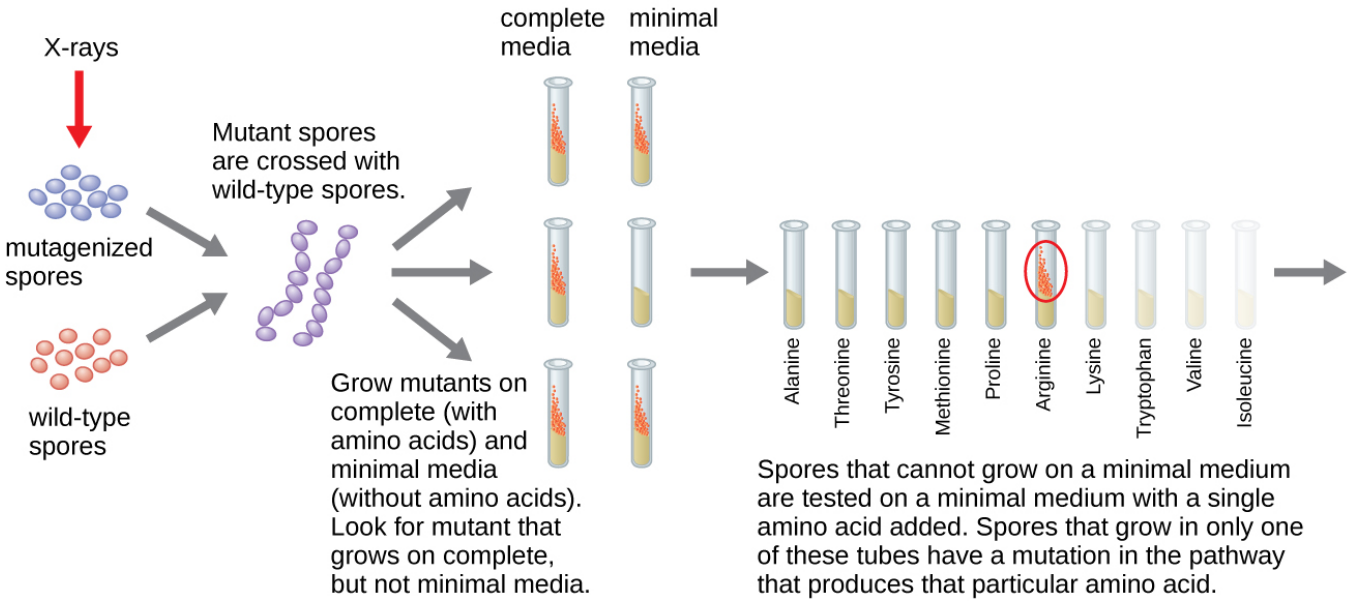


Figure 12.1.4: Beadle and Tatum's experiment involved the mating of irradiated and nonirradiated mold spores. These spores were grown on both complete medium and a minimal medium to determine which amino acid or vitamin the mutant was unable to produce on its own.

Subsequent work by Beadle, Tatum, and colleagues showed that they could isolate different classes of mutants that required a particular supplement, like the amino acid arginine (Figure 12.1.5). With some knowledge of the arginine biosynthesis pathway, they identified three classes of arginine mutants by supplementing the minimal medium with intermediates (citrulline or ornithine) in the pathway. The three mutants differed in their abilities to grow in each of the media, which led the group of scientists to propose, in 1945, that each type of mutant had a defect in a different gene in the arginine biosynthesis pathway. This led to the so-called **one gene–one enzyme hypothesis**, which suggested that each gene encodes one enzyme.

Subsequent knowledge about the processes of transcription and translation led scientists to revise this to the "one gene–one polypeptide" hypothesis. Although there are some genes that do not encode polypeptides (but rather encode for transfer RNAs [tRNAs] or ribosomal RNAs [rRNAs], which we will discuss later), the one gene–one enzyme hypothesis is true in many cases, especially in microbes. Beadle and Tatum's discovery of the link between genes and corresponding characteristics earned them the 1958 Nobel Prize in Physiology and Medicine and has since become the basis for modern molecular genetics.

Beadle and Tatum Experiments				
Bread Mold	**Minimal Medium (MM)**	**MM + Ornithine**	**MM + Citrulline**	**MM + Arginine**
Wild type	grew	grew	grew	grew
Mutant 1	**did not grow**	grew	grew	grew
Mutant 2	**did not grow**	**did not grow**	grew	grew
Mutant 3	**did not grow**	**did not grow**	**did not grow**	grew

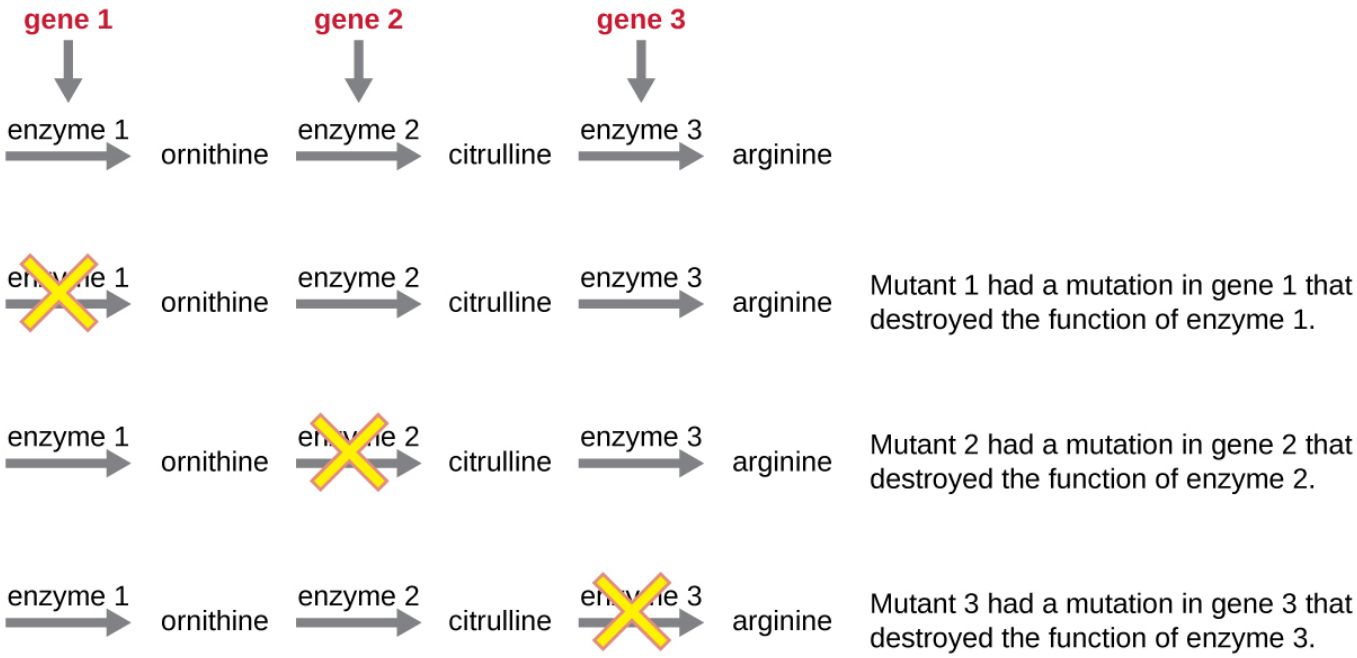


Figure **12.1.5**: Three classes of arginine mutants were identified, each differing in their ability to grow in the presence of intermediates in the arginine biosynthesis pathway. From this, Beadle and Tatum concluded that each mutant was defective in a different gene encoding a different enzyme in the arginine biosynthesis pathway, leading to them to their one gene–one enzyme hypothesis.

Link to Learning

To learn more about the experiments of Beadle and Tatum, visit this website from the DNA Learning Center.

Key Concepts and Summary

- **DNA** was discovered and characterized long before its role in heredity was understood. Microbiologists played significant roles in demonstrating that DNA is the hereditary information found within cells.
- In the 1850s and 1860s, Gregor Mendel experimented with true-breeding garden peas to demonstrate the **heritability** of specific observable traits.
- In 1869, Friedrich Miescher isolated and purified a compound rich in phosphorus from the nuclei of white blood cells; he named the compound nuclein. Miescher's student Richard Altmann discovered its acidic nature, renaming it **nucleic acid**. Albrecht Kossell characterized the **nucleotide bases** found within nucleic acids.

- Although Walter Sutton and Theodor Boveri proposed the **Chromosomal Theory of Inheritance** in 1902, it was not scientifically demonstrated until the 1915 publication of the work of Thomas Hunt Morgan and his colleagues.
- Using *Acetabularia*, a large algal cell, as his model system, Joachim Hämmerling demonstrated in the 1930s and 1940s that the nucleus was the location of hereditary information in these cells.
- In the 1940s, George Beadle and Edward Tatum used the mold *Neurospora crassa* to show that each protein's production was under the control of a single gene, demonstrating the **"one gene–one enzyme" hypothesis**.

Footnotes

1. 1 J.G. Mendel. "Versuche über Pflanzenhybriden." *Verhandlungen des naturforschenden Vereines in Brünn, Bd. Abhandlungen* 4 (1865):3–7. (For English translation, see http://www.mendelweb.org/Mendel.plain.html)
2. 2 G.W. Beadle, E.L. Tatum. "Genetic Control of Biochemical Reactions in *Neurospora.*" *Proceedings of the National Academy of Sciences* 27 no. 11 (1941):499–506.
3. 3 F. Griffith. "The Significance of Pneumococcal Types." *Journal of Hygiene* 27 no. 2 (1928):8–159.
4. 4 A.D. Hershey, M. Chase. "Independent Functions of Viral Protein and Nucleic Acid in Growth of Bacteriophage." *Journal of General Physiology* 36 no. 1 (1952):39–56.

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12.2: Structure and Function of DNA

Learning Objectives

- Describe the biochemical structure of deoxyribonucleotides
- Identify the base pairs used in the synthesis of deoxyribonucleotides
- Explain why the double helix of DNA is described as antiparallel

In Microbial Metabolism, we discussed three classes of macromolecules: proteins, lipids, and carbohydrates. In this chapter, we will discuss a fourth class of macromolecules: nucleic acids. Like other macromolecules, **nucleic acids** are composed of monomers, called **nucleotides**, which are polymerized to form large strands. Each nucleic acid strand contains certain nucleotides that appear in a certain order within the strand, called its **base sequence**. The base sequence of **deoxyribonucleic acid (DNA)** is responsible for carrying and retaining the hereditary information in a cell. In Mechanisms of Microbial Genetics, we will discuss in detail the ways in which DNA uses its own base sequence to direct its own synthesis, as well as the synthesis of RNA and proteins, which, in turn, gives rise to products with diverse structure and function. In this section, we will discuss the basic structure and function of DNA.

DNA Nucleotides

The building blocks of nucleic acids are nucleotides. Nucleotides that compose DNA are called **deoxyribonucleotides**. The three components of a deoxyribonucleotide are a five-carbon sugar called deoxyribose, a phosphate group, and a **nitrogenous base**, a nitrogen-containing ring structure that is responsible for complementary base pairing between nucleic acid strands (Figure 12.2.1). The carbon atoms of the five-carbon deoxyribose are numbered 1′, 2′, 3′, 4′, and 5′ (1′ is read as “one prime”). A nucleoside comprises the five-carbon sugar and nitrogenous base.

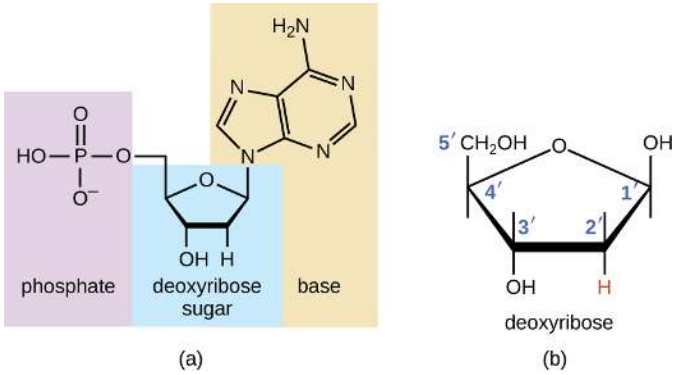


Figure 12.2.1: (a) Each deoxyribonucleotide is made up of a sugar called deoxyribose, a phosphate group, and a nitrogenous base—in this case, adenine. (b) The five carbons within deoxyribose are designated as 1′, 2′, 3′, 4′, and 5′.

The deoxyribonucleotide is named according to the nitrogenous bases (Figure 12.2.2). The nitrogenous bases **adenine (A)** and **guanine (G)** are the purines; they have a double-ring structure with a six-carbon ring fused to a five-carbon ring. The pyrimidines, **cytosine (C)** and **thymine (T)**, are smaller nitrogenous bases that have only a six-carbon ring structure.

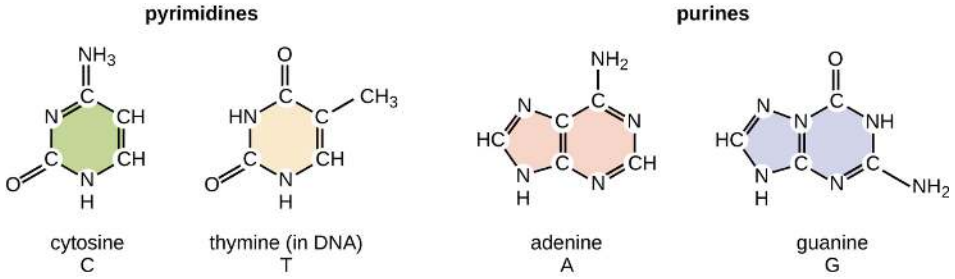


Figure 12.2.2: Nitrogenous bases within DNA are categorized into the two-ringed purines adenine and guanine and the single-ringed pyrimidines cytosine and thymine. Thymine is unique to DNA.

Individual nucleoside triphosphates combine with each other by covalent bonds known as **5′-3′ phosphodiester bonds**, or linkages whereby the phosphate group attached to the 5′ carbon of the sugar of one nucleotide bonds to the hydroxyl group of the 3′ carbon of the sugar of the next nucleotide. Phosphodiester bonding between nucleotides forms the **sugar-phosphate backbone**, the alternating sugar-phosphate structure composing the framework of a nucleic acid strand (Figure **12.2.3**). During the polymerization process, deoxynucleotide triphosphates (dNTP) are used. To construct the sugar-phosphate backbone, the two terminal phosphates are released from the dNTP as a pyrophosphate. The resulting strand of nucleic acid has a free phosphate group at the 5′ carbon end and a free hydroxyl group at the 3′ carbon end. The two unused phosphate groups from the nucleotide triphosphate are released as pyrophosphate during phosphodiester bond formation. Pyrophosphate is subsequently hydrolyzed, releasing the energy used to drive nucleotide polymerization.

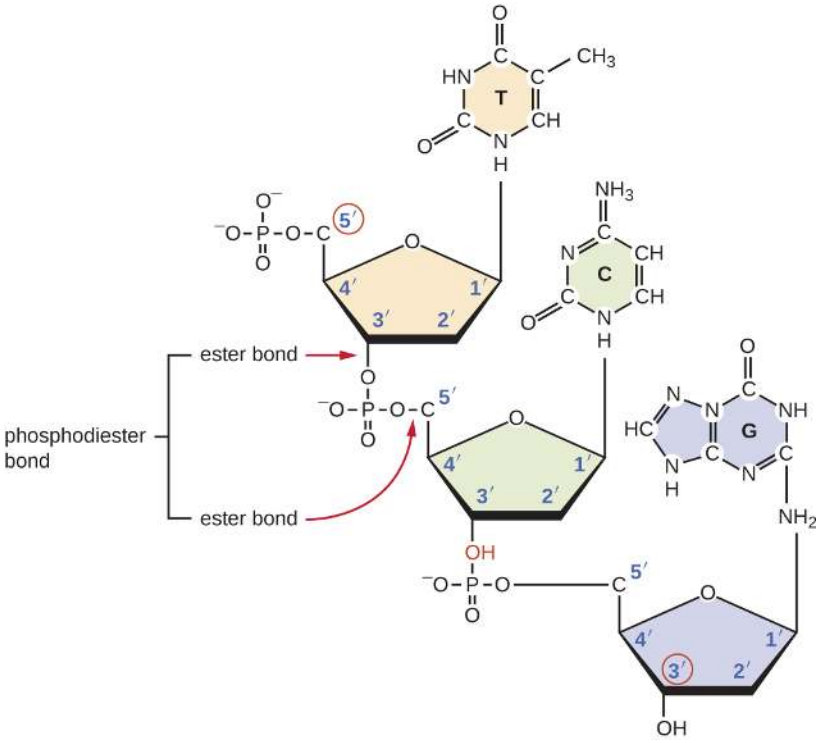


Figure **12.2.3**: Phosphodiester bonds form between the phosphate group attached to the 5′ carbon of one nucleotide and the hydroxyl group of the 3′ carbon in the next nucleotide, bringing about polymerization of nucleotides in to nucleic acid strands. Note the 5′ and 3′ ends of this nucleic acid strand.

Discovering the Double Helix

By the early 1950s, considerable evidence had accumulated indicating that DNA was the genetic material of cells, and now the race was on to discover its three-dimensional structure. Around this time, Austrian biochemist **Erwin Chargaff**[1](1905–2002) examined the content of DNA in different species and discovered that adenine, thymine, guanine, and cytosine were not found in equal quantities, and that it varied from species to species, but not between individuals of the same species. He found that the amount of adenine was very close to equaling the amount of thymine, and the amount of cytosine was very close to equaling the amount of guanine, or A = T and G = C. These relationships are also known as **Chargaff's rules**.

Other scientists were also actively exploring this field during the mid-20th century. In 1952, American scientist **Linus Pauling** (1901–1994) was the world's leading structural chemist and odds-on favorite to solve the structure of DNA. Pauling had earlier discovered the structure of protein α helices, using X-ray diffraction, and, based upon X-ray diffraction images of DNA made in his laboratory, he proposed a triple-stranded model of DNA.[2] At the same time, British researchers **Rosalind Franklin** (1920–1958) and her graduate student R.G. Gosling were also using X-ray diffraction to understand the structure of DNA (Figure **12.2.4**). It was Franklin's scientific expertise that resulted in the production of more well-defined X-ray diffraction images of DNA that would clearly show the overall double-helix structure of DNA.

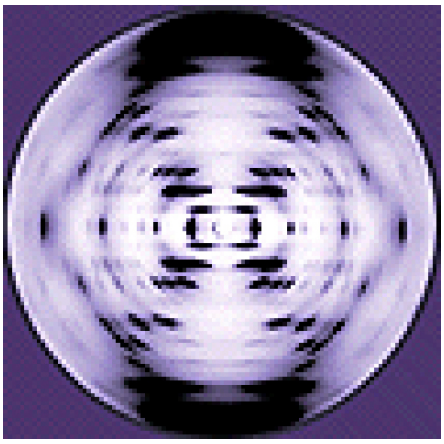


Figure **12.2.4**: The X-ray diffraction pattern of DNA shows its helical nature. (credit: National Institutes of Health)

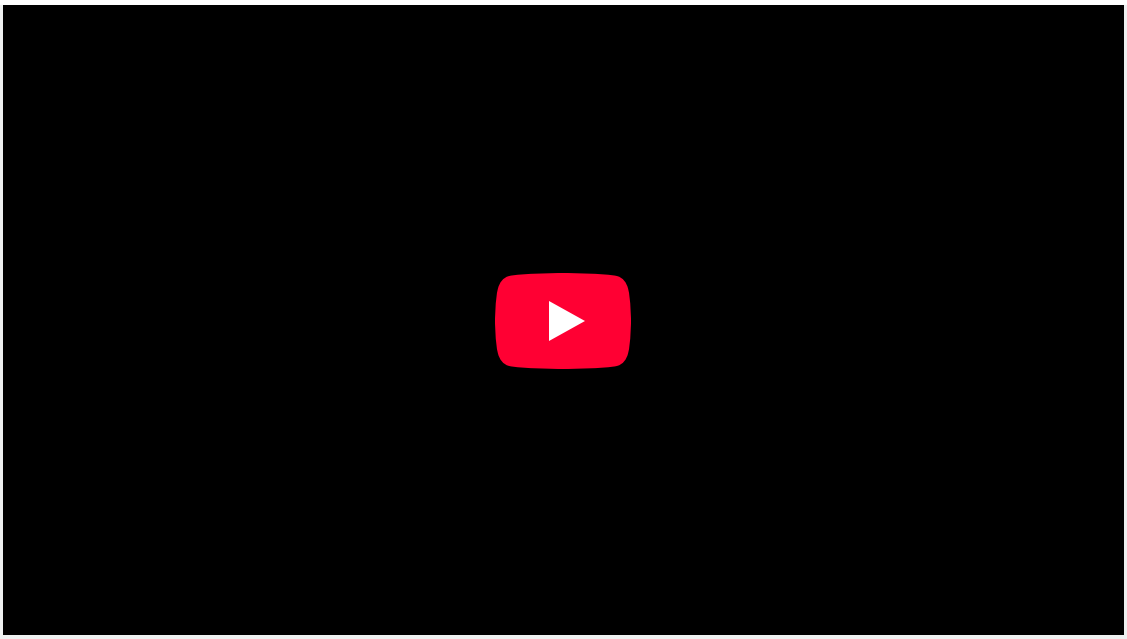
James Watson (1928–), an American scientist, and **Francis Crick** (1916–2004), a British scientist, were working together in the 1950s to discover DNA's structure. They used Chargaff's rules and Franklin and Wilkins' X-ray diffractionimages of DNA fibers to piece together the purine-pyrimidine pairing of the double helical DNA molecule (Figure **12.2.5**). In April 1953, Watson and Crick published their model of the **DNA double helix** in *Nature*.[3] The same issue additionally included papers by Wilkins and colleagues,[4] as well as by Franklin and Gosling,[5] each describing different aspects of the molecular structure of DNA. In 1962, James Watson, Francis Crick, and **Maurice Wilkins** were awarded the Nobel Prize in Physiology and Medicine. Unfortunately, by then Franklin had died, and Nobel prizes at the time were not awarded posthumously. Work continued, however, on learning about the structure of DNA. In 1973, Alexander Rich(1924–2015) and colleagues were able to analyze DNA crystals to confirm and further elucidate DNA structure.[6]



Figure **12.2.5**: In 1953, James Watson and Francis Crick built this model of the structure of DNA, shown here on display at the Science Museum in London.

Video: The DNA Double Helix Discovery - HHMI Biointeractive Video

The discovery of the structure of the DNA double helix was one of the most important of the 20th century. In this educational video, explore Watson and Crick's quest to understand DNA's structure, and Rosalind Franklin's key insights via x-ray crystallography.





DNA Structure

Watson and Crick proposed that DNA is made up of two strands that are twisted around each other to form a right-handed helix. The two DNA strands are **antiparallel**, such that the 3′ end of one strand faces the 5′ end of the other (Figure **12.2.6**). The 3′ end of each strand has a free hydroxyl group, while the 5′ end of each strand has a free phosphate group. The sugar and phosphate of the polymerized nucleotides form the backbone of the structure, whereas the nitrogenous bases are stacked inside. These nitrogenous bases on the interior of the molecule interact with each other, **base pairing**.

Analysis of the diffraction patterns of DNA has determined that there are approximately 10 bases per turn in DNA. The asymmetrical spacing of the sugar-phosphate backbones generates **major grooves** (where the backbone is far apart) and **minor grooves** (where the backbone is close together) (Figure **12.2.6**). These grooves are locations where proteins can bind to DNA. The binding of these proteins can alter the structure of DNA, regulate replication, or regulate transcription of DNA into RNA.

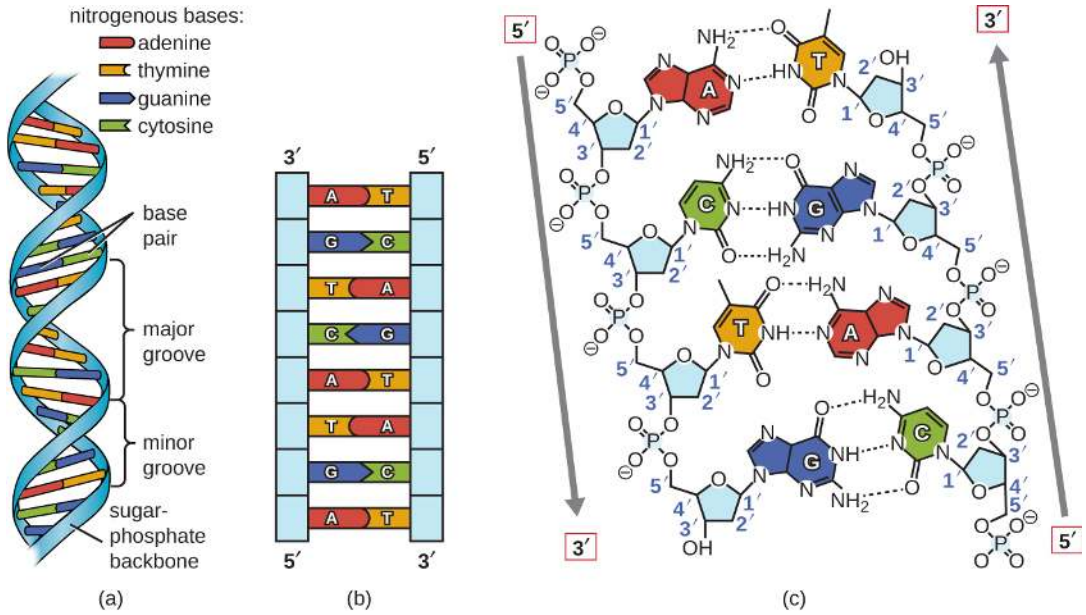


Figure 12.2.6: Watson and Crick proposed the double helix model for DNA. (a) The sugar-phosphate backbones are on the outside of the double helix and purines and pyrimidines form the "rungs" of the DNA helix ladder. (b) The two DNA strands are antiparallel to each other. (c) The direction of each strand is identified by numbering the carbons (1 through 5) in each sugar molecule. The 5′ end is the one where carbon #5 is not bound to another nucleotide; the 3′ end is the one where carbon #3 is not bound to another nucleotide.

Base pairing takes place between a purine and pyrimidine. In DNA, adenine (A) and thymine (T) are complementary base pairs, and cytosine (C) and guanine (G) are also complementary base pairs, explaining Chargaff's rules (Figure 12.2.7). The base pairs are stabilized by **hydrogen bonds**; adenine and thymine form two hydrogen bonds between them, whereas cytosine and guanine form three hydrogen bonds between them.

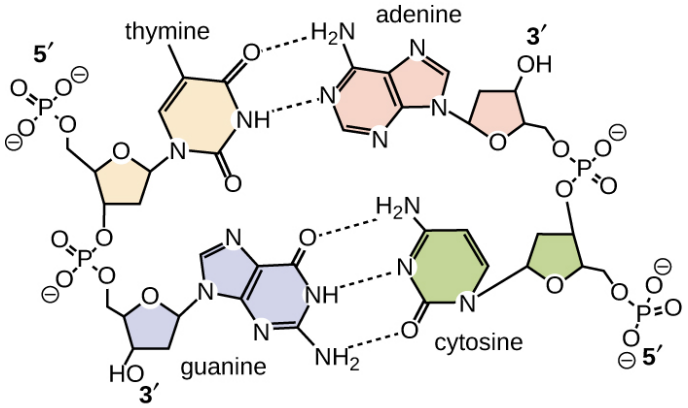


Figure 12.2.7: Hydrogen bonds form between complementary nitrogenous bases on the interior of DNA.

In the laboratory, exposing the two DNA strands of the double helix to high temperatures or to certain chemicals can break the hydrogen bonds between complementary bases, thus separating the strands into two separate single strands of DNA (single-stranded DNA [ssDNA]). This process is called **DNA denaturation** and is analogous to protein denaturation, as described in Proteins. The ssDNA strands can also be put back together as double-stranded DNA (dsDNA), through reannealing or renaturing by cooling or removing the chemical denaturants, allowing these hydrogen bonds to reform. The ability to artificially manipulate DNA in this way is the basis for several important techniques in biotechnology (Figure 12.2.8). Because of the additional hydrogen bonding between the C = G base pair, DNA with a high GC content is more difficult to denature than DNA with a lower GC content.

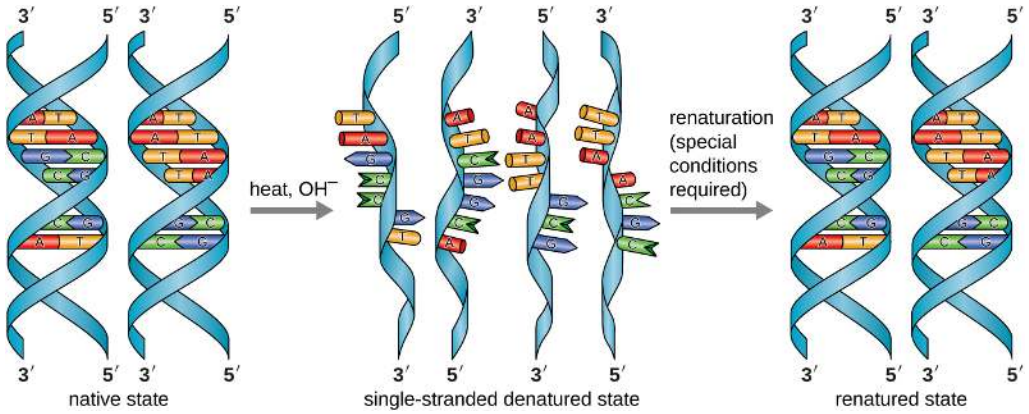


Figure **12.2.8**: In the laboratory, the double helix can be denatured to single-stranded DNA through exposure to heat or chemicals, and then renatured through cooling or removal of chemical denaturants to allow the DNA strands to reanneal. (credit: modification of work by Hernández-Lemus E, Nicasio-Collazo LA, Castañeda-Priego R)

Link to Learning

View an animation on DNA structure from the DNA Learning Center to learn more.

DNA Function

DNA stores the information needed to build and control the cell. The transmission of this information from mother to daughter cells is called vertical gene transfer and it occurs through the process of DNA replication. DNA is replicated when a cell makes a duplicate copy of its DNA, then the cell divides, resulting in the correct distribution of one DNA copy to each resulting cell. DNA can also be enzymatically degraded and used as a source of nucleosides and nucleotides for the cell. Unlike other macromolecules, DNA does not serve a structural role in cells.

Paving the Way for Women in Science and Health Professions

Historically, women have been underrepresented in the sciences and in medicine, and often their pioneering contributions have gone relatively unnoticed. For example, although Rosalind Franklin performed the X-ray diffraction studies demonstrating the double helical structure of DNA, it is Watson and Crick who became famous for this discovery, building on her data. There still remains great controversy over whether their acquisition of her data was appropriate and whether personality conflicts and gender bias contributed to the delayed recognition of her significant contributions. Similarly, Barbara McClintock did pioneering work in maize (corn) genetics from the 1930s through 1950s, discovering transposons (jumping genes), but she was not recognized until much later, receiving a Nobel Prize in Physiology or Medicine in 1983 (Figure **12.2.9**).

Today, women still remain underrepresented in many fields of science and medicine. While more than half of the undergraduate degrees in science are awarded to women, only 46% of doctoral degrees in science are awarded to women. In academia, the number of women at each level of career advancement continues to decrease, with women holding less than one-third of the positions of Ph.D.-level scientists in tenure-track positions, and less than one-quarter of the full professorships at 4-year colleges and universities.[7] Even in the health professions, like nearly all other fields, women are often underrepresented in many medical careers and earn significantly less than their male counterparts, as shown in a 2013 study published by the *Journal of the American Medical Association*.[8]

Why do such disparities continue to exist and how do we break these cycles? The situation is complex and likely results from the combination of various factors, including how society conditions the behaviors of girls from a young age and supports their interests, both professionally and personally. Some have suggested that women do not belong in the laboratory, including Nobel Prize winner Tim Hunt, whose 2015 public comments suggesting that women are too emotional for science[9] were met with widespread condemnation.

Perhaps girls should be supported more from a young age in the areas of science and math (Figure **12.2.9**). Science, technology, engineering, and mathematics (STEM) programs sponsored by the American Association of University Women (AAUW)[10] and National Aeronautics and Space Administration (NASA)[11] are excellent examples of programs that offer such support.

Contributions by women in science should be made known more widely to the public, and marketing targeted to young girls should include more images of historically and professionally successful female scientists and medical professionals, encouraging all bright young minds, including girls and women, to pursue careers in science and medicine.



Figure 12.2.9: (a) Barbara McClintock's work on maize genetics in the 1930s through 1950s resulted in the discovery of transposons, but its significance was not recognized at the time. (b) Efforts to appropriately mentor and to provide continued societal support for women in science and medicine may someday help alleviate some of the issues preventing gender equality at all levels in science and medicine. (credit a: modification of work by Smithsonian Institution; credit b: modification of work by Haynie SL, Hinkle AS, Jones NL, Martin CA, Olsiewski PJ, Roberts MF)

Key Concepts and Summary

- **Nucleic acids** are composed of **nucleotides**, each of which contains a pentose sugar, a phosphate group, and a **nitrogenous base**. **Deoxyribonucleotides** within DNA contain **deoxyribose** as the pentose sugar.
- DNA contains the **pyrimidines cytosine** and **thymine**, and the **purines adenine** and **guanine**.
- **Nucleotides** are linked together by phosphodiester bonds between the 5′ phosphate group of one nucleotide and the 3′ hydroxyl group of another. A **nucleic acid strand** has a free phosphate group at the 5′ end and a free hydroxyl group at the 3′ end.
- Chargaff discovered that the amount of **adenine** is approximately equal to the amount of **thymine** in DNA, and that the amount of the **guanine** is approximately equal to **cytosine**. These relationships were later determined to be due to complementary base pairing.
- Watson and Crick, building on the work of Chargaff, Franklin and Gosling, and Wilkins, proposed the double helix model and base pairing for DNA structure.
- DNA is composed of two complementary strands oriented **antiparallel** to each other with the **phosphodiester backbones** on the exterior of the molecule. The nitrogenous bases of each strand face each other and complementary bases hydrogen bond to each other, stabilizing the double helix.
- Heat or chemicals can break the hydrogen bonds between complementary bases, denaturing DNA. Cooling or removing chemicals can lead to renaturation or reannealing of DNA by allowing hydrogen bonds to reform between complementary bases.
- DNA stores the instructions needed to build and control the cell. This information is transmitted from parent to offspring through **vertical gene transfer**.

Footnotes

1. 1 N. Kresge et al. "Chargaff's Rules: The Work of Erwin Chargaff." *Journal of Biological Chemistry* 280 (2005):e21.
2. 2 L. Pauling, "A Proposed Structure for the Nucleic Acids." *Proceedings of the National Academy of Science of the United States of America* 39 no. 2 (1953):84–97.
3. 3 J.D. Watson, F.H.C. Crick. "A Structure for Deoxyribose Nucleic Acid." *Nature* 171 no. 4356 (1953):737–738.
4. 4 M.H.F. Wilkins et al. "Molecular Structure of Deoxypentose Nucleic Acids." *Nature* 171 no. 4356 (1953):738–740.
5. 5 R. Franklin, R.G. Gosling. "Molecular Configuration in Sodium Thymonucleate." *Nature* 171 no. 4356 (1953):740–741.
6. 6 R.O. Day et al. "A Crystalline Fragment of the Double Helix: The Structure of the Dinucleoside Phosphate Guanylyl-3',5'-Cytidine." *Proceedings of the National Academy of Sciences of the United States of America* 70 no. 3 (1973):849–853.

7. 7 N.H. Wolfinger “For Female Scientists, There's No Good Time to Have Children.” *The Atlantic* July 29, 2013. www.theatlantic.com/sexes/arc...ildren/278165/.
8. 8 S.A. Seabury et al. “Trends in the Earnings of Male and Female Health Care Professionals in the United States, 1987 to 2010.” *Journal of the American Medical Association Internal Medicine* 173 no. 18 (2013):1748–1750.
9. 9 E. Chung. “Tim Hunt, Sexism and Science: The Real 'Trouble With Girls' in Labs.” *CBC News Technology and Science*, June 12, 2015. http://www.cbc.ca/news/technology/ti...labs-1.3110133. Accessed 8/4/2016.
10. 10 American Association of University Women. “Building a STEM Pipeline for Girls and Women.” www.aauw.org/what-we-do/stem-education/. Accessed June 10, 2016.
11. 11 National Aeronautics and Space Administration. “Outreach Programs: Women and Girls Initiative.” http://women.nasa.gov/outreach-programs/. Accessed June 10, 2016.

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12.3: Structure and Function of RNA

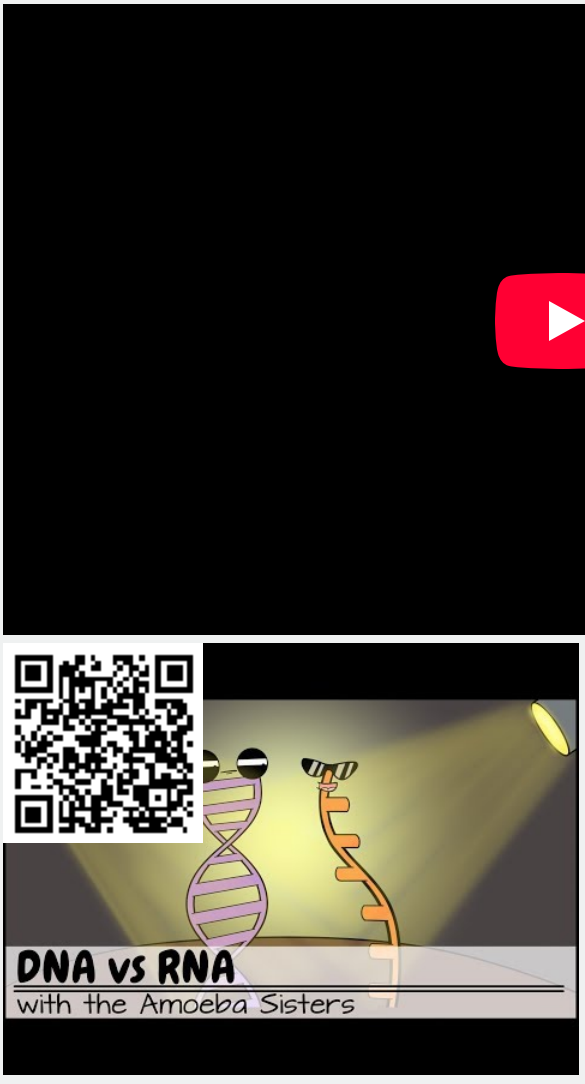
Learning Objectives

- Describe the biochemical structure of ribonucleotides
- Describe the similarities and differences between RNA and DNA
- Describe the functions of the three main types of RNA used in protein synthesis
- Explain how RNA can serve as hereditary information

Structurally speaking, **ribonucleic acid (RNA)**, is quite similar to DNA. However, whereas DNA molecules are typically long and double stranded, RNA molecules are much shorter and are typically single stranded. RNA molecules perform a variety of roles in the cell but are mainly involved in the process of protein synthesis (translation) and its regulation.

Video: DNA vs RNA

Why is RNA just as cool as DNA? Join the Amoeba Sisters as they compare and contrast RNA with DNA and learn why DNA should be sharing the limelight!



RNA Structure

RNA is typically single stranded and is made of **ribonucleotides** that are linked by phosphodiester bonds. A ribonucleotide in the RNA chain contains **ribose** (the pentose sugar), one of the four nitrogenous bases (A, U, G, and C), and a phosphate group. The subtle structural difference between the sugars gives DNA added stability, making DNA more suitable for storage of genetic information, whereas the relative instability of RNA makes it more suitable for its more short-term functions.

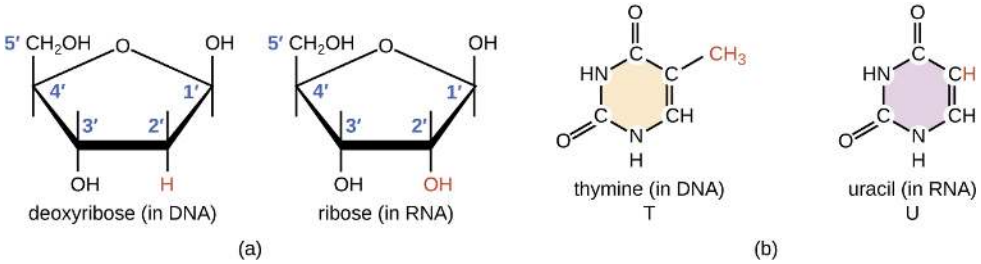


Figure **12.3.1**: (a) Ribonucleotides contain the pentose sugar ribose instead of the deoxyribose found in deoxyribonucleotides. (b) RNA contains the pyrimidine uracil in place of thymine found in DNA.

The RNA-specific pyrimidine **uracil** forms a complementary base pair with adenine and is used instead of the thymine used in DNA. Even though RNA is single stranded, most types of RNA molecules show extensive intramolecular base pairing between complementary sequences within the RNA strand, creating a predictable three-dimensional structure essential for their function (Figure **12.3.1** and Figure **12.3.2**).

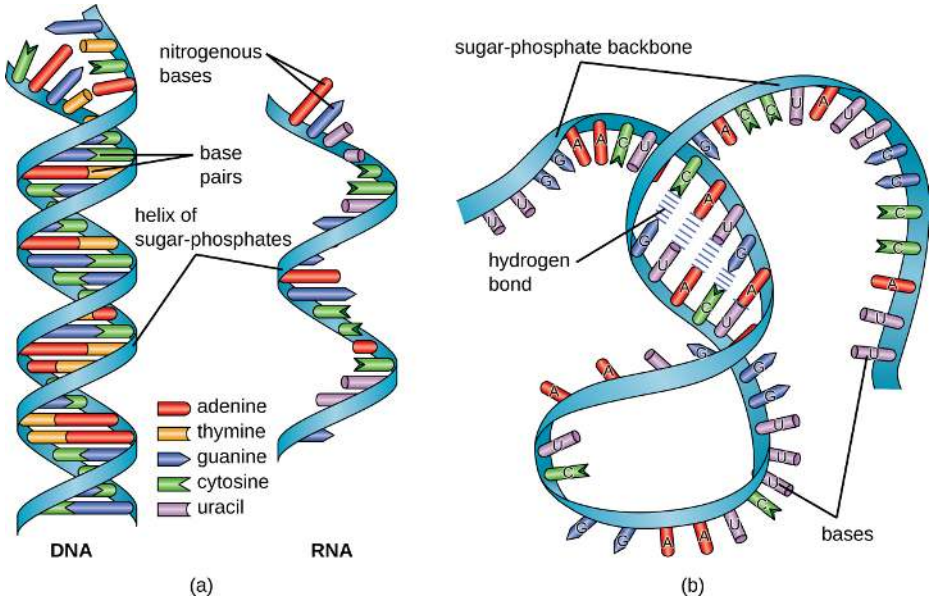


Figure **12.3.2**: (a) DNA is typically double stranded, whereas RNA is typically single stranded. (b) Although it is single stranded, RNA can fold upon itself, with the folds stabilized by short areas of complementary base pairing within the molecule, forming a three-dimensional structure.

Functions of RNA in Protein Synthesis

Cells access the information stored in DNA by creating RNA to direct the synthesis of proteins through the process of translation. Proteins within a cell have many functions, including building cellular structures and serving as enzyme catalysts for cellular chemical reactions that give cells their specific characteristics. The three main types of RNA directly involved in protein synthesis are **messenger RNA (mRNA)**, **ribosomal RNA (rRNA)**, and **transfer RNA (tRNA)**.

In 1961, French scientists François Jacob and Jacques Monod hypothesized the existence of an intermediary between DNA and its protein products, which they called messenger RNA.[1] Evidence supporting their hypothesis was gathered soon afterwards showing that information from DNA is transmitted to the ribosome for protein synthesis using mRNA. If DNA serves as the complete

library of cellular information, mRNA serves as a photocopy of specific information needed at a particular point in time that serves as the instructions to make a protein.

The **mRNA** carries the message from the DNA, which controls all of the cellular activities in a cell. If a cell requires a certain protein to be synthesized, the gene for this product is "turned on" and the mRNA is synthesized through the process of transcription (see RNA Transcription). The mRNA then interacts with ribosomes and other cellular machinery (Figure **12.3.3**) to direct the synthesis of the protein it encodes during the process of translation (see Protein Synthesis). mRNA is relatively unstable and short-lived in the cell, especially in prokaryotic cells, ensuring that proteins are only made when needed.

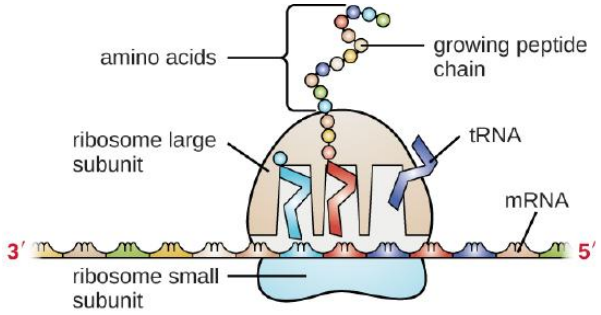


Figure **12.3.3**: A generalized illustration of how mRNA and tRNA are used in protein synthesis within a cell.

rRNA and tRNA are stable types of RNA. In prokaryotes and eukaryotes, tRNA and rRNA are encoded in the DNA, then copied into long RNA molecules that are cut to release smaller fragments containing the individual mature RNA species. In eukaryotes, synthesis, cutting, and assembly of rRNA into ribosomes takes place in the nucleolus region of the nucleus, but these activities occur in the cytoplasm of prokaryotes. Neither of these types of RNA carries instructions to direct the synthesis of a polypeptide, but they play other important roles in protein synthesis.

Ribosomes are composed of **rRNA** and **protein**. As its name suggests, rRNA is a major constituent of ribosomes, composing up to about 60% of the ribosome by mass and providing the location where the mRNA binds. The rRNA ensures the proper alignment of the mRNA, tRNA, and the ribosomes; the rRNA of the ribosome also has an enzymatic activity (peptidyl transferase) and catalyzes the formation of the peptide bonds between two aligned amino acids during protein synthesis. Although rRNA had long been thought to serve primarily a structural role, its catalytic role within the ribosome was proven in 2000.[2] Scientists in the laboratories of Thomas Steitz (1940–) and Peter Moore(1939–) at Yale University were able to crystallize the ribosome structure from *Haloarcula marismortui*, a halophilic archaeon isolated from the Dead Sea. Because of the importance of this work, Steitz shared the 2009 Nobel Prize in Chemistry with other scientists who made significant contributions to the understanding of ribosome structure.

Transfer RNA is the third main type of RNA and one of the smallest, usually only 70–90 nucleotides long. It carries the correct amino acid to the site of protein synthesis in the ribosome. It is the base pairing between the tRNA and mRNA that allows for the correct amino acid to be inserted in the polypeptide chain being synthesized (Figure **12.3.4**). Any mutations in the tRNA or rRNA can result in global problems for the cell because both are necessary for proper protein synthesis (Table **12.3.1**).

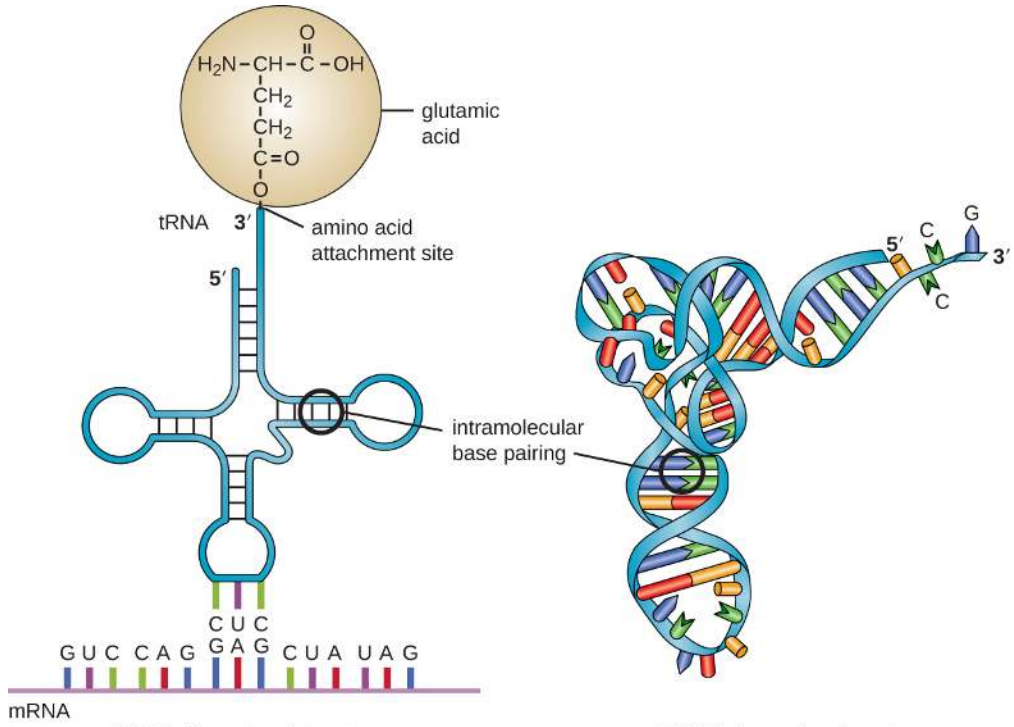


Figure **12.3.4**: A tRNA molecule is a single-stranded molecule that exhibits significant intracellular base pairing, giving it its characteristic three-dimensional shape.

Table **12.3.1**: Structure and Function of RNA

	mRNA	rRNA	tRNA
Structure	Short, unstable, single-stranded RNAcorresponding to a gene encoded within DNA	Longer, stable RNA molecules composing 60% of ribosome's mass	Short (70-90 nucleotides), stable RNA with extensive intramolecular base pairing; contains an amino acid binding site and an mRNA binding site
Function	Serves as intermediary between DNA and protein; used by ribosome to direct synthesis of protein it encodes	Ensures the proper alignment of mRNA, tRNA, and ribosome during protein synthesis; catalyzes peptide bond formation between amino acids	Carries the correct amino acid to the site of protein synthesis in the ribosome

RNA as Hereditary Information

Although RNA does not serve as the hereditary information in most cells, RNA does hold this function for many viruses that do not contain DNA. Thus, RNA clearly does have the additional capacity to serve as genetic information. Although RNA is typically single stranded within cells, there is significant diversity in viruses. Rhinoviruses, which cause the common cold; influenza viruses; and the Ebola virus are single-stranded RNA viruses. Rotaviruses, which cause severe gastroenteritis in children and other immunocompromised individuals, are examples of double-stranded RNA viruses. Because double-stranded RNA is uncommon in eukaryotic cells, its presence serves as an indicator of viral infection. The implications for a virus having an RNA genome instead of a DNA genome are discussed in more detail in Viruses.

Key Concepts and Summary

- **Ribonucleic acid (RNA)** is typically single stranded and contains ribose as its pentose sugar and the pyrimidine uracil instead of thymine. An RNA strand can undergo significant intramolecular base pairing to take on a three-dimensional structure.

- There are three main types of RNA, all involved in protein synthesis.
- Messenger RNA (**mRNA**) serves as the intermediary between DNA and the synthesis of protein products during translation.
- Ribosomal RNA (**rRNA**) is a type of stable RNA that is a major constituent of ribosomes. It ensures the proper alignment of the mRNA and the ribosomes during protein synthesis and catalyzes the formation of the peptide bonds between two aligned amino acids during protein synthesis.
- Transfer RNA (**tRNA**) is a small type of stable RNA that carries an amino acid to the corresponding site of protein synthesis in the ribosome. It is the base pairing between the tRNA and mRNA that allows for the correct amino acid to be inserted in the polypeptide chain being synthesized.
- Although RNA is not used for long-term genetic information in cells, many viruses do use RNA as their genetic material.

Footnotes

1. 1 A. Rich. “The Era of RNA Awakening: Structural Biology of RNA in the Early Years.” *Quarterly Reviews of Biophysics* 42 no. 2 (2009):117–137.
2. 2 P. Nissen et al. “The Structural Basis of Ribosome Activity in Peptide Bond Synthesis.” *Science* 289 no. 5481 (2000):920–930.

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12.4: The Structure and Function of Cellular Genomes

Learning Objectives

- Define gene and genotype and differentiate genotype from phenotype
- Describe chromosome structure and packaging
- Compare prokaryotic and eukaryotic chromosomes
- Explain why extrachromosomal DNA is important in a cell

Thus far, we have discussed the structure and function of individual pieces of DNA and RNA. In this section, we will discuss how all of an organism's genetic material—collectively referred to as its **genome**—is organized inside of the cell. Since an organism's genetics to a large extent dictate its characteristics, it should not be surprising that organisms differ in the arrangement of their DNA and RNA.

Genotype versus Phenotype

All cellular activities are encoded within a cell's DNA. The sequence of bases within a DNA molecule represents the genetic information of the cell. Segments of DNA molecules are called **genes**, and individual genes contain the instructional code necessary for synthesizing various proteins, enzymes, or stable RNA molecules.

The full collection of genes that a cell contains within its genome is called its **genotype**. However, a cell does not express all of its genes simultaneously. Instead, it turns on (expresses) or turns off certain genes when necessary. The set of genes being expressed at any given point in time determines the cell's activities and its observable characteristics, referred to as its **phenotype**. Genes that are always expressed are known as constitutive genes; some constitutive genes are known as housekeeping genes because they are necessary for the basic functions of the cell.

While the genotype of a cell remains constant, the phenotype may change in response to environmental signals (e.g., changes in temperature or nutrient availability) that affect which nonconstitutive genes are expressed. For example, the oral bacterium *Streptococcus mutans* produces a sticky slime layer that allows it to adhere to teeth, forming dental plaque; however, the genes that control the production of the slime layer are only expressed in the presence of sucrose (table sugar). Thus, while the genotype of *S. mutans* is constant, its phenotype changes depending on the presence and absence of sugar in its environment. Temperature can also regulate gene expression. For example, the gram-negative bacterium *Serratia marcescens*, a pathogen frequently associated with hospital-acquired infections, produces a red pigment at 28 °C but not at 37 °C, the normal internal temperature of the human body (Figure **12.4.1**).

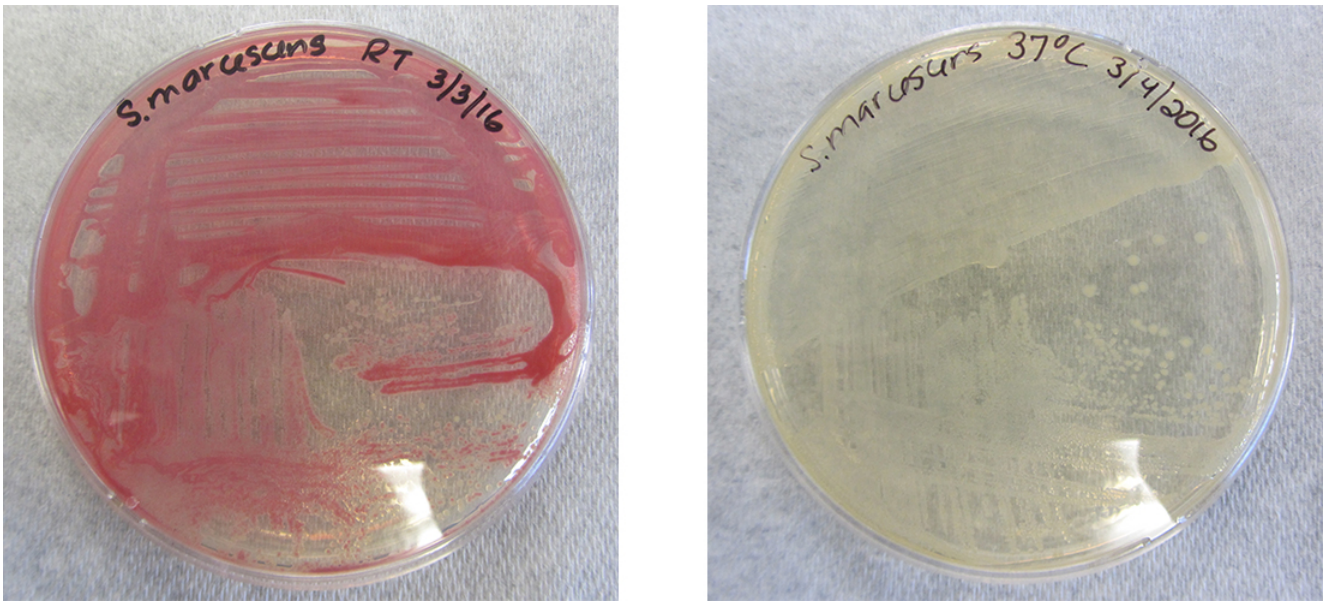


Figure 12.4.1: Both plates contain strains of *Serratia marcescens* that have the gene for red pigment. However, this gene is expressed at 28 °C (left) but not at 37 °C (right). (credit: modification of work by Ann Auman)

Organization of Genetic Material

The vast majority of an organism's genome is organized into the cell's **chromosomes**, which are discrete DNA structures within cells that control cellular activity. Recall that while eukaryotic chromosomes are housed in the membrane-bound nucleus, most prokaryotes contain a single, circular chromosome that is found in an area of the cytoplasm called the nucleoid (see Unique Characteristics of Prokaryotic Cells). A chromosome may contain several thousand genes.

Organization of Eukaryotic Chromosome

Chromosome structure differs somewhat between eukaryotic and prokaryotic cells. Eukaryotic chromosomes are typically linear, and eukaryotic cells contain multiple distinct chromosomes. Many eukaryotic cells contain two copies of each chromosome and, therefore, are **diploid**.

The length of a chromosome greatly exceeds the length of the cell, so a chromosome needs to be packaged into a very small space to fit within the cell. For example, the combined length of all of the 3 billion base pairs[1] of DNA of the human genome would measure approximately 2 meters if completely stretched out, and some eukaryotic genomes are many times larger than the human genome. DNA **supercoiling** refers to the process by which DNA is twisted to fit inside the cell. Supercoiling may result in DNA that is either underwound (less than one turn of the helix per 10 base pairs) or overwound (more than one turn per 10 base pairs) from its normal relaxed state. Proteins known to be involved in supercoiling include **topoisomerases**; these enzymes help maintain the structure of supercoiled chromosomes, preventing overwinding of DNA during certain cellular processes like DNA replication.

During **DNA packaging**, DNA-binding proteins called **histones** perform various levels of DNA wrapping and attachment to scaffolding proteins. The combination of DNA with these attached proteins is referred to as **chromatin**. In eukaryotes, the packaging of DNA by histones may be influenced by environmental factors that affect the presence of methyl groups on certain cytosine nucleotides of DNA. The influence of environmental factors on DNA packaging is called epigenetics. Epigenetics is another mechanism for regulating gene expression without altering the sequence of nucleotides. Epigenetic changes can be maintained through multiple rounds of cell division and, therefore, can be heritable.

Link to Learning

View this animation from the DNA Learning Center to learn more about on DNA packaging in eukaryotes.

Organization of Prokaryotic Chromosomes

Chromosomes in bacteria and archaea are usually circular, and a prokaryotic cell typically contains only a single chromosome within the nucleoid. Because the chromosome contains only one copy of each gene, prokaryotes are haploid. As in eukaryotic cells, DNA supercoiling is necessary for the genome to fit within the prokaryotic cell. The DNA in the bacterial chromosome is arranged in several supercoiled domains. As with eukaryotes, topoisomerases are involved in supercoiling DNA. DNA gyrase is a type of topoisomerase, found in bacteria and some archaea, that helps prevent the overwinding of DNA. (Some antibiotics kill bacteria by targeting DNA gyrase.) In addition, histone-like proteins bind DNA and aid in DNA packaging. Other proteins bind to the origin of replication, the location in the chromosome where DNA replication initiates. Because different regions of DNA are packaged differently, some regions of chromosomal DNA are more accessible to enzymes and thus may be used more readily as templates for gene expression. Interestingly, several bacteria, including *Helicobacter pylori* and *Shigella flexneri*, have been shown to induce epigenetic changes in their hosts upon infection, leading to chromatin remodeling that may cause long-term effects on host immunity.[2]

Noncoding DNA

In addition to genes, a genome also contains many regions of noncoding DNA that do not encode proteins or stable RNA products. Noncoding DNA is commonly found in areas prior to the start of coding sequences of genes as well as in intergenic regions (i.e., DNA sequences located between genes) (Figure **12.4.2**).

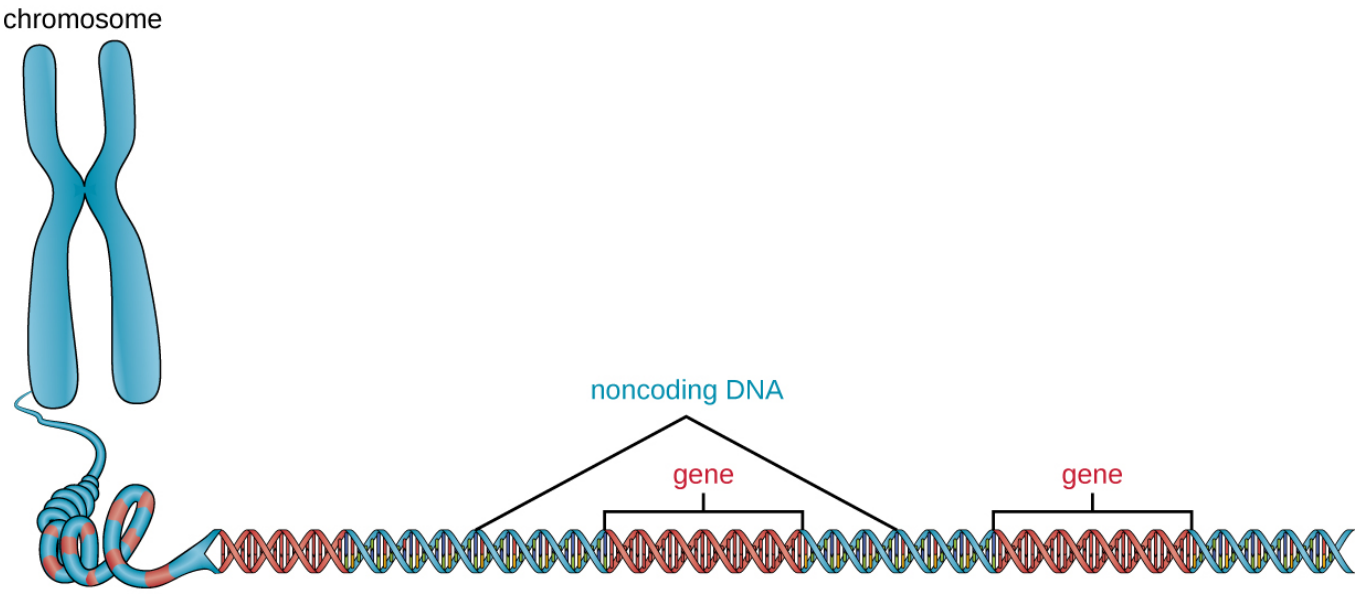


Figure **12.4.2**: Chromosomes typically have a significant amount of noncoding DNA, often found in intergenic regions.

Prokaryotes appear to use their genomes very efficiently, with only an average of 12% of the genome being taken up by noncoding sequences. In contrast, noncoding DNA can represent about 98% of the genome in eukaryotes, as seen in humans, but the percentage of noncoding DNA varies between species.[3] These noncoding DNA regions were once referred to as “junk DNA”; however, this terminology is no longer widely accepted because scientists have since found roles for some of these regions, many of which contribute to the regulation of transcription or translation through the production of small noncoding RNA molecules, DNA packaging, and chromosomal stability. Although scientists may not fully understand the roles of all noncoding regions of DNA, it is generally believed that they do have purposes within the cell.

Extrachromosomal DNA

Although most DNA is contained within a cell’s chromosomes, many cells have additional molecules of DNA outside the chromosomes, called extrachromosomal DNA, that are also part of its genome. The genomes of eukaryotic cells would also include

the chromosomes from any organelles such as mitochondria and/or chloroplasts that these cells maintain (Figure **12.4.3**). The maintenance of circular chromosomes in these organelles is a vestige of their prokaryotic origins and supports the endosymbiotic theory (see Foundations of Modern Cell Theory). In some cases, genomes of certain DNA viruses can also be maintained independently in host cells during latent viral infection. In these cases, these viruses are another form of extrachromosomal DNA. For example, the human papillomavirus (HPV) may be maintained in infected cells in this way.

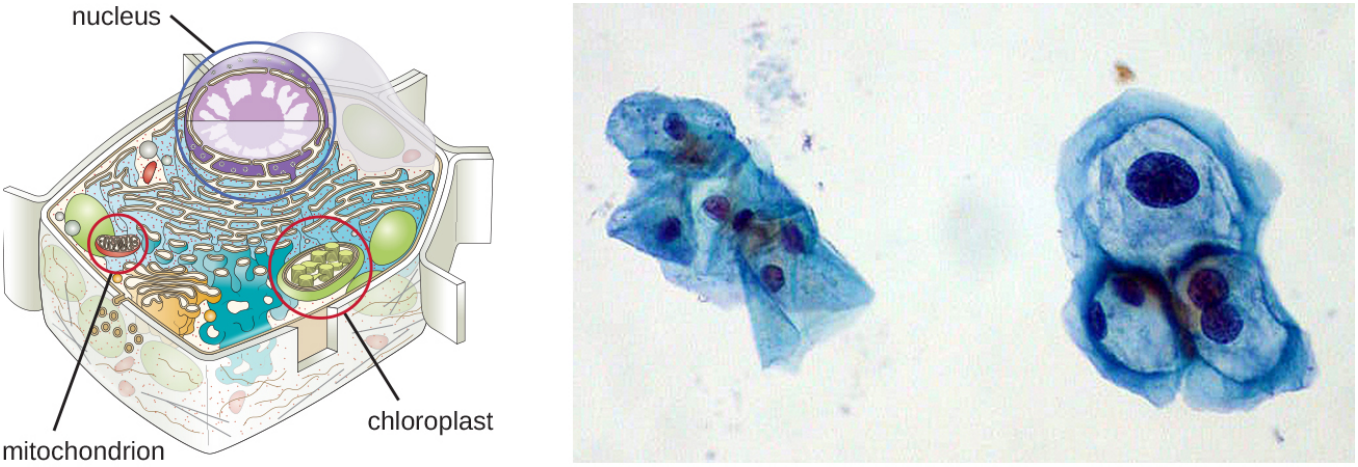


Figure **12.4.3**: The genome of a eukaryotic cell consists of the chromosome housed in the nucleus, and extrachromosomal DNA found in the mitochondria (all cells) and chloroplasts (plants and algae).

Besides chromosomes, some prokaryotes also have smaller loops of DNA called **plasmids** that may contain one or a few genes not essential for normal growth (Figure 3.3.1). Bacteria can exchange these plasmids with other bacteria in a process known as horizontal gene transfer (HGT). The exchange of genetic material on plasmids sometimes provides microbes with new genes beneficial for growth and survival under special conditions. In some cases, genes obtained from plasmids may have clinical implications, encoding virulence factors that give a microbe the ability to cause disease or make a microbe resistant to certain antibiotics. Plasmids are also used heavily in genetic engineering and biotechnology as a way to move genes from one cell to another. The role of plasmids in horizontal gene transfer and biotechnology will be discussed further in Mechanisms of Microbial Genetics and Modern Applications of Microbial Genetics.

Lethal Plasmids

Maria, a 20-year-old anthropology student from Texas, recently became ill in the African nation of Botswana, where she was conducting research as part of a study-abroad program. Maria's research was focused on traditional African methods of tanning hides for the production of leather. Over a period of three weeks, she visited a tannery daily for several hours to observe and participate in the tanning process. One day, after returning from the tannery, Maria developed a fever, chills, and a headache, along with chest pain, muscle aches, nausea, and other flu-like symptoms. Initially, she was not concerned, but when her fever spiked and she began to cough up blood, her African host family became alarmed and rushed her to the hospital, where her condition continued to worsen.

After learning about her recent work at the tannery, the physician suspected that Maria had been exposed to anthrax. He ordered a chest X-ray, a blood sample, and a spinal tap, and immediately started her on a course of intravenous penicillin. Unfortunately, lab tests confirmed the physician's presumptive diagnosis. Maria's chest X-ray exhibited pleural effusion, the accumulation of fluid in the space between the pleural membranes, and a Gram stain of her blood revealed the presence of gram-positive, rod-shaped bacteria in short chains, consistent with *Bacillus anthracis*. Blood and bacteria were also shown to be present in her cerebrospinal fluid, indicating that the infection had progressed to meningitis. Despite supportive treatment and aggressive antibiotic therapy, Maria slipped into an unresponsive state and died three days later.

Anthrax is a disease caused by the introduction of endospores from the gram-positive bacterium *B. anthracis* into the body. Once infected, patients typically develop meningitis, often with fatal results. In Maria's case, she inhaled the endospores while handling the hides of animals that had been infected.

The genome of *B. anthracis* illustrates how small structural differences can lead to major differences in virulence. In 2003, the genomes of *B. anthracis* and *Bacillus cereus*, a similar but less pathogenic bacterium of the same genus, were sequenced and compared.[4] Researchers discovered that the 16S rRNA gene sequences of these bacteria are more than 99% identical, meaning that they are actually members of the same species despite their traditional classification as separate species. Although their chromosomal sequences also revealed a great deal of similarity, several virulence factors of *B. anthracis* were found to be encoded on two large plasmids not found in *B. cereus*. The plasmid pX01 encodes a three-part toxin that suppresses the host immune system, whereas the plasmid pX02 encodes a capsular polysaccharide that further protects the bacterium from the host immune system (Figure **12.4.4**). Since *B. cereus* lacks these plasmids, it does not produce these virulence factors, and although it is still pathogenic, it is typically associated with mild cases of diarrhea from which the body can quickly recover. Unfortunately for Maria, the presence of these toxin-encoding plasmids in *B. anthracis* gives it its lethal virulence.

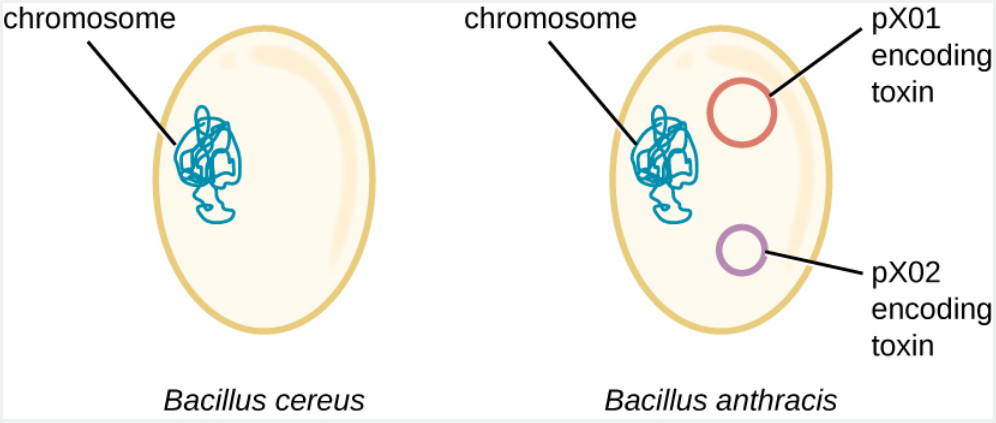


Figure **12.4.4**: Genome sequencing of Bacillus anthracis and its close relative *B. cereus* reveals that the pathogenicity of *B. anthracis* is due to the maintenance of two plasmids, pX01 and pX02, which encode virulence factors.

Viral Genomes

Viral genomes exhibit significant diversity in structure. Some viruses have genomes that consist of DNA as their genetic material. This DNA may be single stranded, as exemplified by human parvoviruses, or double stranded, as seen in the herpesviruses and poxviruses. Additionally, although all cellular life uses DNA as its genetic material, some viral genomes are made of either single-stranded or double-stranded RNA molecules, as we have discussed. Viral genomes are typically smaller than most bacterial genomes, encoding only a few genes, because they rely on their hosts to carry out many of the functions required for their replication. The diversity of viral genome structures and their implications for viral replication life cycles are discussed in more detail in The Viral Life Cycle.

Genome Size Matters

There is great variation in size of genomes among different organisms. Most eukaryotes maintain multiple chromosomes; humans, for example have 23 pairs, giving them 46 chromosomes. Despite being large at 3 billion base pairs, the human genome is far from the largest genome. Plants often maintain very large genomes, up to 150 billion base pairs, and commonly are polyploid, having multiple copies of each chromosome.

The size of bacterial genomes also varies considerably, although they tend to be smaller than eukaryotic genomes (Figure **12.4.5**). Some bacterial genomes may be as small as only 112,000 base pairs. Often, the size of a bacterium's genome directly relates to how much the bacterium depends on its host for survival. When a bacterium relies on the host cell to carry out certain functions, it loses the genes encoding the abilities to carry out those functions itself. These types of bacterial endosymbionts are reminiscent of the prokaryotic origins of mitochondria and chloroplasts.

From a clinical perspective, obligate intracellular pathogens also tend to have small genomes (some around 1 million base pairs). Because host cells supply most of their nutrients, they tend to have a reduced number of genes encoding metabolic functions. Due to their small sizes, the genomes of organisms like *Mycoplasma genitalium* (580,000 base pairs), *Chlamydia trachomatis* (1.0 million), *Rickettsia prowazekii* (1.1 million), and *Treponema pallidum* (1.1 million) were some of the earlier

bacterial genomes sequenced. Respectively, these pathogens cause urethritis and pelvic inflammation, chlamydia, typhus, and syphilis.

Whereas obligate intracellular pathogens have unusually small genomes, other bacteria with a great variety of metabolic and enzymatic capabilities have unusually large bacterial genomes. *Pseudomonas aeruginosa*, for example, is a bacterium commonly found in the environment and is able to grow on a wide range of substrates. Its genome contains 6.3 million base pairs, giving it a high metabolic ability and the ability to produce virulence factors that cause several types of opportunistic infections.

Interestingly, there has been significant variability in genome size in viruses as well, ranging from 3,500 base pairs to 2.5 million base pairs, significantly exceeding the size of many bacterial genomes. The great variation observed in viral genome sizes further contributes to the great diversity of viral genome characteristics already discussed.

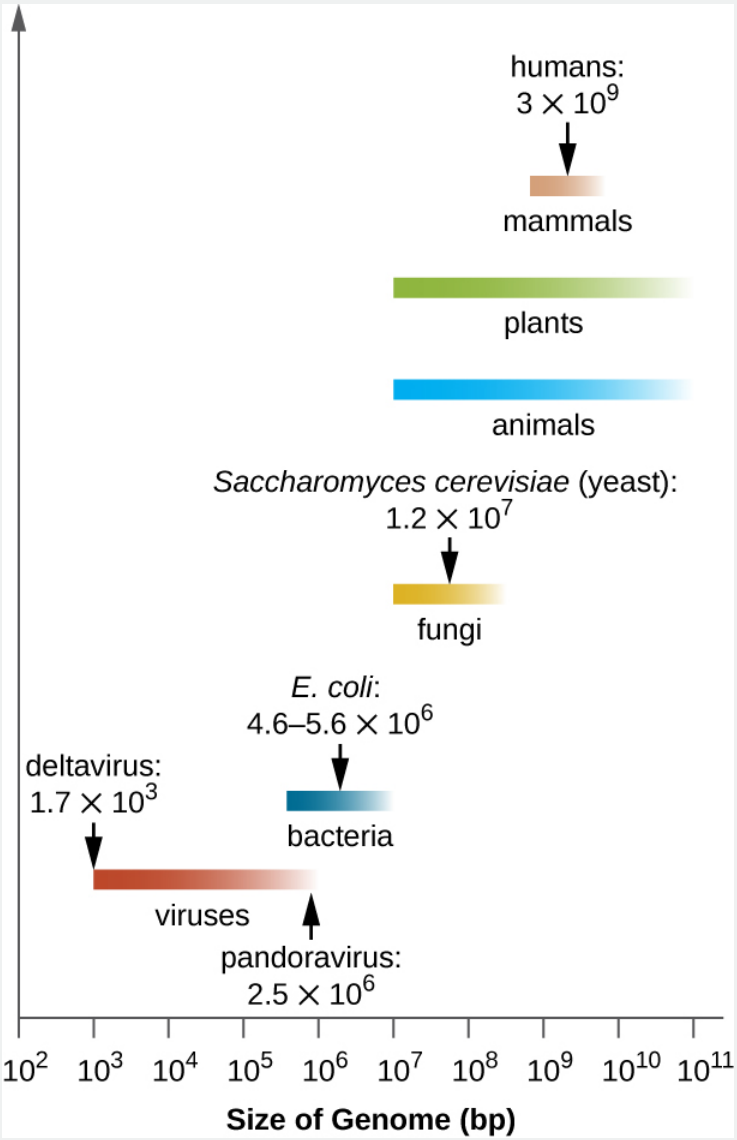


Figure **12.4.5**: There is great variability as well as overlap among the genome sizes of various groups of organisms and viruses.

Link to Learning

Visit the genome database of the National Center for Biotechnology Information (NCBI) to see the genomes that have been sequenced and their sizes.

Key Concepts and Summary

- The entire genetic content of a cell is its **genome**.
- **Genes** code for proteins, or stable RNA molecules, each of which carries out a specific function in the cell.
- Although the **genotype** that a cell possesses remains constant, expression of genes is dependent on environmental conditions.
- A **phenotype** is the observable characteristics of a cell (or organism) at a given point in time and results from the complement of genes currently being used.
- The majority of genetic material is organized into **chromosomes** that contain the DNA that controls cellular activities.
- Prokaryotes are typically haploid, usually having a single circular chromosome found in the nucleoid. Eukaryotes are diploid; DNA is organized into multiple linear chromosomes found in the nucleus.
- Supercoiling and DNA packaging using DNA binding proteins allows lengthy molecules to fit inside a cell. Eukaryotes and archaea use histone proteins, and bacteria use different proteins with similar function.
- Prokaryotic and eukaryotic genomes both contain **noncoding DNA**, the function of which is not well understood. Some noncoding DNA appears to participate in the formation of small noncoding RNA molecules that influence gene expression; some appears to play a role in maintaining chromosomal structure and in DNA packaging.
- **Extrachromosomal DNA** in eukaryotes includes the chromosomes found within organelles of prokaryotic origin (mitochondria and chloroplasts) that evolved by endosymbiosis. Some viruses may also maintain themselves extrachromosomally.
- Extrachromosomal DNA in prokaryotes is commonly maintained as **plasmids** that encode a few nonessential genes that may be helpful under specific conditions. Plasmids can be spread through a bacterial community by horizontal gene transfer.
- Viral genomes show extensive variation and may be composed of either RNA or DNA, and may be either double or single stranded.

Footnotes

1. 1 National Human Genome Research Institute. “The Human Genome Project Completion: Frequently Asked Questions.” https://www.genome.gov/11006943. Accessed June 10, 2016
2. 2 H. Bierne et al. “Epigenetics and Bacterial Infections.” *Cold Spring Harbor Perspectives in Medicine* 2 no. 12 (2012):a010272.
3. 3 R.J. Taft et al. “The Relationship between Non-Protein-Coding DNA and Eukaryotic Complexity.” *Bioessays* 29 no. 3 (2007):288–299.
4. 4 N. Ivanova et al. “Genome Sequence of *Bacillus cereus* and Comparative Analysis with *Bacillus anthracis.*” *Nature* 423 no. 6935 (2003):87–91.

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12.E: Biochemistry of the Genome (Exercises)

11.1: Using Microbiology to Discover the Secrets of Life

DNA was discovered and characterized long before its role in heredity was understood. Microbiologists played significant roles in demonstrating that DNA is the hereditary information found within cells. In the 1850s and 1860s, Gregor Mendel experimented with true-breeding garden peas to demonstrate the heritability of specific observable traits. In 1869, Friedrich Miescher isolated and purified a compound rich in phosphorus from the nuclei of white blood cells; he named the compound nuclein.

Multiple Choice

Frederick Griffith infected mice with a combination of dead R and live S bacterial strains. What was the outcome, and why did it occur?

A. The mice will live. Transformation was not required.
B. The mice will die. Transformation of genetic material from R to S was required.
C. The mice will live. Transformation of genetic material from S to R was required.
D. The mice will die. Transformation was not required.

Answer

D

Why was the alga *Acetabularia* a good model organism for Joachim Hämmerling to use to identify the location of genetic material?

A. It lacks a nuclear membrane.
B. It self-fertilizes.
C. It is a large, asymmetrical, single cell easy to see with the naked eye.
D. It makes a protein capsid.

Answer

C

Which of the following best describes the results from Hershey and Chase's experiment using bacterial viruses with ^{35}S-labeled proteins or ^{32}P-labeled DNA that are consistent with protein being the molecule responsible for hereditary?

A. After infection with the ^{35}S-labeled viruses and centrifugation, only the pellet would be radioactive.
B. After infection with the ^{35}S-labeled viruses and centrifugation, both the pellet and the supernatant would be radioactive.
C. After infection with the ^{32}P-labeled viruses and centrifugation, only the pellet would be radioactive.
D. After infection with the ^{32}P-labeled viruses and centrifugation, both the pellet and the supernatant would be radioactive.

Answer

A

Which method did Morgan and colleagues use to show that hereditary information was carried on chromosomes?

A. statistical predictions of the outcomes of crosses using true-breeding parents
B. correlations between microscopic observations of chromosomal movement and the characteristics of offspring
C. transformation of nonpathogenic bacteria to pathogenic bacteria
D. mutations resulting in distinct defects in metabolic enzymatic pathways

Answer

B

According to Beadle and Tatum's "one gene–one enzyme" hypothesis, which of the following enzymes will eliminate the transformation of hereditary material from pathogenic bacteria to nonpathogenic bacteria?

A. carbohydrate-degrading enzymes
B. proteinases
C. ribonucleases
D. deoxyribonucleases

Answer

D

Fill in the Blank

The element _____ is unique to nucleic acids compared with other macromolecules.

Answer

phosphorus

In the late 1800s and early 1900s, the macromolecule thought to be responsible for heredity was _____.

Answer

protein

Short Answer

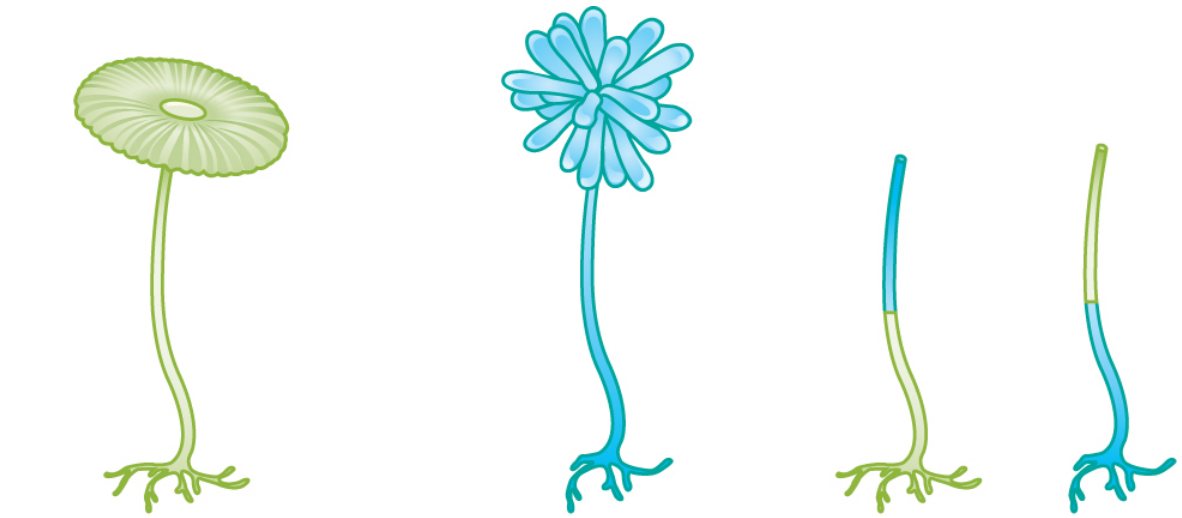
Why do bacteria and viruses make good model systems for various genetic studies?

Why was nucleic acid disregarded for so long as the molecule responsible for the transmission of hereditary information?

Bacteriophages inject their genetic material into host cells, whereas animal viruses enter host cells completely. Why was it important to use a bacteriophage in the Hershey–Chase experiment rather than an animal virus?

Critical Thinking

In the figure shown, if the nuclei were contained within the stalks of *Acetabularia*, what types of caps would you expect from the pictured grafts?



Why are Hershey and Chase credited with identifying DNA as the carrier of heredity even though DNA had been discovered many years before?

11.2: Structure and Function of DNA

Nucleic acids are composed of nucleotides, each of which contains a pentose sugar, a phosphate group, and a nitrogenous base. Deoxyribonucleotides within DNA contain deoxyribose as the pentose sugar. DNA contains the pyrimidines cytosine and thymine,

and the purines adenine and guanine. Nucleotides are linked together by phosphodiester bonds between the 5′ phosphate group of one nucleotide and the 3′ hydroxyl group of another.

Multiple Choice

Which of the following is not found within DNA?

A. thymine
B. phosphodiester bonds
C. complementary base pairing
D. amino acids

Answer

D

If 30% of the bases within a DNA molecule are adenine, what is the percentage of thymine?

A. 20%
B. 25%
C. 30%
D. 35%

Answer

C

Which of the following statements about base pairing in DNA is incorrect?

A. Purines always base pairs with pyrimidines.
B. Adenine binds to guanine.
C. Base pairs are stabilized by hydrogen bonds.
D. Base pairing occurs at the interior of the double helix.

Answer

B

If a DNA strand contains the sequence 5′-ATTCCGGATCGA-3′, which of the following is the sequence of the complementary strand of DNA?

A. 5′-TAAGGCCTAGCT-3′
B. 5′-ATTCCGGATCGA-3′
C. 3′-TAACCGGTACGT-5′
D. 5′-TCGATCCGGAAT-3′

Answer

D

During denaturation of DNA, which of the following happens?

A. Hydrogen bonds between complementary bases break.
B. Phosphodiester bonds break within the sugar-phosphate backbone.
C. Hydrogen bonds within the sugar-phosphate backbone break.
D. Phosphodiester bonds between complementary bases break.

Answer

A.

Fill in the Blank

The end of a nucleic acid strand with a free phosphate group is called the _____.

Answer

5′ end

True/False

The work of Rosalind Franklin and R.G. Gosling was important in demonstrating the helical nature of DNA.

Answer

True

The A-T base pair has more hydrogen bonding than the C-G base pair.

Answer

False

Short Answer

What is the role of phosphodiester bonds within the sugar-phosphate backbone of DNA?

What is meant by the term “antiparallel?”

Why is DNA with a high GC content more difficult to denature than that with a low GC content?

Critical Thinking

A certain DNA sample is found to have a makeup consisting of 22% thymine. Use Chargaff’s rules to fill in the percentages for the other three nitrogenous bases.

adenine	guanine	thymine	cytosine
___%	___%	22%	___%

In considering the structure of the DNA double helix, how would you expect the structure to differ if there was base pairing between two purines? Between two pyrimidines?

11.3: Structure and Function of RNA

Ribonucleic acid (RNA) is typically single stranded and contains ribose as its pentose sugar and the pyrimidine uracil instead of thymine. An RNA strand can undergo significant intramolecular base pairing to take on a three-dimensional structure. There are three main types of RNA, all involved in protein synthesis. Messenger RNA (mRNA) serves as the intermediary between DNA and the synthesis of protein products during translation.

Multiple Choice

Which of the following types of RNA codes for a protein?

A. dsRNA
B. mRNA
C. rRNA
D. tRNA

Answer

B

A nucleic acid is purified from a mixture. The molecules are relatively small, contain uracil, and most are covalently bound to an amino acid. Which of the following was purified?

A. DNA
B. mRNA
C. rRNA
D. tRNA

Answer

D

Which of the following types of RNA is known for its catalytic abilities?

A. dsRNA
B. mRNA
C. rRNA
D. tRNA

Answer

C

Ribosomes are composed of rRNA and what other component?

A. protein
B. polypeptides
C. DNA
D. mRNA

Answer

A

Which of the following may use RNA as its genome?

A. a bacterium
B. an archaeon
C. a virus
D. a eukaryote

Answer

C

Matching

Match the correct molecule with its description:

___tRNA	A. is a major component of ribosome
___rRNA	B. is a copy of the information in a gene
___mRNA	C. carries an amino acid to the ribosome

Answer

C, A, B

True/False

Ribosomes are composed mostly of RNA.

Answer

True

Double-stranded RNA is commonly found inside cells.

Answer

False

Short Answer

What are the differences between DNA nucleotides and RNA nucleotides?

How is the information stored within the base sequence of DNA used to determine a cell's properties?

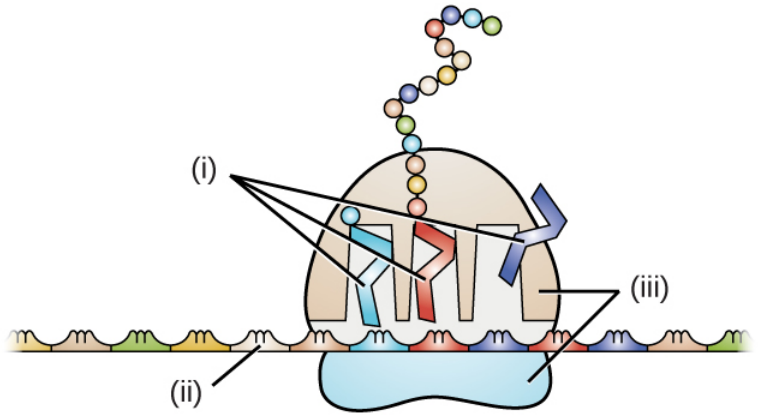
How do complementary base pairs contribute to intramolecular base pairing within an RNA molecule?

If an antisense RNA has the sequence 5′AUUCGAAUGC3′, what is the sequence of the mRNA to which it will bind? Be sure to label the 5′ and 3′ ends of the molecule you draw.

Why does double-stranded RNA (dsRNA) stimulate RNA interference?

Critical Thinking

Identify the location of mRNA, rRNA, and tRNA in the figure.



Why does it make sense that tRNA and rRNA molecules are more stable than mRNA molecules?

11.4: The Structure and Function of Cellular Genomes

The entire genetic content of a cell is its genome. Genes code for proteins, or stable RNA molecules, each of which carries out a specific function in the cell. Although the genotype that a cell possesses remains constant, expression of genes is dependent on environmental conditions. A phenotype is the observable characteristics of a cell (or organism) at a given point in time and results from the complement of genes currently being used.

Multiple Choice

Which of the following correctly describes the structure of the typical eukaryotic genome?

A. diploid
B. linear
C. singular
D. double stranded

Answer

A

Which of the following is typically found as part of the prokaryotic genome?

A. chloroplast DNA
B. linear chromosomes
C. plasmids
D. mitochondrial DNA

Answer

C

Serratia marcescens cells produce a red pigment at room temperature. The red color of the colonies is an example of which of the following?

A. genotype
B. phenotype
C. change in DNA base composition
D. adaptation to the environment

Answer

B

Which of the following genes would not likely be encoded on a plasmid?

A. genes encoding toxins that damage host tissue
B. genes encoding antibacterial resistance
C. gene encoding enzymes for glycolysis
D. genes encoding enzymes for the degradation of an unusual substrate

Answer

C

Histones are DNA binding proteins that are important for DNA packaging in which of the following?

A. double-stranded and single-stranded DNA viruses
B. archaea and bacteria
C. bacteria and eukaryotes
D. eukaryotes and archaea

Answer

D

True/False

Within an organism, phenotypes may change while genotypes remain constant.

Answer

True

Noncoding DNA has no biological purpose.

Answer

False

Fill in the Blank

Plasmids are typically transferred among members of a bacterial community by _____ gene transfer.

Answer

horizontal

Short Answer

What are some differences in chromosomal structures between prokaryotes and eukaryotes?

How do prokaryotes and eukaryotes manage to fit their lengthy DNA inside of cells? Why is this necessary?

What are some functions of noncoding DNA?

In the chromatin of eukaryotic cells, which regions of the chromosome would you expect to be more compact: the regions that contain genes being actively copied into RNA or those that contain inactive genes?

Critical Thinking

A new type of bacteriophage has been isolated and you are in charge of characterizing its genome. The base composition of the bacteriophage is A (15%), C (20%), T (35%), and G (30%). What can you conclude about the genome of the virus?

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CHAPTER OVERVIEW

13: DNA Replication, Transcription and Translation

In 1954, French scientist and future Nobel laureate Jacques Monod (1910–1976) famously said, “What is true in *E. coli* is true in the elephant,” suggesting that the biochemistry of life was maintained throughout evolution and is shared in all forms of known life. Since Monod’s famous statement, we have learned a great deal about the mechanisms of gene regulation, expression, and replication in living cells. All cells use DNA for information storage, share the same genetic code, and use similar mechanisms to replicate and express it. Although many aspects of genetics are universally shared, variations do exist among contemporary genetic systems. We now know that within the shared overall theme of the genetic mechanism, there are significant differences among the three domains of life: Eukarya, Archaea, and Bacteria. Additionally, viruses, cellular parasites but not themselves living cells, show dramatic variation in their genetic material and the replication and gene expression processes. Some of these differences have allowed us to engineer clinical tools such as antibiotics and antiviral drugs that specifically inhibit the reproduction of pathogens yet are harmless to their hosts.

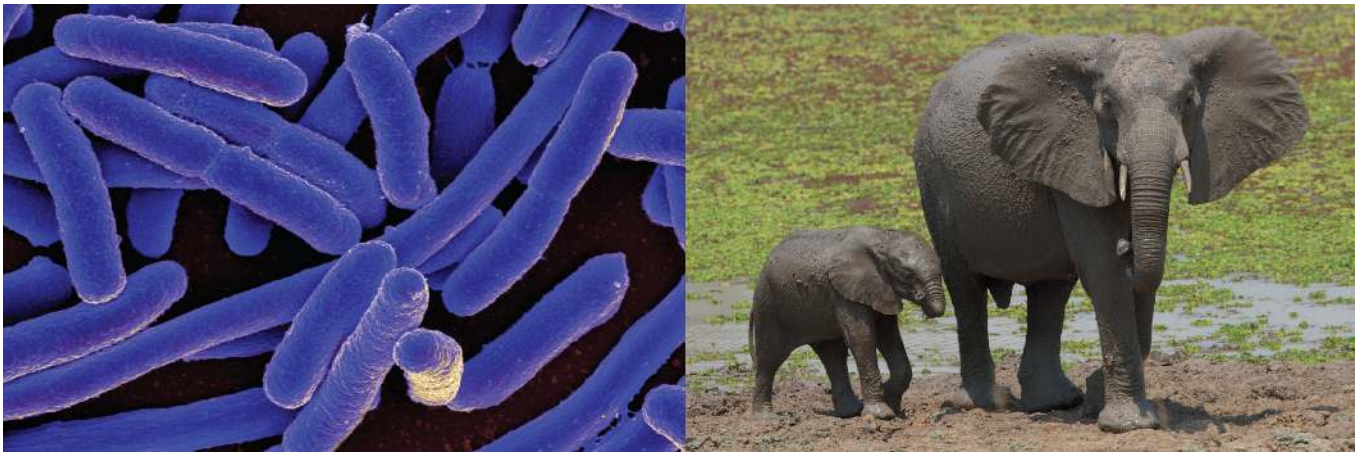


Figure 13.1: *Escherichia coli* (left) may not appear to have much in common with an elephant (right), but the genetic blueprints for these vastly different organisms are both encoded in DNA. (credit left: modification of work by NIAID; credit right: modification of work by Tom Lubbock)

13.1: What Are Genes?

13.2: DNA Replication

13.3: RNA Transcription

13.4: Protein Synthesis (Translation)

13.E: Mechanisms of Microbial Genetics (Exercises)

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13.1: What Are Genes?

Learning Objectives

- Explain the two functions of the genome
- Explain the meaning of the central dogma of molecular biology
- Differentiate between genotype and phenotype and explain how environmental factors influence phenotype

DNA serves two essential functions that deal with cellular information. First, DNA is the genetic material responsible for inheritance and is passed from parent to offspring for all life on earth. To preserve the integrity of this genetic information, DNA must be replicated with great accuracy, with minimal errors that introduce changes to the DNA sequence. A genome contains the full complement of DNA within a cell and is organized into smaller, discrete units called genes that are arranged on chromosomes and plasmids. The second function of DNA is to direct and regulate the construction of the proteins necessary to a cell for growth and reproduction in a particular cellular environment.

A gene is composed of DNA that is "read" or transcribed to produce an RNA molecule during the process of transcription. One major type of RNA molecule, called messenger RNA (mRNA), provides the information for the ribosome to catalyze protein synthesis in a process called translation. The processes of transcription and translation are collectively referred to as **gene expression**. Gene expression is the synthesis of a specific protein with a sequence of amino acids that is encoded in the gene. The flow of genetic information from DNA to RNA to protein is described by the **central dogma** (Figure 13.1.1). This central dogma of molecular biology further elucidates the mechanism behind Beadle and Tatum's "one gene-one enzyme" hypothesis (see Using Microbiology to Discover the Secrets of Life). Each of the processes of replication, transcription, and translation includes the stages of 1) initiation, 2) elongation (polymerization), and 3) termination. These stages will be described in more detail in this chapter.



Figure 13.1.1: The central dogma states that DNA encodes messenger RNA, which, in turn, encodes protein.

A cell's **genotype** is the full collection of genes it contains, whereas its **phenotype** is the set of observable characteristics that result from those genes. The phenotype is the product of the array of proteins being produced by the cell at a given time, which is influenced by the cell's genotype as well as interactions with the cell's environment. Genes code for proteins that have functions in the cell. Production of a specific protein encoded by an individual gene often results in a distinct phenotype for the cell compared with the phenotype without that protein. For this reason, it is also common to refer to the genotype of an individual gene and its phenotype. Although a cell's genotype remains constant, not all genes are used to direct the production of their proteins simultaneously. Cells carefully regulate expression of their genes, only using genes to make specific proteins when those proteins are needed (Figure 13.1.2).

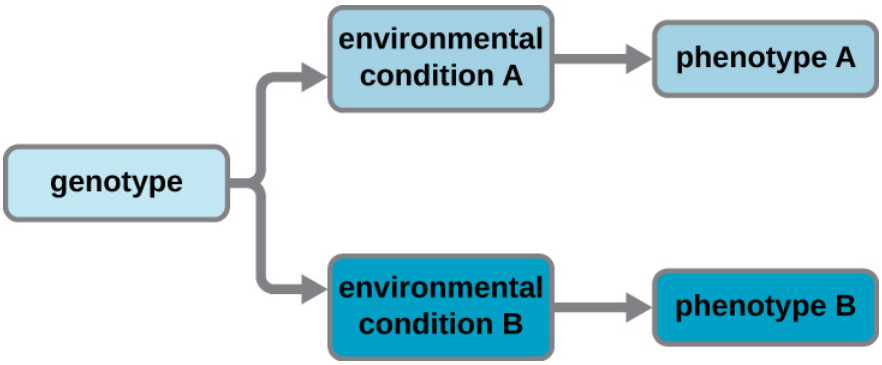
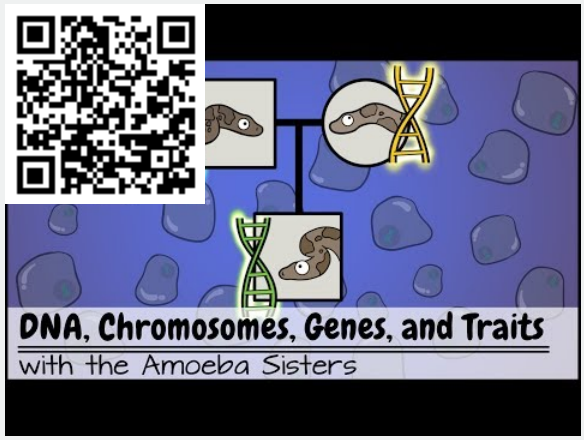


Figure 13.1.2: Phenotype is determined by the specific genes within a genotype that are expressed under specific conditions. Although multiple cells may have the same genotype, they may exhibit a wide range of phenotypes resulting from differences in patterns of gene expression in response to different environmental conditions.

DNA, Chromosomes, Genes, and Traits

Explore DNA structure/function, chromosomes, genes, and traits and how this relates to heredity! Video can replace old DNA structure & function video and in addition covers foundational concepts of heredity.





>

Use and Abuse of Genome Data

Why can some humans harbor opportunistic pathogens like *Haemophilus influenzae*, *Staphylococcus aureus*, or *Streptococcus pyogenes*, in their upper respiratory tracts but remain asymptomatic carriers, while other individuals become seriously ill when infected? There is evidence suggesting that differences in susceptibility to infection between patients may be a result, at least in part, of genetic differences between human hosts. For example, genetic differences in human leukocyte antigens (HLAs) and red blood cell antigens among hosts have been implicated in different immune responses and resulting disease progression from infection with *H. influenzae*.

Because the genetic interplay between pathogen and host may contribute to disease outcomes, understanding differences in genetic makeup between individuals may be an important clinical tool. Ecological genomics is a relatively new field that seeks to understand how the genotypes of different organisms interact with each other in nature. The field answers questions about how gene expression of one organism affects gene expression of another. Medical applications of ecological genomics will

focus on how pathogens interact with specific individuals, as opposed to humans in general. Such analyses would allow medical professionals to use knowledge of an individual's genotype to apply more individualized plans for treatment and prevention of disease.

With the advent of next-generation sequencing, it is relatively easy to obtain the entire genomic sequences of pathogens; a bacterial genome can be sequenced in as little as a day.[1] The speed and cost of sequencing the human genome has also been greatly reduced and, already, individuals can submit samples to receive extensive reports on their personal genetic traits, including ancestry and carrier status for various genetic diseases. As sequencing technologies progress further, such services will continue to become less expensive, more extensive, and quicker.

However, as this day quickly approaches, there are many ethical concerns with which society must grapple. For example, should genome sequencing be a standard practice for everybody? Should it be required by law or by employers if it will lower health-care costs? If one refuses genome sequencing, does he or she forfeit his or her right to health insurance coverage? For what purposes should the data be used? Who should oversee proper use of these data? If genome sequencing reveals predisposition to a particular disease, do insurance companies have the right to increase rates? Will employers treat an employee differently? Knowing that environmental influences also affect disease development, how should the data on the presence of a particular disease-causing allele in an individual be used ethically? The Genetic Information Nondiscrimination Act of 2008 (GINA) currently prohibits discriminatory practices based on genetic information by both health insurance companies and employers. However, GINA does not cover life, disability, or long-term care insurance policies. Clearly, all members of society must continue to engage in conversations about these issues so that such genomic data can be used to improve health care while simultaneously protecting an individual's rights.

Key Concepts and Summary

- DNA serves two important cellular functions: It is the genetic material passed from parent to offspring and it serves as the information to direct and regulate the construction of the proteins necessary for the cell to perform all of its functions.
- The **central dogma** states that DNA organized into genes specifies the sequences of messenger RNA (mRNA), which, in turn, specifies the amino acid sequence of proteins.
- The genotype of a cell is the full collection of genes a cell contains. Not all genes are used to make proteins simultaneously. The phenotype is a cell's observable characteristics resulting from the proteins it is producing at a given time under specific environmental conditions.

Footnotes

1. 1 D.J. Edwards, K.E. Holt. "Beginner's Guide to Comparative Bacterial Genome Analysis Using Next-Generation Sequence Data." *Microbial Informatics and Experimentation* 3 no. 1 (2013):2.

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13.2: DNA Replication

Learning Objectives

- Explain the meaning of semiconservative DNA replication
- Explain why DNA replication is bidirectional and includes both a leading and lagging strand
- Explain why Okazaki fragments are formed
- Describe the process of DNA replication and the functions of the enzymes involved

The elucidation of the structure of the double helix by James Watson and Francis Crick in 1953 provided a hint as to how DNA is copied during the process of **replication**. Separating the strands of the double helix would provide two templates for the synthesis of new complementary strands, but exactly how new DNA molecules were constructed was still unclear. In one model, **semiconservative replication**, the two strands of the double helix separate during DNA replication, and each strand serves as a template from which the new complementary strand is copied; after replication, each double-stranded DNA includes one parental or "old" strand and one "new" strand. There were two competing models also suggested: conservative and dispersive, which are shown in Figure **13.2.1**.

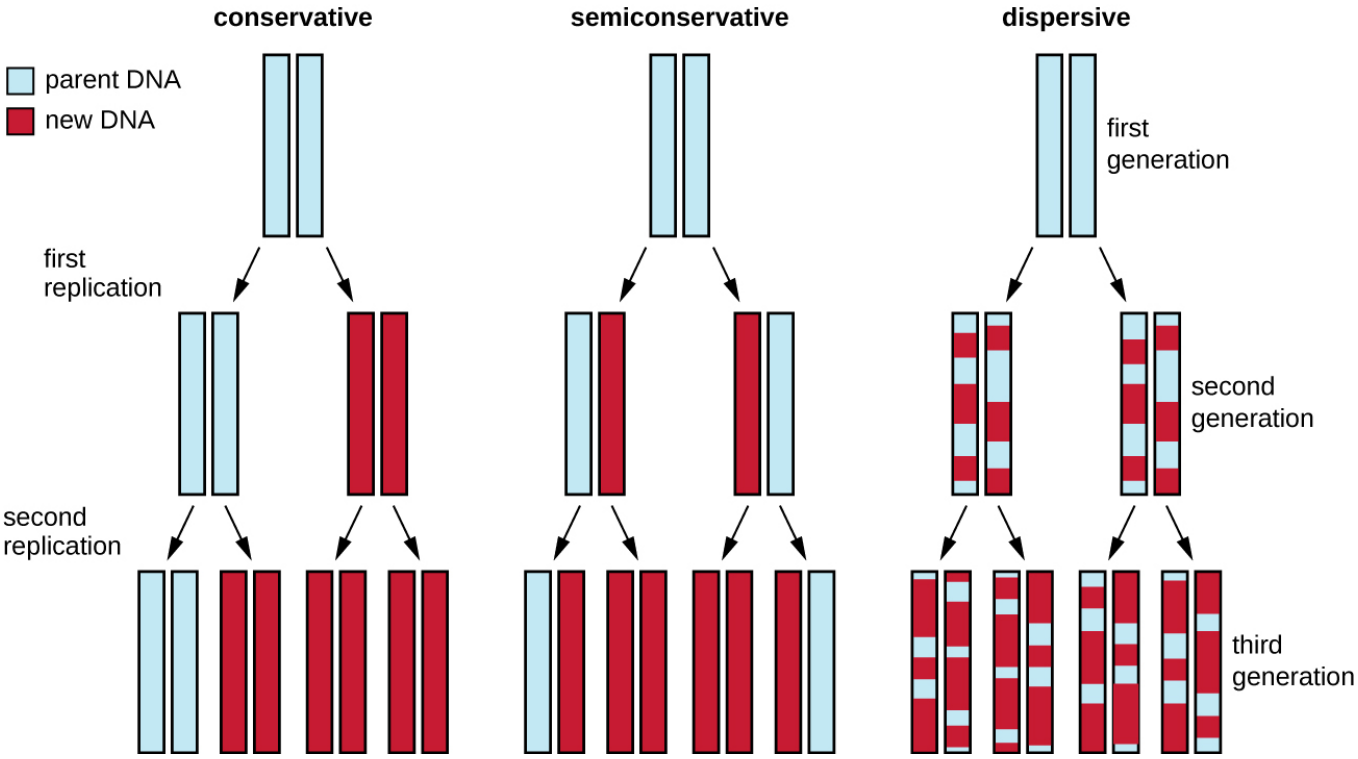


Figure **13.2.1**: There were three models suggested for DNA replication. In the conservative model, parental DNA strands (blue) remained associated in one DNA molecule while new daughter strands (red) remained associated in newly formed DNA molecules. In the semiconservative model, parental strands separated and directed the synthesis of a daughter strand, with each resulting DNA molecule being a hybrid of a parental strand and a daughter strand. In the dispersive model, all resulting DNA strands have regions of double-stranded parental DNA and regions of double-stranded daughter DNA.

Matthew Meselson (1930–) and Franklin Stahl (1929–) devised an experiment in 1958 to test which of these models correctly represents DNA replication (Figure **13.2.2**). They grew *E. coli* for several generations in a medium containing a "heavy" isotope of nitrogen (^{15}N) that was incorporated into nitrogenous bases and, eventually, into the DNA. This labeled the parental DNA. The *E. coli* culture was then shifted into a medium containing ^{14}N and allowed to grow for one generation. The cells were harvested and the DNA was isolated. The DNA was separated by ultracentrifugation, during which the DNA formed bands according to its density. DNA grown in ^{15}N would be expected to form a band at a higher density position than that grown in ^{14}N. Meselson and Stahl noted that after one generation of growth in ^{14}N, the single band observed was intermediate in position in between DNA of cells grown exclusively in ^{15}N or ^{14}N. This suggested either a semiconservative or dispersive mode of replication. Some cells were

allowed to grow for one more generation in ^{14}N and spun again. The DNA harvested from cells grown for two generations in ^{14}N formed two bands: one DNA band was at the intermediate position between ^{15}N and ^{14}N, and the other corresponded to the band of ^{14}N DNA. These results could only be explained if DNA replicates in a semiconservative manner. Therefore, the other two models were ruled out. As a result of this experiment, we now know that during DNA replication, each of the two strands that make up the double helix serves as a template from which new strands are copied. The new strand will be complementary to the parental or "old" strand. The resulting DNA molecules have the same sequence and are divided equally into the two daughter cells.

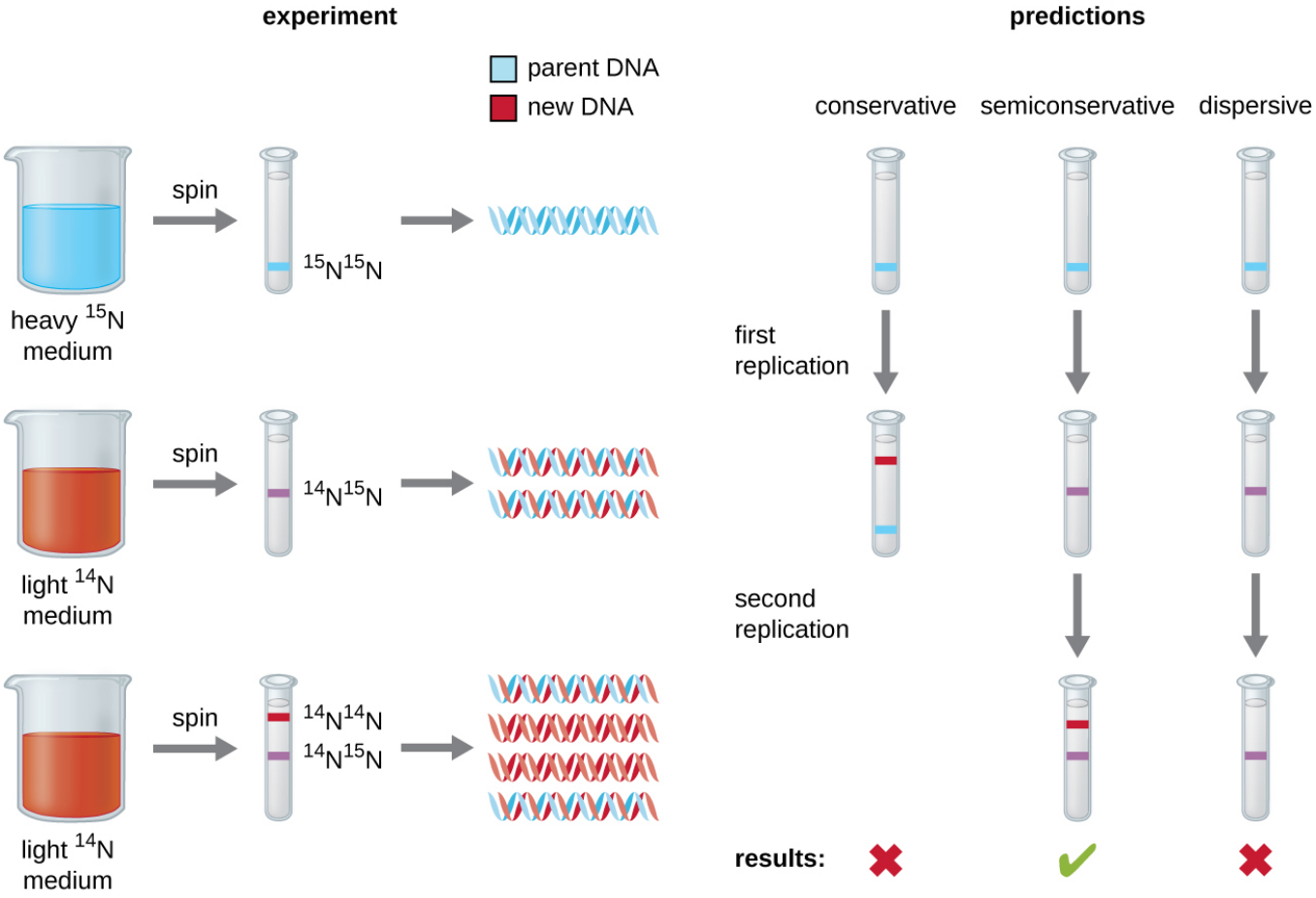


Figure **13.2.2**: Meselson and Stahl experimented with *E. coli* grown first in heavy nitrogen (^{15}N) then in ^{14}N. DNA grown in ^{15}N (blue band) was heavier than DNA grown in ^{14}N (red band), and sedimented to a lower level on ultracentrifugation. After one round of replication, the DNA sedimented halfway between the ^{15}N and ^{14}N levels (purple band), ruling out the conservative model of replication. After a second round of replication, the dispersive model of replication was ruled out. These data supported the semiconservative replication model.

DNA Replication in Bacteria

DNA replication has been well studied in bacteria primarily because of the small size of the genome and the mutants that are available. *E. coli* has 4.6 million base pairs (Mbp) in a single circular chromosome and all of it is replicated in approximately 42 minutes, starting from a single origin of replication and proceeding around the circle bidirectionally (i.e., in both directions). This means that approximately 1000 nucleotides are added per second. The process is quite rapid and occurs with few errors.

DNA replication uses a large number of proteins and enzymes (Table **13.2.1**). One of the key players is the enzyme **DNA polymerase**, also known as DNA pol. In bacteria, three main types of DNA polymerases are known: DNA pol I, DNA pol II, and DNA pol III. It is now known that DNA pol III is the enzyme required for DNA synthesis; DNA pol I and DNA pol II are primarily required for repair. DNA pol III adds deoxyribonucleotides each complementary to a nucleotide on the template strand, one by one to the 3'-OH group of the growing DNA chain. The addition of these nucleotides requires energy. This energy is present in the

bonds of three phosphate groups attached to each nucleotide (a triphosphate nucleotide), similar to how energy is stored in the phosphate bonds of adenosine triphosphate (ATP) (Figure **13.2.3**). When the bond between the phosphates is broken and diphosphate is released, the energy released allows for the formation of a covalent phosphodiester bond by dehydration synthesis between the incoming nucleotide and the free 3'-OH group on the growing DNA strand.

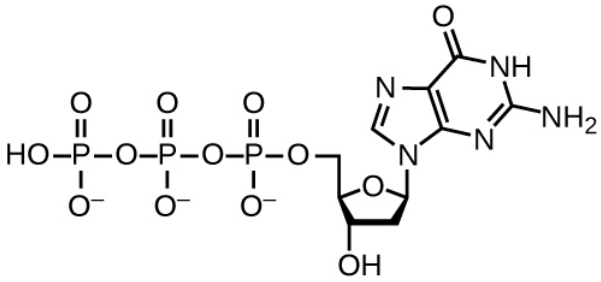
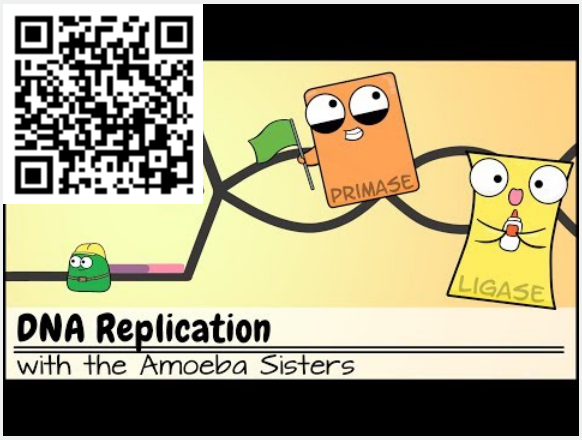


Figure **13.2.3**: This structure shows the guanosine triphosphate deoxyribonucleotide that is incorporated into a growing DNA strand by cleaving the two end phosphate groups from the molecule and transferring the energy to the sugar phosphate bond. The other three nucleotides form analogous structures.

Video: DNA Replication

Explore the steps of DNA replication, the enzymes involved, and the difference between the leading and lagging strand!



Initiation

The **initiation of replication** occurs at specific nucleotide sequence called the **origin of replication**, where various proteins bind to begin the replication process. *E. coli* has a single origin of replication (as do most prokaryotes), called *oriC*, on its one chromosome. The origin of replication is approximately 245 base pairs long and is rich in adenine-thymine (AT) sequences.

Some of the proteins that bind to the origin of replication are important in making single-stranded regions of DNA accessible for replication. Chromosomal DNA is typically wrapped around histones (in eukaryotes and archaea) or histone-like proteins (in bacteria), and is supercoiled, or extensively wrapped and twisted on itself. This packaging makes the information in the DNA molecule inaccessible. However, enzymes called topoisomerases change the shape and supercoiling of the chromosome. For bacterial DNA replication to begin, the supercoiled chromosome is relaxed by **topoisomerase II**, also called **DNA gyrase**. An enzyme called **helicase** then separates the DNA strands by breaking the hydrogen bonds between the nitrogenous base pairs. Recall that AT sequences have fewer hydrogen bonds and, hence, have weaker interactions than guanine-cytosine (GC) sequences. These enzymes require ATP hydrolysis. As the DNA opens up, Y-shaped structures called **replication forks** are formed. Two replication forks are formed at the origin of replication, allowing for bidirectional replication and formation of a structure that looks like a bubble when viewed with a transmission electron microscope; as a result, this structure is called a **replication bubble**. The DNA near each replication fork is coated with **single-stranded binding proteins** to prevent the single-stranded DNA from rewinding into a double helix.

Once single-stranded DNA is accessible at the origin of replication, DNA replication can begin. However, DNA pol III is able to add nucleotides only in the 5’ to 3’ direction (a new DNA strand can be only extended in this direction). This is because DNA polymerase requires a free 3’-OH group to which it can add nucleotides by forming a covalent phosphodiester bond between the 3’-OH end and the 5’ phosphate of the next nucleotide. This also means that it cannot add nucleotides if a free 3’-OH group is not available, which is the case for a single strand of DNA. The problem is solved with the help of an RNA sequence that provides the free 3’-OH end. Because this sequence allows the start of DNA synthesis, it is appropriately called the primer. The primer is five to 10 nucleotides long and complementary to the parental or template DNA. It is synthesized by **RNA primase**, which is an RNA polymerase. Unlike DNA polymerases, RNA polymerases do not need a free 3’-OH group to synthesize an RNA molecule. Now that the primer provides the free 3’-OH group, **DNA polymerase III** can now extend this RNA primer, adding DNA nucleotides one by one that are complementary to the template strand (Figure **13.2.1**).

Elongation

During **elongation in DNA replication**, the addition of nucleotides occurs at its maximal rate of about 1000 nucleotides per second. DNA polymerase III can only extend in the 5’ to 3’ direction, which poses a problem at the replication fork. The DNA double helix is antiparallel; that is, one strand is oriented in the 5’ to 3’ direction and the other is oriented in the 3’ to 5’ direction (see Structure and Function of DNA). During replication, one strand, which is complementary to the 3’ to 5’ parental DNA strand, is synthesized continuously toward the replication fork because polymerase can add nucleotides in this direction. This continuously synthesized strand is known as the **leading strand**. The other strand, complementary to the 5’ to 3’ parental DNA, grows away from the replication fork, so the polymerase must move back toward the replication fork to begin adding bases to a new primer, again in the direction away from the replication fork. It does so until it bumps into the previously synthesized strand and then it moves back again (Figure **13.2.4**). These steps produce small DNA sequence fragments known as **Okazaki fragments**, each separated by RNA primer. Okazaki fragments are named after the Japanese research team and married couple Reiji and Tsuneko Okazaki, who first discovered them in 1966. The strand with the Okazaki fragments is known as the **lagging strand**, and its synthesis is said to be discontinuous.

The leading strand can be extended from one primer alone, whereas the lagging strand needs a new primer for each of the short Okazaki fragments. The overall direction of the lagging strand will be 3’ to 5’, and that of the leading strand 5’ to 3’. A protein called the sliding clamp holds the DNA polymerase in place as it continues to add nucleotides. The sliding clamp is a ring-shaped protein that binds to the DNA and holds the polymerase in place. Beyond its role in initiation, topoisomerase also prevents the overwinding of the DNA double helix ahead of the replication fork as the DNA is opening up; it does so by causing temporary nicks in the DNA helix and then resealing it. As synthesis proceeds, the RNA primers are replaced by DNA. The primers are removed by the **exonuclease** activity of **DNA polymerase I**, and the gaps are filled in. The nicks that remain between the newly synthesized DNA (that replaced the RNA primer) and the previously synthesized DNA are sealed by the enzyme **DNA ligase** that

catalyzes the formation of covalent phosphodiester linkage between the 3’-OH end of one DNA fragment and the 5’ phosphate end of the other fragment, stabilizing the sugar-phosphate backbone of the DNA molecule.

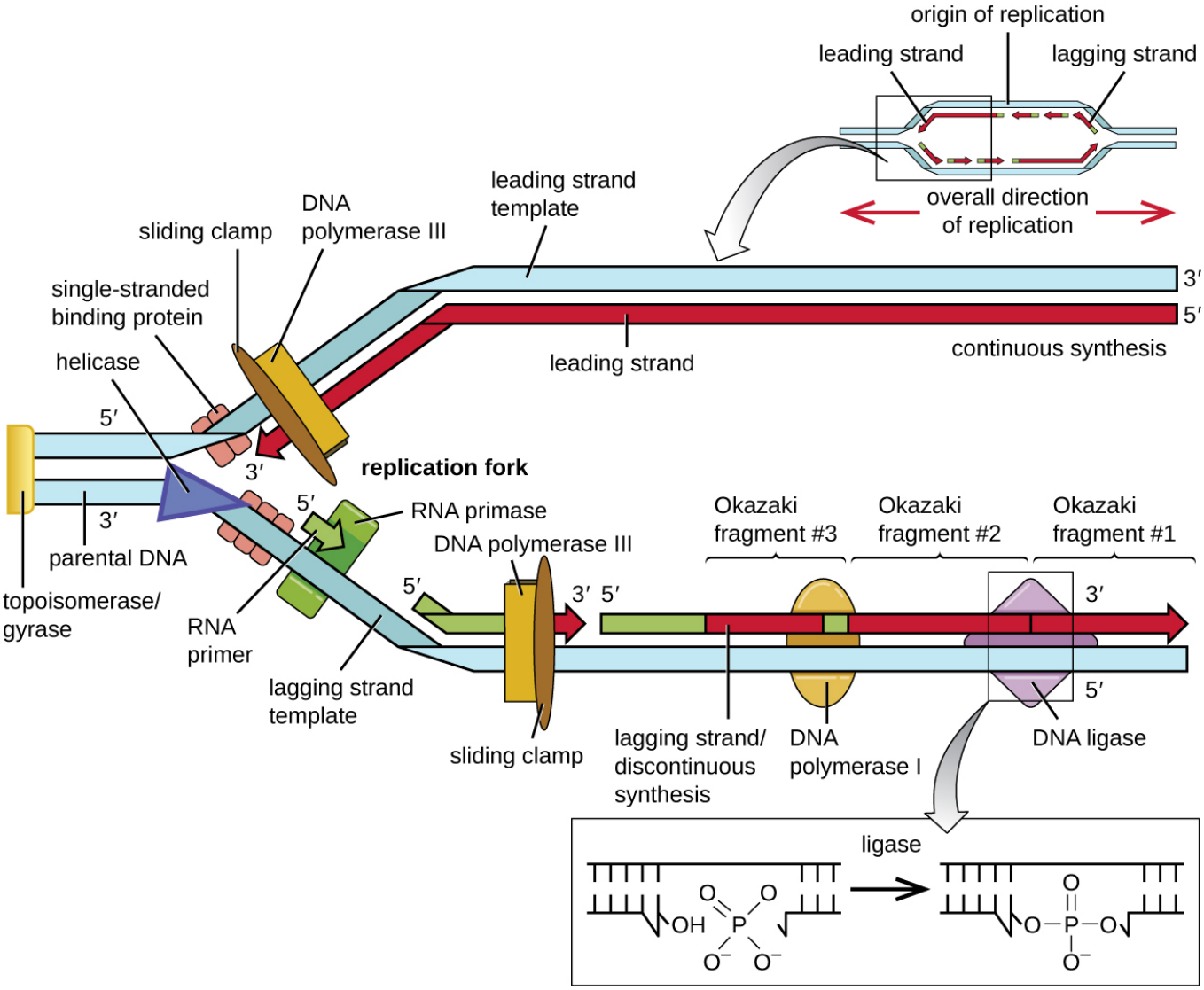


Figure 13.2.4: At the origin of replication, topoisomerase II relaxes the supercoiled chromosome. Two replication forks are formed by the opening of the double-stranded DNA at the origin, and helicase separates the DNA strands, which are coated by single-stranded binding proteins to keep the strands separated. DNA replication occurs in both directions. An RNA primer complementary to the parental strand is synthesized by RNA primase and is elongated by DNA polymerase III through the addition of nucleotides to the 3’-OH end. On the leading strand, DNA is synthesized continuously, whereas on the lagging strand, DNA is synthesized in short stretches called Okazaki fragments. RNA primers within the lagging strand are removed by the exonuclease activity of DNA polymerase I, and the Okazaki fragments are joined by DNA ligase.

Termination

Once the complete chromosome has been replicated, **termination of DNA replication** must occur. Although much is known about initiation of replication, less is known about the termination process. Following replication, the resulting complete circular genomes of prokaryotes are concatenated, meaning that the circular DNA chromosomes are interlocked and must be separated from each other. This is accomplished through the activity of bacterial topoisomerase IV, which introduces double-stranded breaks into DNA molecules, allowing them to separate from each other; the enzyme then reseals the circular chromosomes. The resolution of concatemers is an issue unique to prokaryotic DNA replication because of their circular chromosomes. Because both bacterial DNA gyrase and topoisomerase IV are distinct from their eukaryotic counterparts, these enzymes serve as targets for a class of antimicrobial drugs called quinolones.

Table 13.2.1: The Molecular Machinery Involved in Bacterial DNA Replication

Enzyme or Factor	Function
DNA pol I	Exonuclease activity removes RNA primer and replaces it with newly synthesized DNA
DNA pol III	Main enzyme that adds nucleotides in the 5’ to 3’ direction
Helicase	Opens the DNA helix by breaking hydrogen bonds between the nitrogenous bases
Ligase	Seals the gaps between the Okazaki fragments on the lagging strand to create one continuous DNA strand
Primase	Synthesizes RNA primers needed to start replication
Single-stranded binding proteins	Bind to single-stranded DNA to prevent hydrogen bonding between DNA strands, reforming double-stranded DNA
Sliding clamp	Helps hold DNA pol III in place when nucleotides are being added
Topoisomerase II (DNA gyrase)	Relaxes supercoiled chromosome to make DNA more accessible for the initiation of replication; helps relieve the stress on DNA when unwinding, by causing breaks and then resealing the DNA
Topoisomerase IV	Introduces single-stranded break into concatenated chromosomes to release them from each other, and then reseals the DNA

Key Concepts and Summary

- The DNA replication process is **semiconservative**, which results in two DNA molecules, each having one parental strand of DNA and one newly synthesized strand.
- In bacteria, the **initiation of replication** occurs at the **origin of replication**, where **supercoiled** DNA is unwound by **DNA gyrase**, made single-stranded by **helicase**, and bound by **single-stranded binding protein** to maintain its single-stranded state. **Primase** synthesizes a short RNA **primer**, providing a free 3’-OH group to which **DNA polymerase III** can add DNA nucleotides.
- During **elongation**, the **leading strand** of DNA is synthesized continuously from a single primer. The **lagging strand** is synthesized discontinuously in short **Okazaki fragments**, each requiring its own primer. The RNA primers are removed and replaced with DNA nucleotides by bacterial **DNA polymerase I**, and **DNA ligase** seals the gaps between these fragments.
- **Termination** of replication in bacteria involves the resolution of circular DNA concatemers by topoisomerase IV to release the two copies of the circular chromosome.

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13.3: RNA Transcription

Learning Objectives

- Explain how RNA is synthesized using DNA as a template
- Distinguish between transcription in prokaryotes and eukaryotes
- Interpret the DNA template strand to predict the sequence of the corresponding mRNA

During the process of **transcription**, the information encoded within the DNA sequence of one or more genes is transcribed into a strand of RNA, also called an **RNA transcript**. The resulting single-stranded RNA molecule, composed of ribonucleotides containing the bases adenine (A), cytosine (C), guanine (G), and uracil (U), acts as a mobile molecular copy of the original DNA sequence. Transcription in prokaryotes and in eukaryotes requires the DNA double helix to partially unwind in the region of RNA synthesis. The unwound region is called a **transcription bubble**. Transcription of a particular gene always proceeds from one of the two DNA strands that acts as a template, the so-called **antisense strand**. The RNA product is complementary to the template strand of DNA and is almost identical to the nontemplate DNA strand, or the **sense strand**. The only difference is that in RNA, all of the T nucleotides are replaced with U nucleotides; during RNA synthesis, U is incorporated when there is an A in the complementary antisense strand.

Transcription in Bacteria

Bacteria use the same RNA polymerase to transcribe all of their genes. Like DNA polymerase, **RNA polymerase** adds nucleotides one by one to the 3'-OH group of the growing nucleotide chain. One critical difference in activity between DNA polymerase and RNA polymerase is the requirement for a 3'-OH onto which to add nucleotides: DNA polymerase requires such a 3'-OH group, thus necessitating a primer, whereas RNA polymerase does not. During transcription, a ribonucleotide complementary to the DNA template strand is added to the growing RNA strand and a covalent phosphodiester bond is formed by dehydration synthesis between the new nucleotide and the last one added. In *E. coli*, RNA polymerase comprises six polypeptide subunits, five of which compose the polymerase core enzyme responsible for adding RNA nucleotides to a growing strand. The sixth subunit is known as sigma (σ). The σ factor enables RNA polymerase to bind to a specific promoter, thus allowing for the transcription of various genes. There are various σ factors that allow for transcription of various genes.

Initiation

The **initiation of transcription** begins at a **promoter**, a DNA sequence onto which the transcription machinery binds and initiates transcription. The nucleotide pair in the DNA double helix that corresponds to the site from which the first 5' RNA nucleotide is transcribed is the initiation site. Nucleotides preceding the initiation site are designated "upstream," whereas nucleotides following the initiation site are called "downstream" nucleotides. In most cases, promoters are located just upstream of the genes they regulate. Although promoter sequences vary among bacterial genomes, a few elements are conserved. At the –10 and –35 positions within the DNA prior to the initiation site (designated +1), there are two promoter consensus sequences, or regions that are similar across all promoters and across various bacterial species. The –10 consensus sequence, called the TATA box, is TATAAT. The –35 sequence is recognized and bound by σ.

Elongation

The **elongation in transcription** phase begins when the σ subunit dissociates from the polymerase, allowing the core enzyme to synthesize RNA complementary to the DNA template in a 5' to 3' direction at a rate of approximately 40 nucleotides per second. As elongation proceeds, the DNA is continuously unwound ahead of the core enzyme and rewound behind it (Figure 13.3.1).

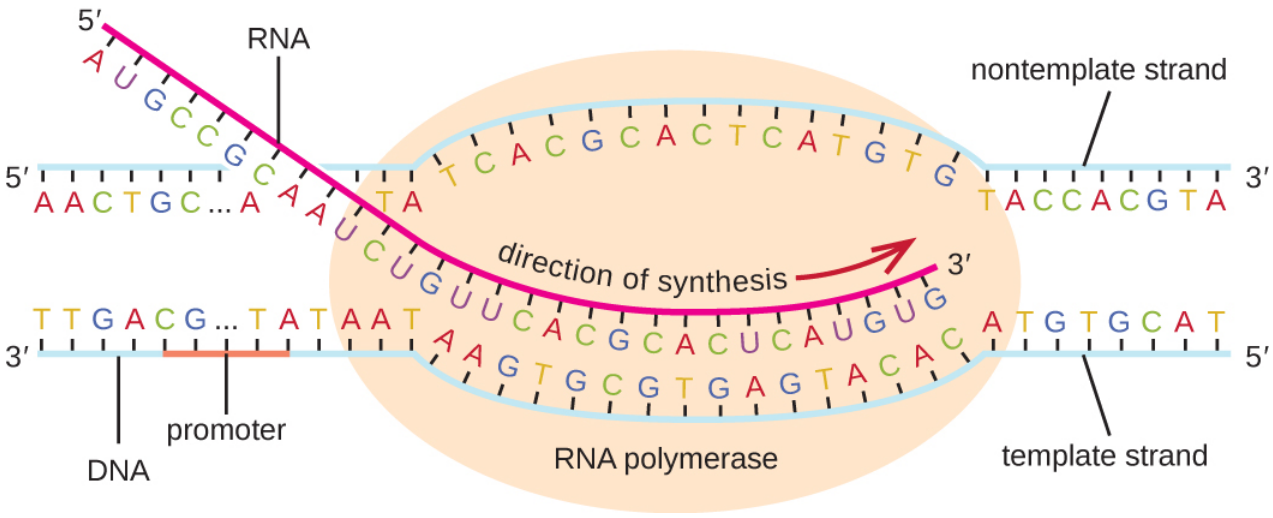


Figure **13.3.1**: During elongation, the bacterial RNA polymerase tracks along the DNA template, synthesizes mRNA in the 5' to 3' direction, and unwinds and rewinds the DNA as it is read.

Termination

Once a gene is transcribed, the bacterial polymerase must dissociate from the DNA template and liberate the newly made RNA. This is referred to as **termination of transcription**. The DNA template includes repeated nucleotide sequences that act as termination signals, causing RNA polymerase to stall and release from the DNA template, freeing the RNA transcript.

Transcription in Eukaryotes

Prokaryotes and eukaryotes perform fundamentally the same process of transcription, with a few significant differences (see Table **13.3.1**). Eukaryotes use three different polymerases, RNA polymerases I, II, and III, all structurally distinct from the bacterial RNA polymerase. Each transcribes a different subset of genes. Interestingly, archaea contain a single RNA polymerase that is more closely related to eukaryotic RNA polymerase II than to its bacterial counterpart. Eukaryotic mRNAs are also usually monocistronic, meaning that they each encode only a single polypeptide, whereas prokaryotic mRNAs of bacteria and archaea are commonly **polycistronic**, meaning that they encode multiple polypeptides.

The most important difference between prokaryotes and eukaryotes is the latter's membrane-bound nucleus, which influences the ease of use of RNA molecules for protein synthesis. With the genes bound in a nucleus, the eukaryotic cell must transport protein-encoding RNA molecules to the cytoplasm to be translated. Protein-encoding **primary transcripts**, the RNA molecules directly synthesized by RNA polymerase, must undergo several processing steps to protect these RNA molecules from degradation during the time they are transferred from the nucleus to the cytoplasm and translated into a protein. For example, eukaryotic mRNAs may last for several hours, whereas the typical prokaryotic mRNA lasts no more than 5 seconds.

The primary transcript (also called pre-mRNA) is first coated with RNA-stabilizing proteins to protect it from degradation while it is processed and exported out of the nucleus. The first type of processing begins while the primary transcript is still being synthesized; a special 7-methylguanosine nucleotide, called the **5' cap**, is added to the 5' end of the growing transcript. In addition to preventing degradation, factors involved in subsequent protein synthesis recognize the cap, which helps initiate translation by ribosomes. Once elongation is complete, another processing enzyme then adds a string of approximately 200 adenine nucleotides to the 3' end, called the **poly-A tail**. This modification further protects the pre-mRNA from degradation and signals to cellular factors that the transcript needs to be exported to the cytoplasm.

Eukaryotic genes that encode polypeptides are composed of coding sequences called **exons** (*ex*-on signifies that they are *ex*pressed) and intervening sequences called **introns** (*int*-ron denotes their *int*ervening role). Transcribed RNA sequences corresponding to introns do not encode regions of the functional polypeptide and are removed from the pre-mRNA during processing. It is essential that all of the intron-encoded RNA sequences are completely and precisely removed from a pre-mRNA before protein synthesis so that the exon-encoded RNA sequences are properly joined together to code for a functional polypeptide. If the process errs by even

a single nucleotide, the sequences of the rejoined exons would be shifted, and the resulting polypeptide would be nonfunctional. The process of removing intron-encoded RNA sequences and reconnecting those encoded by exons is called **RNA splicing** and is facilitated by the action of a **spliceosome** containing small nuclear ribonucleo proteins (snRNPs). Intron-encoded RNA sequences are removed from the pre-mRNA while it is still in the nucleus. Although they are not translated, introns appear to have various functions, including gene regulation and mRNA transport. On completion of these modifications, the mature transcript, the mRNA that encodes a polypeptide, is transported out of the nucleus, destined for the cytoplasm for translation. Introns can be spliced out differently, resulting in various exons being included or excluded from the final mRNA product. This process is known as alternative splicing. The advantage of alternative splicing is that different types of mRNA transcripts can be generated, all derived from the same DNA sequence. In recent years, it has been shown that some archaea also have the ability to splice their pre-mRNA.

Table **13.3.1**: Comparison of Transcription in Bacteria Versus Eukaryotes

Property	Bacteria	Eukaryotes
Number of polypeptides encoded per mRNA	Monocistronic or polycistronic	Exclusively monocistronic
Strand elongation	core + σ = holoenzyme	RNA polymerases I, II, or III
Addition of 5’ cap	No	Yes
Addition of 3’ poly-A tail	No	Yes
Splicing of pre-mRNA	No	Yes

Link to Learning

Visualize how mRNA splicing happens by watching the process in action in this video. See how introns are removed during RNA splicing here.

Key Concepts and Summary

- During **transcription**, the information encoded in DNA is used to make RNA.
- **RNA polymerase** synthesizes RNA, using the antisense strand of the DNA as template by adding complementary RNA nucleotides to the 3’ end of the growing strand.
- RNA polymerase binds to DNA at a sequence called a **promoter** during the **initiation of transcription**.
- Genes encoding proteins of related functions are frequently transcribed under the control of a single promoter in prokaryotes, resulting in the formation of a **polycistronic mRNA** molecule that encodes multiple polypeptides.
- Unlike DNA polymerase, RNA polymerase does not require a 3’-OH group to add nucleotides, so a **primer** is not needed during initiation.
- **Termination of transcription** in bacteria occurs when the RNA polymerase encounters specific DNA sequences that lead to stalling of the polymerase. This results in release of RNA polymerase from the DNA template strand, freeing the **RNA transcript**.
- Eukaryotes have three different RNA polymerases. Eukaryotes also have monocistronic mRNA, each encoding only a single polypeptide.
- Eukaryotic primary transcripts are processed in several ways, including the addition of a **5’ cap** and a 3′-**poly-A tail**, as well as **splicing**, to generate a mature mRNA molecule that can be transported out of the nucleus and that is protected from degradation.

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13.4: Protein Synthesis (Translation)

Learning Objectives

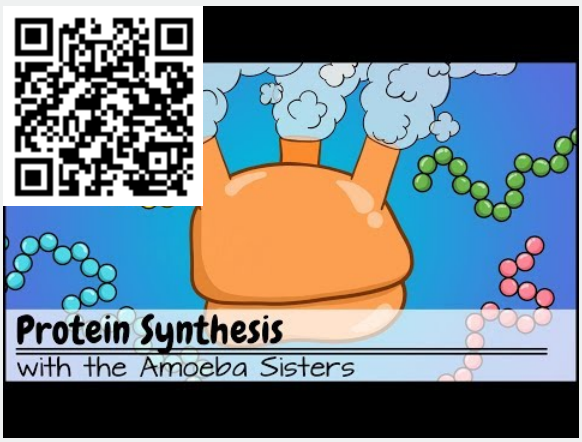
- Describe the genetic code and explain why it is considered almost universal
- Interpret a codon chart to determine the amino acid sequence encoded by an mRNA strand
- Describe the components of the translation machinery

The synthesis of proteins consumes more of a cell's energy than any other metabolic process. In turn, proteins account for more mass than any other macromolecule of living organisms. They perform virtually every function of a cell, serving as both functional (e.g., enzymes) and structural elements. The process of **translation**, or **protein synthesis**, the second part of gene expression, involves the decoding by a ribosome of an mRNA message into a polypeptide product.

Video: Protein Synthesis

Explore the steps of transcription and translation in protein synthesis! This video explains several reasons why proteins are so important before explaining the roles of mRNA, rRNA, and tRNA in the steps of protein synthesis! Expand details for contents and resources.





The Genetic Code

Translation of the mRNA template converts nucleotide-based genetic information into the “language” of amino acids to create a protein product. A protein sequence consists of 20 commonly occurring amino acids. Each amino acid is defined within the mRNA by a triplet of nucleotides called a **codon**. The relationship between an mRNA codon and its corresponding amino acid is called the **genetic code**.

The three-nucleotide code means that there is a total of 64 possible combinations (4^3, with four different nucleotides possible at each of the three different positions within the codon). This number is greater than the number of amino acids and a given amino acid is encoded by more than one codon (Figure 13.4.1). This redundancy in the genetic code is called **degeneracy**. Typically, whereas the first two positions in a codon are important for determining which amino acid will be incorporated into a growing polypeptide, the third position, called the **wobble position**, is less critical. In some cases, if the nucleotide in the third position is changed, the same amino acid is still incorporated.

Whereas 61 of the 64 possible triplets code for amino acids, three of the 64 codons do not code for an amino acid; they terminate protein synthesis, releasing the polypeptide from the translation machinery. These are called **stop codons** or **nonsense codons**. Another codon, AUG, also has a special function. In addition to specifying the amino acid methionine, it also typically serves as the **start codon** to initiate translation. The **reading frame**, the way nucleotides in mRNA are grouped into codons, for translation is set by the AUG start codon near the 5’ end of the mRNA. Each set of three nucleotides following this start codon is a codon in the mRNA message.

The genetic code is nearly universal. With a few exceptions, virtually all species use the same genetic code for protein synthesis, which is powerful evidence that all extant life on earth shares a common origin. However, unusual amino acids such as selenocysteine and pyrrolysine have been observed in archaea and bacteria. In the case of selenocysteine, the codon used is UGA (normally a stop codon). However, UGA can encode for selenocysteine using a stem-loop structure (known as the selenocysteine insertion sequence, or SECIS element), which is found at the 3’ untranslated region of the mRNA. Pyrrolysine uses a different stop codon, UAG. The incorporation of pyrrolysine requires the *pylS* gene and a unique transfer RNA (tRNA) with a CUA anticodon.

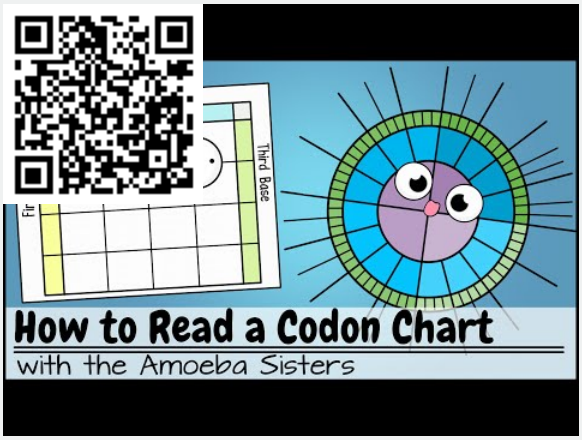
second letter

first letter	U	C	A	G	third letter
U	UUU, UUC: Phe UUA, UUG: Leu	UCU, UCC, UCA, UCG: Ser	UAU, UAC: Tyr UAA: stop UAG: stop	UGU, UGC: Cys UGA: stop UGG: Trp	U C A G
C	CUU, CUC, CUA, CUG: Leu	CCU, CCC, CCA, CCG: Pro	CAU, CAC: His CAA, CAG: Gln	CGU, CGC, CGA, CGG: Arg	U C A G
A	AUU, AUC, AUA: Ile AUG: Met	ACU, ACC, ACA, ACG: Thr	AAU, AAC: Asn AAA, AAG: Lys	AGU, AGC: Ser AGA, AGG: Arg	U C A G
G	GUU, GUC, GUA, GUG: Val	GCU, GCC, GCA, GCG: Ala	GAU, GAC: Asp GAA, GAG: Glu	GGU, GGC, GGA, GGG: Gly	U C A G

Figure 13.4.1: This figure shows the genetic code for translating each nucleotide triplet in mRNA into an amino acid or a termination signal in a nascent protein. The first letter of a codon is shown vertically on the left, the second letter of a codon is shown horizontally across the top, and the third letter of a codon is shown vertically on the right. (credit: modification of work by National Institutes of Health)

Video: How to Read a Codon Chart

Join the Amoeba Sisters as they show how to work through a rectangular and circular codon chart. Also, find our video companion under the topic "codon charts" on our handout page https://www.amoebasisters.com/handouts



The Protein Synthesis Machinery

In addition to the mRNA template, many molecules and macromolecules contribute to the process of translation. The composition of each component varies across taxa; for instance, ribosomes may consist of different numbers of ribosomal RNAs (rRNAs) and polypeptides depending on the organism. However, the general structures and functions of the protein synthesis machinery are comparable from bacteria to human cells. Translation requires the input of an mRNA template, ribosomes, tRNAs, and various enzymatic factors.

Ribosomes

A **ribosome** is a complex macromolecule composed of catalytic rRNAs (called **ribozymes**) and structural rRNAs, as well as many distinct polypeptides. Mature rRNAs make up approximately 50% of each ribosome. **Prokaryotes** have **70S ribosomes**, whereas **eukaryotes** have **80S ribosomes** in the cytoplasm and rough endoplasmic reticulum, and 70S ribosomes in mitochondria and chloroplasts. Ribosomes dissociate into large and small subunits when they are not synthesizing proteins and reassociate during the **initiation of translation**. In *E. coli*, the **small subunit** is described as 30S (which contains the 16S rRNA subunit), and the **large subunit** is 50S (which contains the 5S and 23S rRNA subunits), for a total of 70S (Svedberg units are not additive). Eukaryote

ribosomes have a small 40S subunit (which contains the 18S rRNA subunit) and a large 60S subunit (which contains the 5S, 5.8S and 28S rRNA subunits), for a total of 80S. The small subunit is responsible for binding the mRNA template, whereas the large subunit binds tRNAs (discussed in the next subsection).

Each mRNA molecule is simultaneously translated by many ribosomes, all synthesizing protein in the same direction: reading the mRNA from 5' to 3' and synthesizing the polypeptide from the N terminus to the C terminus. The complete structure containing an mRNA with multiple associated ribosomes is called a **polyribosome** (or **polysome**). In both bacteria and archaea, before transcriptional termination occurs, each protein-encoding transcript is already being used to begin synthesis of numerous copies of the encoded polypeptide(s) because the processes of transcription and translation can occur concurrently, forming polyribosomes (Figure **13.4.2**). The reason why transcription and translation can occur simultaneously is because both of these processes occur in the same 5' to 3' direction, they both occur in the cytoplasm of the cell, and because the RNA transcript is not processed once it is transcribed. This allows a prokaryotic cell to respond to an environmental signal requiring new proteins very quickly. In contrast, in eukaryotic cells, simultaneous transcription and translation is not possible. Although polyribosomes also form in eukaryotes, they cannot do so until RNA synthesis is complete and the RNA molecule has been modified and transported out of the nucleus.

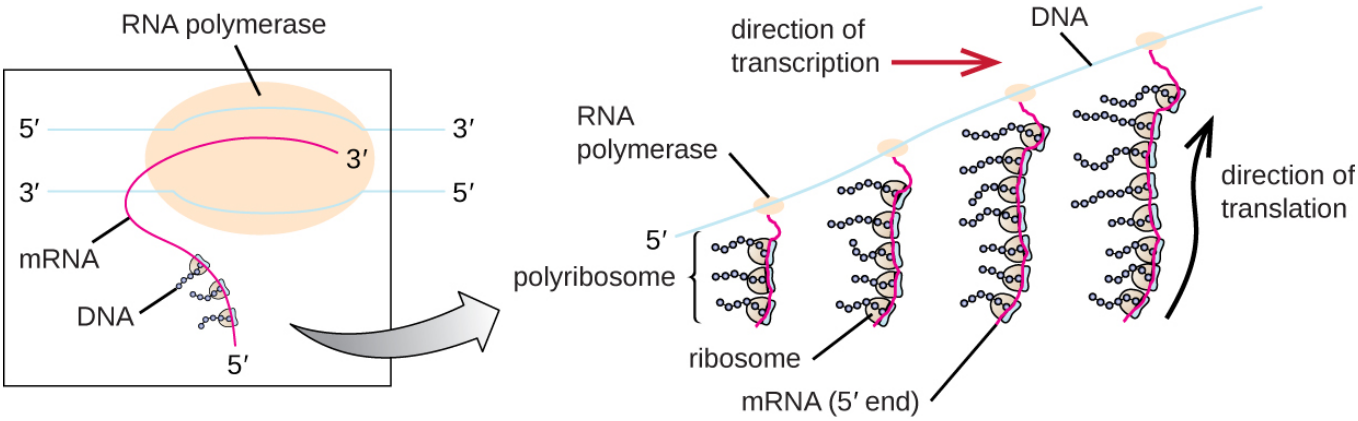


Figure **13.4.2**: In prokaryotes, multiple RNA polymerases can transcribe a single bacterial gene while numerous ribosomes concurrently translate the mRNA transcripts into polypeptides. In this way, a specific protein can rapidly reach a high concentration in the bacterial cell.

Transfer RNAs

Transfer RNAs (tRNAs) are structural RNA molecules and, depending on the species, many different types of tRNAs exist in the cytoplasm. Bacterial species typically have between 60 and 90 types. Serving as adaptors, each tRNA type binds to a specific codon on the mRNA template and adds the corresponding amino acid to the polypeptide chain. Therefore, tRNAs are the molecules that actually "translate" the language of RNA into the language of proteins. As the adaptor molecules of translation, it is surprising that tRNAs can fit so much specificity into such a small package. The tRNA molecule interacts with three factors: aminoacyl tRNA synthetases, ribosomes, and mRNA.

Mature tRNAs take on a three-dimensional structure when complementary bases exposed in the single-stranded RNA molecule hydrogen bond with each other (Figure **13.4.3**). This shape positions the amino-acid binding site, called the **CCA amino acid binding end**, which is a cytosine-cytosine-adenine sequence at the 3' end of the tRNA, and the anticodonat the other end. The **anticodon** is a three-nucleotide sequence that bonds with an mRNA codon through complementary base pairing.

An amino acid is added to the end of a tRNA molecule through the process of tRNA "charging," during which each tRNA molecule is linked to its correct or **cognate amino acid** by a group of enzymes called **aminoacyl tRNA synthetases**. At least one type of aminoacyl tRNA synthetase exists for each of the 20 amino acids. During this process, the amino acid is first activated by the addition of adenosine monophosphate (AMP) and then transferred to the tRNA, making it a **charged tRNA**, and AMP is released.

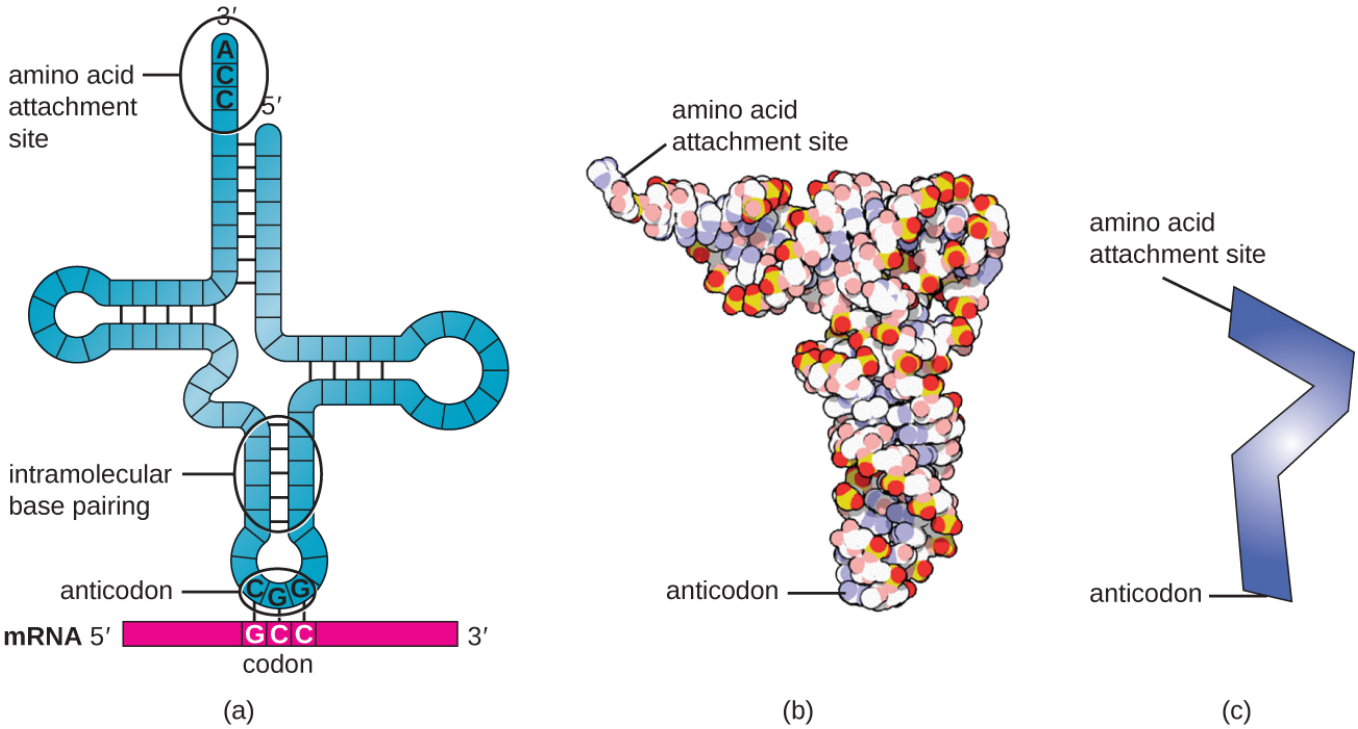


Figure **13.4.3**: (a) After folding caused by intramolecular base pairing, a tRNA molecule has one end that contains the anticodon, which interacts with the mRNA codon, and the CCA amino acid binding end. (b) A space-filling model is helpful for visualizing the three-dimensional shape of tRNA. (c) Simplified models are useful when drawing complex processes such as protein synthesis.

Key Concepts and Summary

- In **translation**, polypeptides are synthesized using mRNA sequences and cellular machinery, including tRNAs that match mRNA **codons** to specific amino acids and ribosomes composed of RNA and proteins that catalyze the reaction.
- The **genetic code** is **degenerate** in that several mRNA codons code for the same amino acids. The genetic code is almost universal among living organisms.
- Prokaryotic (70S) and cytoplasmic eukaryotic (80S) ribosomes are each composed of a large subunit and a small subunit of differing sizes between the two groups. Each subunit is composed of rRNA and protein. Organelle ribosomes in eukaryotic cells resemble prokaryotic ribosomes.
- Some 60 to 90 species of tRNA exist in bacteria. Each tRNA has a three-nucleotide **anticodon** as well as a binding site for a **cognate amino acid**. All tRNAs with a specific anticodon will carry the same amino acid.
- **Initiation** of translation occurs when the small ribosomal subunit binds with **initiation factors** and an initiator tRNA at the **start codon** of an mRNA, followed by the binding to the initiation complex of the large ribosomal subunit.
- In prokaryotic cells, the start codon codes for N-formyl-methionine carried by a special initiator tRNA. In eukaryotic cells, the start codon codes for methionine carried by a special initiator tRNA. In addition, whereas ribosomal binding of the mRNA in prokaryotes is facilitated by the Shine-Dalgarno sequence within the mRNA, eukaryotic ribosomes bind to the 5’ cap of the mRNA.
- During the **elongation** stage of translation, a **charged tRNA** binds to mRNA in the **A site** of the ribosome; a peptide bond is catalyzed between the two adjacent amino acids, breaking the bond between the first amino acid and its tRNA; the ribosome moves one codon along the mRNA; and the first tRNA is moved from the **P site** of the ribosome to the **E site** and leaves the ribosomal complex.
- **Termination** of translation occurs when the ribosome encounters a **stop codon**, which does not code for a tRNA. Release factors cause the polypeptide to be released, and the ribosomal complex dissociates.

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13.E: Mechanisms of Microbial Genetics (Exercises)

12.1: What Are Genes?

A gene is composed of DNA that is “read” or transcribed to produce an RNA molecule during the process of transcription. One major type of RNA molecule, called messenger RNA (mRNA), provides the information for the ribosome to catalyze protein synthesis in a process called translation. The processes of transcription and translation are collectively referred to as gene expression.

Multiple Choice

DNA does all but which of the following?

A. serves as the genetic material passed from parent to offspring
B. remains constant despite changes in environmental conditions
C. provides the instructions for the synthesis of messenger RNA
D. is read by ribosomes during the process of translation

Answer

D

According to the central dogma, which of the following represents the flow of genetic information in cells?

A. protein to DNA to RNA
B. DNA to RNA to protein
C. RNA to DNA to protein
D. DNA to protein to RNA

Answer

B

True/False

Cells are always producing proteins from every gene they possess.

Answer

False

Fill in the Blank

The process of making an RNA copy of a gene is called _____.

Answer

transcription

A cell’s _____ remains constant whereas its phenotype changes in response to environmental influences.

Answer

genotype or genome

Short Answer

Can two observably different cells have the same genotype? Explain.

Critical Thinking

A pure culture of an unknown bacterium was streaked onto plates of a variety of media. You notice that the colony morphology is strikingly different on plates of minimal media with glucose compared to that seen on trypticase soy agar plates. How can you

explain these differences in colony morphology?

12.2: DNA Replication

The DNA replication process is semiconservative, which results in two DNA molecules, each having one parental strand of DNA and one newly synthesized strand. In bacteria, the initiation of replication occurs at the origin of replication, where supercoiled DNA is unwound by DNA gyrase, made single-stranded by helicase, and bound by single-stranded binding protein to maintain its single-stranded state.

Multiple Choice

Which of the following is the enzyme that replaces the RNA nucleotides in a primer with DNA nucleotides?

A. DNA polymerase III
B. DNA polymerase I
C. primase
D. helicase

Answer

B

Which of the following is not involved in the initiation of replication?

A. ligase
B. DNA gyrase
C. single-stranded binding protein
D. primase

Answer

A

Which of the following enzymes involved in DNA replication is unique to eukaryotes?

A. helicase
B. DNA polymerase
C. ligase
D. telomerase

Answer

D

Which of the following would be synthesized using 5′-CAGTTCGGA-3′ as a template?

A. 3′-AGGCTTGAC-4′
B. 3′-TCCGAACTG-5′
C. 3′-GTCAAGCCT-5′
D. 3′-CAGTTCGGA-5′

Answer

C

Fill in the Blank

The enzyme responsible for relaxing supercoiled DNA to allow for the initiation of replication is called _____.

Answer

DNA gyrase or topoisomerase II

Unidirectional replication of a circular DNA molecule like a plasmid that involves nicking one DNA strand and displacing it while synthesizing a new strand is called _____.

Answer

rolling circle replication

True/False

More primers are used in lagging strand synthesis than in leading strand synthesis.

Answer

True

Short Answer

Why is primase required for DNA replication?

What is the role of single-stranded binding protein in DNA replication?

Below is a DNA sequence. Envision that this is a section of a DNA molecule that has separated in preparation for replication, so you are only seeing one DNA strand. Construct the complementary DNA sequence (indicating 5' and 3' ends).

DNA sequence: 3'-T A C T G A C T G A C G A T C-5'

Critical Thinking

Review Figure 11.2.1 and Figure 11.2.2. Why was it important that Meselson and Stahl continue their experiment to at least two rounds of replication after isotopic labeling of the starting DNA with ^{15}N, instead of stopping the experiment after only one round of replication?

If deoxyribonucleotides that lack the 3'-OH groups are added during the replication process, what do you expect will occur?

12.3: RNA Transcription

During the process of transcription, the information encoded within the DNA sequence of one or more genes is transcribed into a strand of RNA, also called an RNA transcript. The resulting single-stranded RNA molecule, composed of ribonucleotides containing the bases adenine, cytosine, guanine, and uracil, acts as a mobile molecular copy of the original DNA sequence. Transcription in prokaryotes and in eukaryotes requires the DNA double helix to partially unwind in the region of RNA synthesis.

Multiple Choice

During which stage of bacterial transcription is the σ subunit of the RNA polymerase involved?

A. initiation
B. elongation
C. termination
D. splicing

Answer

A

Which of the following components is involved in the initiation of transcription?

A. primer
B. origin
C. promoter
D. start codon

Answer

C

Which of the following is not a function of the 5’ cap and 3’ poly-A tail of a mature eukaryotic mRNA molecule?

A. to facilitate splicing
B. to prevent mRNA degradation
C. to aid export of the mature transcript to the cytoplasm
D. to aid ribosome binding to the transcript

Answer

A

Mature mRNA from a eukaryote would contain each of these features except which of the following?

A. exon-encoded RNA
B. intron-encoded RNA
C. 5’ cap
D. 3’ poly-A tail

Answer

B

Fill in the Blank

A _____ mRNA is one that codes for multiple polypeptides.

Answer

polycistronic

The protein complex responsible for removing intron-encoded RNA sequences from primary transcripts in eukaryotes is called the _____.

Answer

Spliceosome

Short Answer

What is the purpose of RNA processing in eukaryotes? Why don’t prokaryotes require similar processing?

Below is a DNA sequence. Envision that this is a section of a DNA molecule that has separated in preparation for transcription, so you are only seeing the antisense strand. Construct the mRNA sequence transcribed from this template.

Antisense DNA strand: 3’-T A C T G A C T G A C G A T C-5’

Critical Thinking

Predict the effect of an alteration in the sequence of nucleotides in the –35 region of a bacterial promoter.

12.4: Protein Synthesis (Translation)

The synthesis of proteins consumes more of a cell’s energy than any other metabolic process. In turn, proteins account for more mass than any other macromolecule of living organisms. They perform virtually every function of a cell, serving as both functional (e.g., enzymes) and structural elements. The process of translation, or protein synthesis, the second part of gene expression, involves the decoding by a ribosome of an mRNA message into a polypeptide product.

Multiple Choice

Which of the following is the name of the three-base sequence in the mRNA that binds to a tRNA molecule?

A. P site
B. codon

C. anticodon
D. CCA binding site

Answer

B

Which component is the last to join the initiation complex during the initiation of translation?

A. the mRNA molecule
B. the small ribosomal subunit
C. the large ribosomal subunit
D. the initiator tRNA

Answer

C

During elongation in translation, to which ribosomal site does an incoming charged tRNA molecule bind?

A. A site
B. P site
C. E site
D. B site

Answer

A

Which of the following is the amino acid that appears at the N-terminus of all newly translated prokaryotic and eukaryotic polypeptides?

A. tryptophan
B. methionine
C. selenocysteine
D. glycine

Answer

B

When the ribosome reaches a nonsense codon, which of the following occurs?

A. a methionine is incorporated
B. the polypeptide is released
C. a peptide bond forms
D. the A site binds to a charged tRNA

Answer

B

Fill in the Blank

The third position within a codon, in which changes often result in the incorporation of the same amino acid into the growing polypeptide, is called the _____.

Answer

wobble position

The enzyme that adds an amino acid to a tRNA molecule is called _____.

Answer

aminoacyl-tRNA synthetase

True/False

Each codon within the genetic code encodes a different amino acid.

Answer

False

Short Answer

Why does translation terminate when the ribosome reaches a stop codon? What happens?

How does the process of translation differ between prokaryotes and eukaryotes?

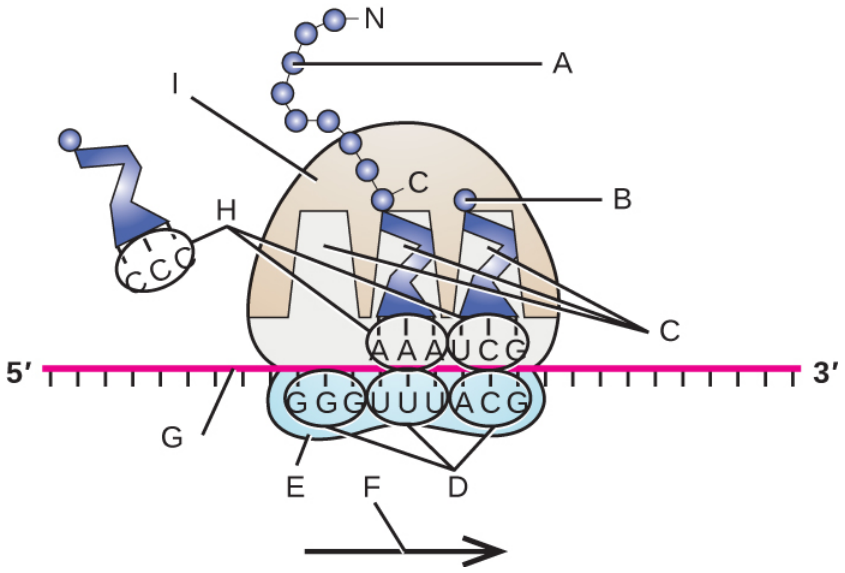
What is meant by the genetic code being nearly universal?

Below is an antisense DNA sequence. Translate the mRNA molecule synthesized using the genetic code, recording the resulting amino acid sequence, indicating the N and C termini.

Antisense DNA strand: 3'-T A C T G A C T G A C G A T C-5'

Critical Thinking

Label the following in the figure: ribosomal E, P, and A sites; mRNA; codons; anticodons; growing polypeptide; incoming amino acid; direction of translocation; small ribosomal unit; large ribosomal unit.



Prior to the elucidation of the genetic code, prominent scientists, including Francis Crick, had predicted that each mRNA codon, coding for one of the 20 amino acids, needed to be at least three nucleotides long. Why is it not possible for codons to be any shorter?

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CHAPTER OVERVIEW

14: Horizontal Gene Transfer and Operons

In 1954, French scientist and future Nobel laureate Jacques Monod (1910–1976) famously said, “What is true in *E. coli* is true in the elephant,” suggesting that the biochemistry of life was maintained throughout evolution and is shared in all forms of known life. Since Monod’s famous statement, we have learned a great deal about the mechanisms of gene regulation, expression, and replication in living cells. All cells use DNA for information storage, share the same genetic code, and use similar mechanisms to replicate and express it. Although many aspects of genetics are universally shared, variations do exist among contemporary genetic systems. We now know that within the shared overall theme of the genetic mechanism, there are significant differences among the three domains of life: Eukarya, Archaea, and Bacteria. Additionally, viruses, cellular parasites but not themselves living cells, show dramatic variation in their genetic material and the replication and gene expression processes. Some of these differences have allowed us to engineer clinical tools such as antibiotics and antiviral drugs that specifically inhibit the reproduction of pathogens yet are harmless to their hosts.

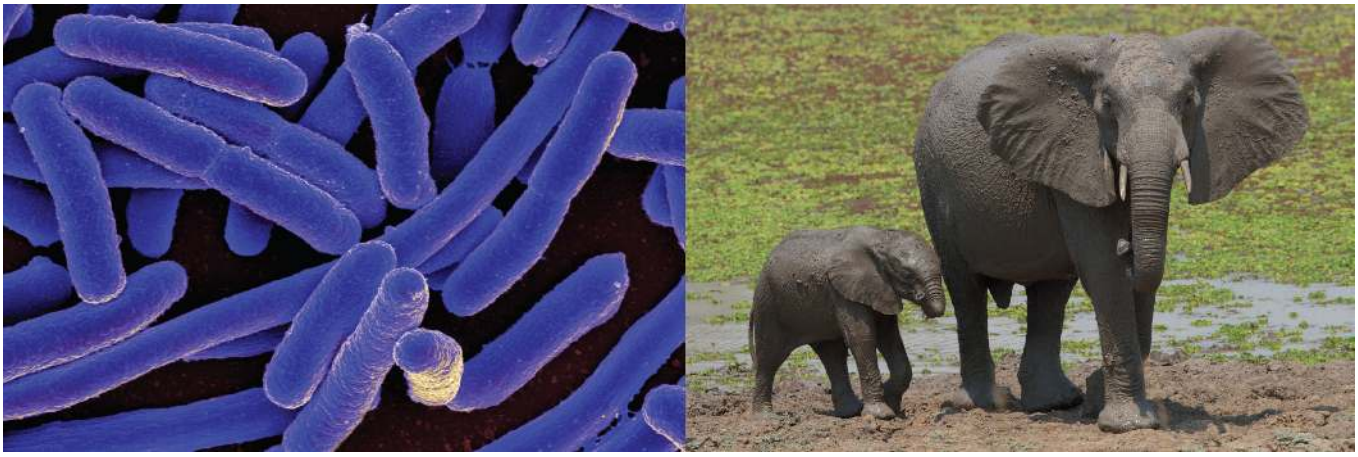


Figure **14.1**: *Escherichia coli* (left) may not appear to have much in common with an elephant (right), but the genetic blueprints for these vastly different organisms are both encoded in DNA. (credit left: modification of work by NIAID; credit right: modification of work by Tom Lubbock)

14.1: Mutations

14.2: How Asexual Prokaryotes Achieve Genetic Diversity

14.3: Gene Regulation - Operon Theory

14.E: Mechanisms of Microbial Genetics (Exercises)

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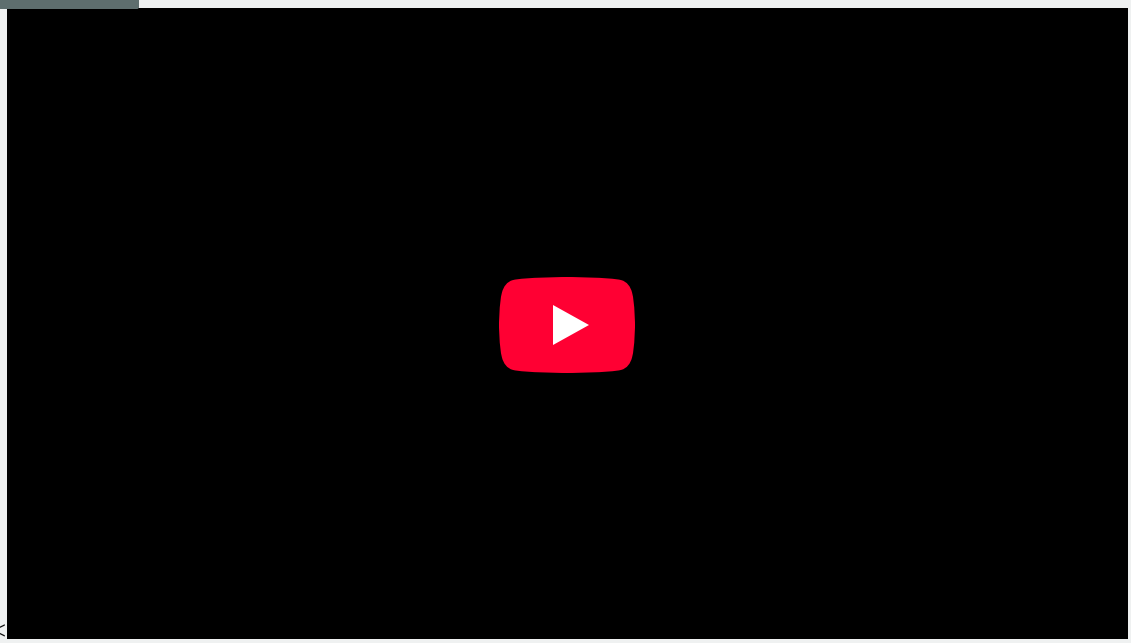
14.1: Mutations

Learning Objectives

- Compare point mutations and frameshift mutations
- Describe the differences between missense, nonsense, and silent mutations
- Explain how different mutagens act
- Analyze sequences of DNA and identify examples of types of mutations

A **mutation** is a heritable change in the DNA sequence of an organism. The resulting organism, called a **mutant**, may have a recognizable change in phenotype compared to the **wild type**, which is the phenotype most commonly observed in nature. A change in the DNA sequence is conferred to mRNA through transcription, and may lead to an altered amino acid sequence in a protein on translation. Because proteins carry out the vast majority of cellular functions, a change in amino acid sequence in a protein may lead to an altered phenotype for the cell and organism.

Mutations



Join the Amoeba Sisters as they explain gene and chromosome mutations, and explore the significance of these changes.

Effects of Mutations on DNA Sequence

There are several types of mutations that are classified according to how the DNA molecule is altered. One type, called a **point mutation**, affects a single base and most commonly occurs when one base is substituted or replaced by another. Mutations also result from the addition of one or more bases, known as an **insertion**, or the removal of one or more bases, known as a **deletion**.

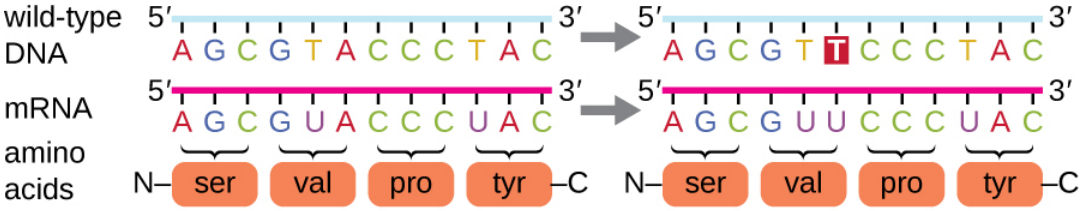
Effects of Mutations on Protein Structure and Function

Point mutations may have a wide range of effects on protein function (Figure **14.1.1**). As a consequence of the degeneracy of the genetic code, a point mutation will commonly result in the same amino acid being incorporated into the resulting polypeptide despite the sequence change. This change would have no effect on the protein's structure, and is thus called a **silent mutation**. A **missense mutation** results in a different amino acid being incorporated into the resulting polypeptide. The effect of a missense mutation depends on how chemically different the new amino acid is from the wild-type amino acid. The location of the changed amino acid within the protein also is important. For example, if the changed amino acid is part of the enzyme's active site, then the effect of the missense mutation may be significant. Many missense mutations result in proteins that are still functional, at least to some degree. Sometimes the effects of missense mutations may be only apparent under certain environmental conditions; such missense mutations are called **conditional mutations**. Rarely, a missense mutation may be beneficial. Under the right environmental conditions, this type of mutation may give the organism that harbors it a selective advantage. Yet another type of point mutation, called a **nonsense mutation**, converts a codon encoding an amino acid (a sense codon) into a stop codon (a nonsense codon). Nonsense mutations result in the synthesis of proteins that are shorter than the wild type and typically not functional.

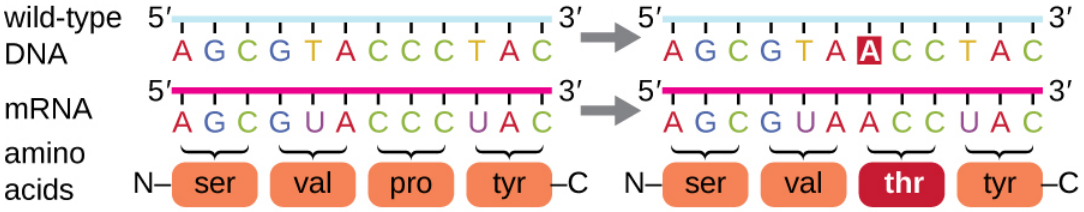
Deletions and insertions also cause various effects. Because codons are triplets of nucleotides, insertions or deletions in groups of three nucleotides may lead to the insertion or deletion of one or more amino acids and may not cause significant effects on the resulting protein's functionality. However, **frameshift mutations**, caused by insertions or deletions of a number of nucleotides that are not a multiple of three are extremely problematic because a shift in the reading frame results (Figure **14.1.1**). Because ribosomes read the mRNA in triplet codons, frameshift mutations can change every amino acid after the point of the mutation. The new reading frame may also include a stop codon before the end of the coding sequence. Consequently, proteins made from genes containing frameshift mutations are nearly always nonfunctional.

point mutation: substitution of a single base

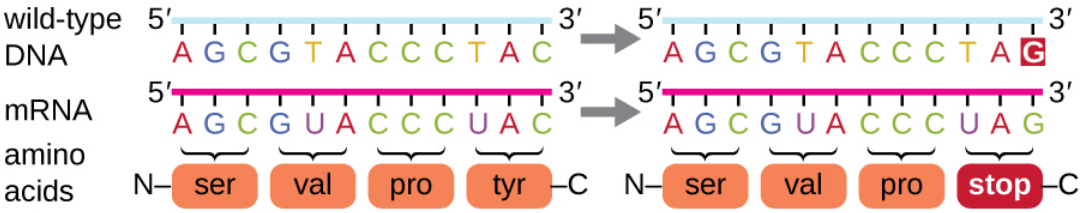
silent: has no effect on the protein sequence



missense: results in an amino acid substitution



nonsense: substitutes a stop codon for an amino acid



frameshift mutation: insertion or deletion of one or more bases

Insertion or **deletion** results in a shift in the reading frame.

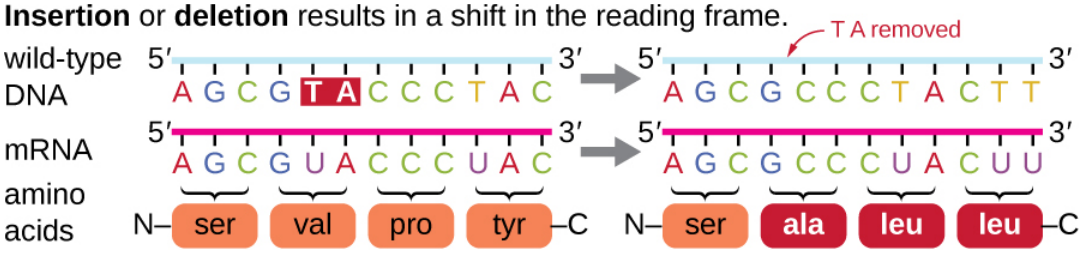


Figure **14.1.1**: Mutations can lead to changes in the protein sequence encoded by the DNA.

A Beneficial Mutation

Since the first case of infection with human immunodeficiency virus (HIV) was reported in 1981, nearly 40 million people have died from HIV infection,[1] the virus that causes acquired immune deficiency syndrome (AIDS). The virus targets helper T cells that play a key role in bridging the innate and adaptive immune response, infecting and killing cells normally involved in the body's response to infection. There is no cure for HIV infection, but many drugs have been developed to slow or block the progression of the virus. Although individuals around the world may be infected, the highest prevalence among people 15–49 years old is in sub-Saharan Africa, where nearly one person in 20 is infected, accounting for greater than 70% of the infections

worldwide[2] (Figure 14.1.2). Unfortunately, this is also a part of the world where prevention strategies and drugs to treat the infection are the most lacking.

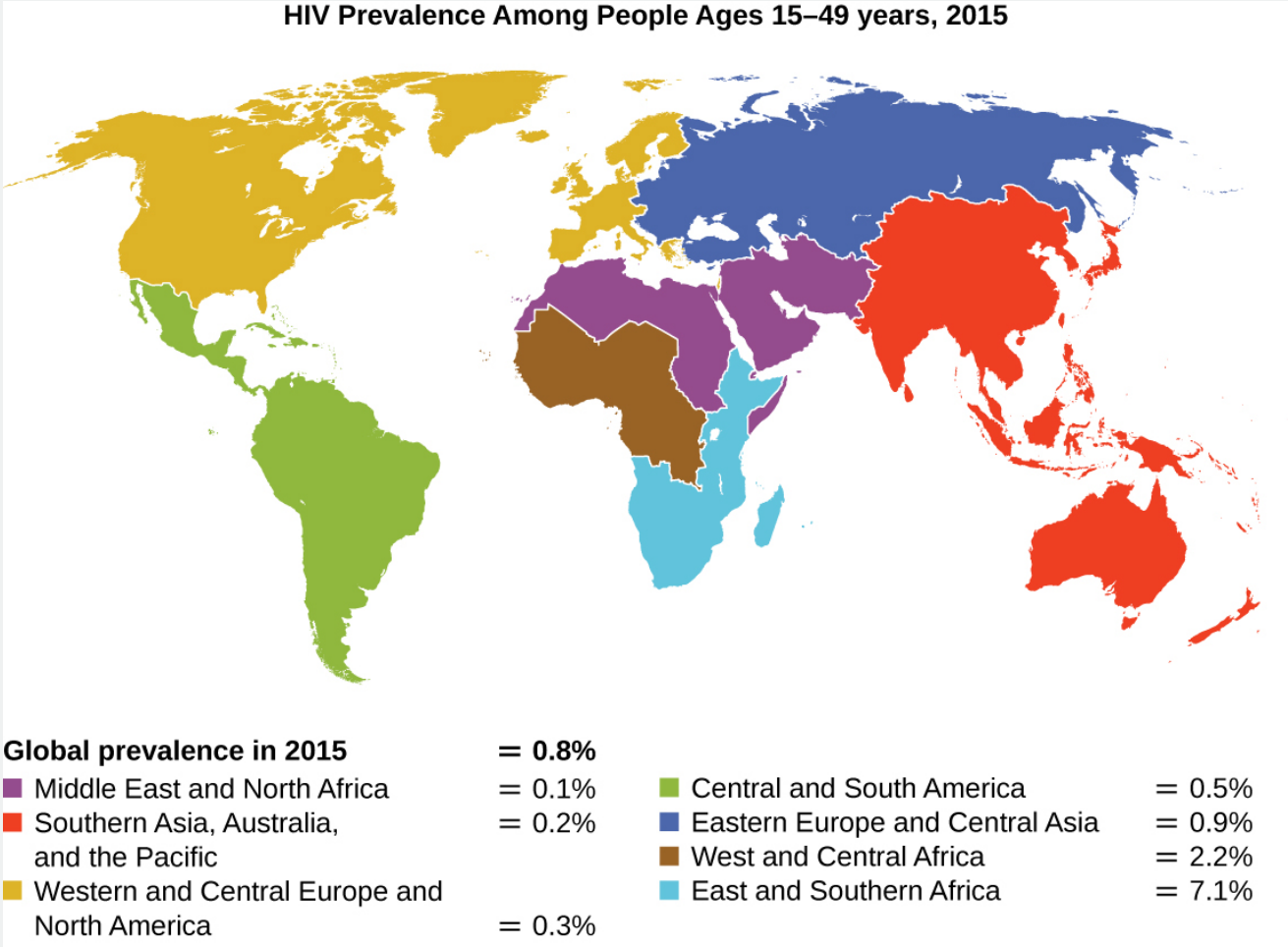


Figure 14.1.2: HIV is highly prevalent in sub-Saharan Africa, but its prevalence is quite low in some other parts of the world.

In recent years, scientific interest has been piqued by the discovery of a few individuals from northern Europe who are resistant to HIV infection. In 1998, American geneticist Stephen J. O'Brien at the National Institutes of Health (NIH) and colleagues published the results of their genetic analysis of more than 4,000 individuals. These indicated that many individuals of Eurasian descent (up to 14% in some ethnic groups) have a deletion mutation, called CCR5-delta 32, in the gene encoding CCR5. CCR5 is a coreceptor found on the surface of T cells that is necessary for many strains of the virus to enter the host cell. The mutation leads to the production of a receptor to which HIV cannot effectively bind and thus blocks viral entry. People homozygous for this mutation have greatly reduced susceptibility to HIV infection, and those who are heterozygous have some protection from infection as well.

It is not clear why people of northern European descent, specifically, carry this mutation, but its prevalence seems to be highest in northern Europe and steadily decreases in populations as one moves south. Research indicates that the mutation has been present since before HIV appeared and may have been selected for in European populations as a result of exposure to the plague or smallpox. This mutation may protect individuals from plague (caused by the bacterium *Yersinia pestis*) and smallpox (caused by the variola virus) because this receptor may also be involved in these diseases. The age of this mutation is a matter of debate, but estimates suggest it appeared between 1875 years to 225 years ago, and may have been spread from Northern Europe through Viking invasions.

This exciting finding has led to new avenues in HIV research, including looking for drugs to block CCR5 binding to HIV in individuals who lack the mutation. Although DNA testing to determine which individuals carry the CCR5-delta 32 mutation is possible, there are documented cases of individuals homozygous for the mutation contracting HIV. For this reason, DNA

testing for the mutation is not widely recommended by public health officials so as not to encourage risky behavior in those who carry the mutation. Nevertheless, inhibiting the binding of HIV to CCR5 continues to be a valid strategy for the development of drug therapies for those infected with HIV.

Causes of Mutations

Mistakes in the process of DNA replication can cause **spontaneous mutations** to occur. The error rate of DNA polymerase is one incorrect base per billion base pairs replicated. Exposure to **mutagens** can cause **induced mutations**, which are various types of chemical agents or radiation (Table **14.1.1**). Exposure to a mutagen can increase the rate of mutation more than 1000-fold. Mutagens are often also carcinogens, agents that cause cancer. However, whereas nearly all carcinogens are mutagenic, not all mutagens are necessarily **carcinogens**.

Chemical Mutagens

Various types of chemical mutagens interact directly with DNA either by acting as nucleoside analogs or by modifying nucleotide bases. Chemicals called **nucleoside analogs** are structurally similar to normal nucleotide bases and can be incorporated into DNA during replication (Figure **14.1.3**). These base analogs induce mutations because they often have different base-pairing rules than the bases they replace. Other chemical mutagens can modify normal DNA bases, resulting in different base-pairing rules. For example, nitrous acid deaminates cytosine, converting it to uracil. Uracil then pairs with adenine in a subsequent round of replication, resulting in the conversion of a GC base pair to an AT base pair. Nitrous acid also deaminates adenine to hypoxanthine, which base pairs with cytosine instead of thymine, resulting in the conversion of a TA base pair to a CG base pair.

Chemical mutagens known as **intercalating agents** work differently. These molecules slide between the stacked nitrogenous bases of the DNA double helix, distorting the molecule and creating atypical spacing between nucleotide base pairs (Figure **14.1.4**). As a result, during DNA replication, DNA polymerase may either skip replicating several nucleotides (creating a deletion) or insert extra nucleotides (creating an insertion). Either outcome may lead to a frameshift mutation. Combustion products like polycyclic aromatic hydrocarbons are particularly dangerous intercalating agents that can lead to mutation-caused cancers. The intercalating agents ethidium bromide and acridine orange are commonly used in the laboratory to stain DNA for visualization and are potential mutagens.

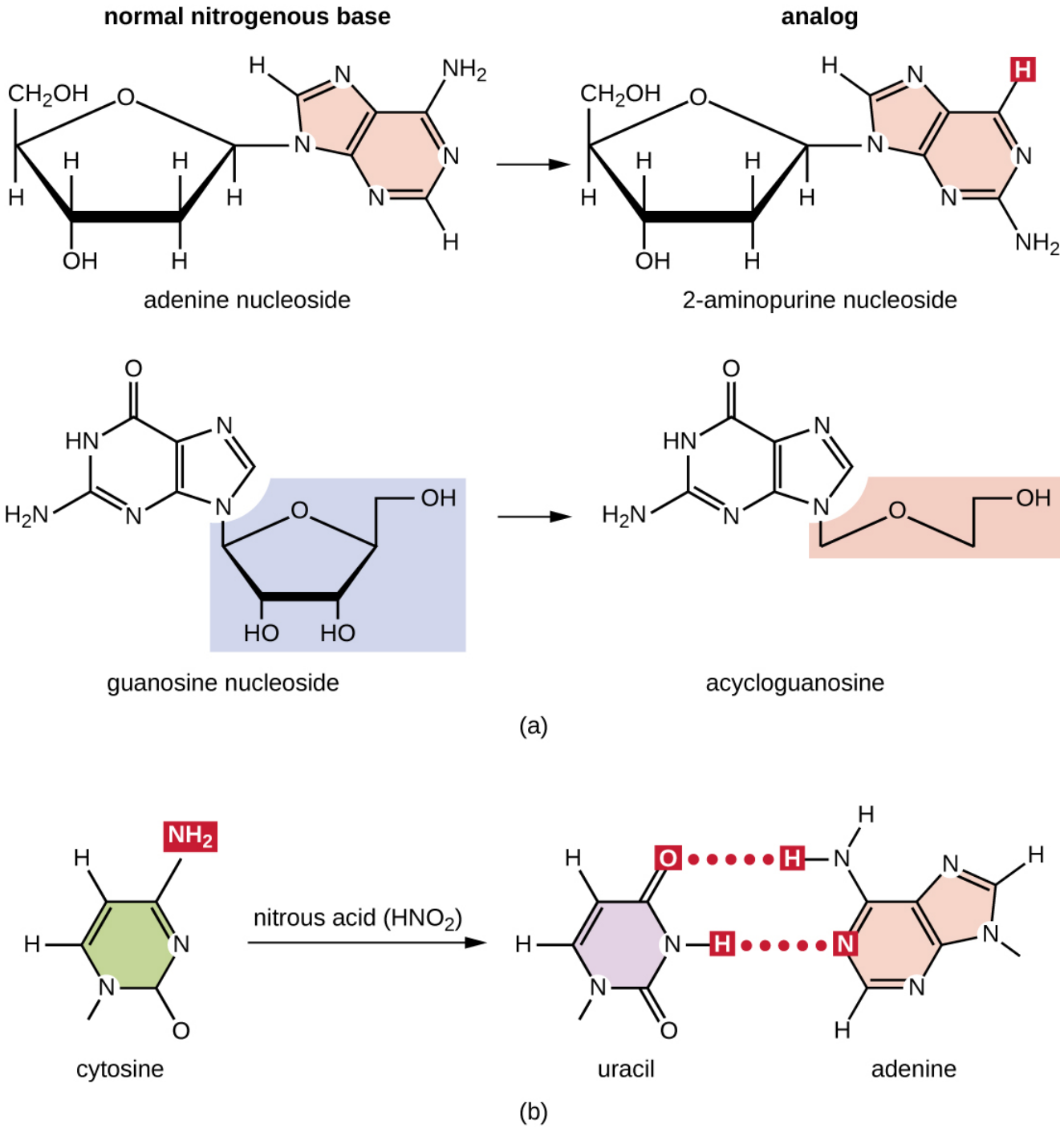


Figure **14.1.3**: (a) 2-aminopurine nucleoside (2AP) structurally is a nucleoside analog to adenine nucleoside, whereas 5-bromouracil (5BU) is a nucleoside analog to thymine nucleoside. 2AP base pairs with C, converting an AT base pair to a GC base pair after several rounds of replication. 5BU pairs with G, converting an AT base pair to a GC base pair after several rounds of replication. (b) Nitrous acid is a different type of chemical mutagen that modifies already existing nucleoside bases like C to produce U, which base pairs with A. This chemical modification, as shown here, results in converting a CG base pair to a TA base pair.

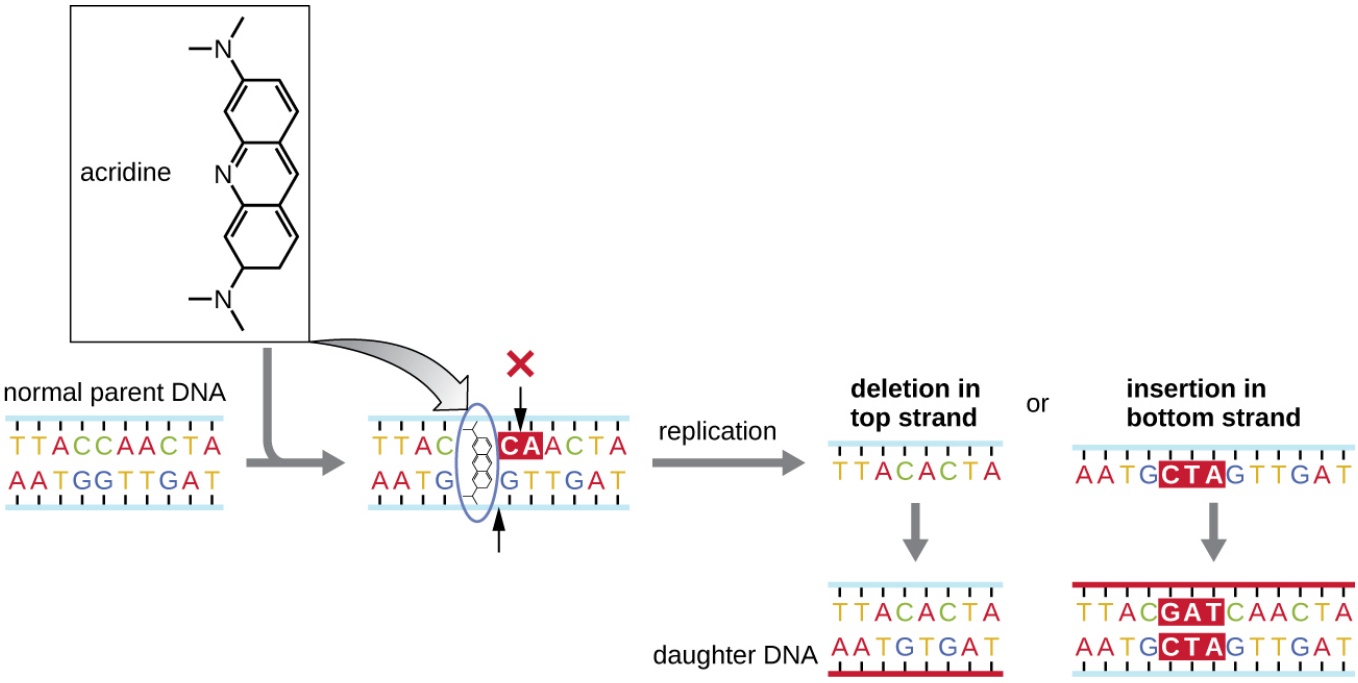
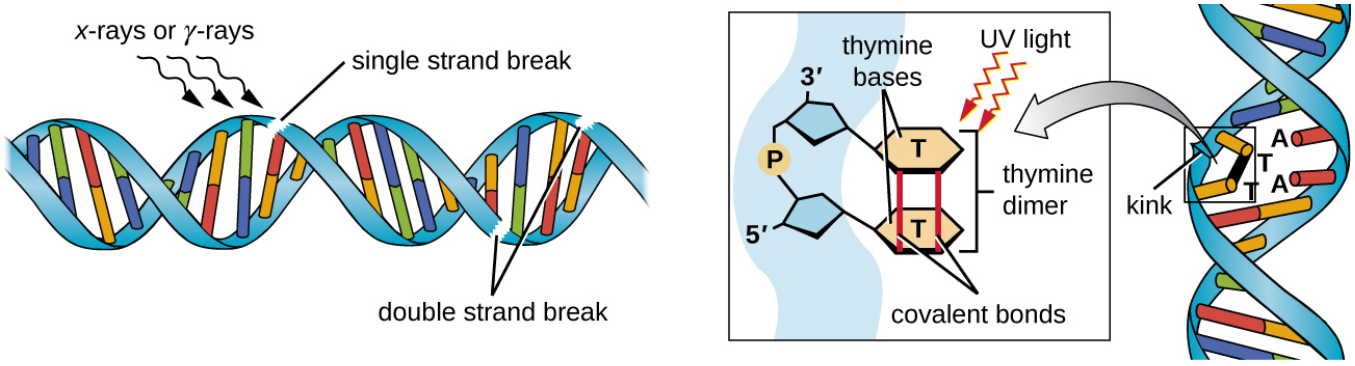


Figure **14.1.4**: Intercalating agents, such as acridine, introduce atypical spacing between base pairs, resulting in DNA polymerase introducing either a deletion or an insertion, leading to a potential frameshift mutation.

Radiation

Exposure to either ionizing or nonionizing radiation can each induce mutations in DNA, although by different mechanisms. Strong **ionizing radiation** like X-rays and gamma rays can cause single- and double-stranded breaks in the DNA backbone through the formation of hydroxyl radicals on radiation exposure (Figure **14.1.5**). Ionizing radiation can also modify bases; for example, the deamination of cytosine to uracil, analogous to the action of nitrous acid.[3] Ionizing radiation exposure is used to kill microbes to sterilize medical devices and foods, because of its dramatic nonspecific effect in damaging DNA, proteins, and other cellular components (see Using Physical Methods to Control Microorganisms).

Nonionizing radiation, like ultraviolet light, is not energetic enough to initiate these types of chemical changes. However, **nonionizing radiation** can induce dimer formation between two adjacent pyrimidine bases, commonly two thymines, within a nucleotide strand. During **thymine dimer** formation, the two adjacent thymines become covalently linked and, if left unrepaired, both DNA replication and transcription are stalled at this point. DNA polymerase may proceed and replicate the dimer incorrectly, potentially leading to frameshift or point mutations.



(a) Ionizing radiation (b) Non-ionizing radiation

Figure **14.1.5**: (a) Ionizing radiation may lead to the formation of single-stranded and double-stranded breaks in the sugar-phosphate backbone of DNA, as well as to the modification of bases (not shown). (b) Nonionizing radiation like ultraviolet light can lead to the formation of thymine dimers, which can stall replication and transcription and introduce frameshift or point mutations.

Table **14.1.1**: A Summary of Mutagenic Agents

Mutagenic Agents	Mode of Action	Effect on DNA	Resulting Type of Mutation
Nucleoside analogs			
2-aminopurine	Is inserted in place of A but base pairs with C	Converts AT to GC base pair	Point
5-bromouracil	Is inserted in place of T but base pairs with G	Converts AT to GC base pair	Point
Nucleotide-modifying agent			
Nitrous oxide	Deaminates C to U	Converts GC to AT base pair	Point
Intercalating agents			
Acridine orange, ethidium bromide, polycyclic aromatic hydrocarbons	Distorts double helix, creates unusual spacing between nucleotides	Introduces small deletions and insertions	Frameshift
Ionizing radiation			
X-rays, γ-rays	Forms hydroxyl radicals	Causes single- and double-strand DNA breaks	Repair mechanisms may introduce mutations
X-rays, γ-rays	Modifies bases (e.g., deaminating C to U)	Converts GC to AT base pair	Point
Nonionizing radiation			
Ultraviolet	Forms pyrimidine (usually thymine) dimers	Causes DNA replication errors	Frameshift or point

Key Concepts and Summary

- A **mutation** is a heritable change in DNA. A mutation may lead to a change in the amino-acid sequence of a protein, possibly affecting its function.
- A **point mutation** affects a single base pair. A point mutation may cause a **silent mutation** if the mRNA codon codes for the same amino acid, a **missense mutation** if the mRNA codon codes for a different amino acid, or a **nonsense mutation** if the mRNA codon becomes a stop codon.
- Missense mutations may retain function, depending on the chemistry of the new amino acid and its location in the protein. Nonsense mutations produce truncated and frequently nonfunctional proteins.
- A **frameshift mutation** results from an insertion or deletion of a number of nucleotides that is not a multiple of three. The change in reading frame alters every amino acid after the point of the mutation and results in a nonfunctional protein.
- **Spontaneous mutations** occur through DNA replication errors, whereas **induced mutations** occur through exposure to a **mutagen**.
- Mutagenic agents are frequently carcinogenic but not always. However, nearly all carcinogens are mutagenic.
- Chemical mutagens include base analogs and chemicals that modify existing bases. In both cases, mutations are introduced after several rounds of DNA replication.
- **Ionizing radiation,** such as X-rays and γ-rays, leads to breakage of the phosphodiester backbone of DNA and can also chemically modify bases to alter their base-pairing rules.
- **Nonionizing radiation** like ultraviolet light may introduce pyrimidine (thymine) dimers, which, during DNA replication and transcription, may introduce frameshift or point mutations.

Footnotes

1. 1 World Health Organization. “ Global Health Observatory (GHO) Data, HIV/AIDS.” http://www.who.int/gho/hiv/en/. Accessed August 5, 2016.
2. 2 World Health Organization. “ Global Health Observatory (GHO) Data, HIV/AIDS.” http://www.who.int/gho/hiv/en/. Accessed August 5, 2016.
3. 3 K.R. Tindall et al. “Changes in DNA Base Sequence Induced by Gamma-Ray Mutagenesis of Lambda Phage and Prophage.” *Genetics* 118 no. 4 (1988):551–560.

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14.2: How Asexual Prokaryotes Achieve Genetic Diversity

Learning Objectives

- Compare the processes of transformation and transduction
- Explain how asexual gene transfer results in prokaryotic genetic diversity
- Evaluate how scientists use horizontal gene transfer mechanisms in genetic engineering and recombinant DNA technology

Typically, when we consider genetic transfer, we think of **vertical gene transfer**, the transmission of genetic information from generation to generation. Vertical gene transfer is by far the main mode of transmission of genetic information in all cells. In sexually reproducing organisms, crossing-over events and independent assortment of individual chromosomes during meiosis contribute to genetic diversity in the population. Genetic diversity is also introduced during sexual reproduction, when the genetic information from two parents, each with different complements of genetic information, are combined, producing new combinations of parental genotypes in the diploid offspring. The occurrence of mutations also contributes to genetic diversity in a population. Genetic diversity of offspring is useful in changing or inconsistent environments and may be one reason for the evolutionary success of sexual reproduction.

When prokaryotes and eukaryotes reproduce asexually, they transfer a nearly identical copy of their genetic material to their offspring through vertical gene transfer. Although asexual reproduction produces more offspring more quickly, any benefits of diversity among those offspring are lost. How then do organisms whose dominant reproductive mode is asexual create genetic diversity? In prokaryotes, **horizontal gene transfer (HGT)**, the introduction of genetic material from one organism to another organism within the same generation, is an important way to introduce genetic diversity. HGT allows even distantly related species to share genes, influencing their phenotypes. It is thought that HGT is more prevalent in prokaryotes but that only a small fraction of the prokaryotic genome may be transferred by this type of transfer at any one time. As the phenomenon is investigated more thoroughly, it may be revealed to be even more common. Many scientists believe that HGT and mutation are significant sources of genetic variation, the raw material for the process of natural selection, in prokaryotes. Although HGT is more common among evolutionarily related organisms, it may occur between any two species that live together in a natural community.

HGT in prokaryotes is known to occur by the three primary mechanisms that are illustrated in Figure **14.2.1**:

1. Transformation: naked DNA is taken up from the environment
2. Transduction: genes are transferred between cells in a virus (see The Viral Life Cycle)
3. Conjugation: use of a hollow tube called a conjugation pilus to transfer genes between cells

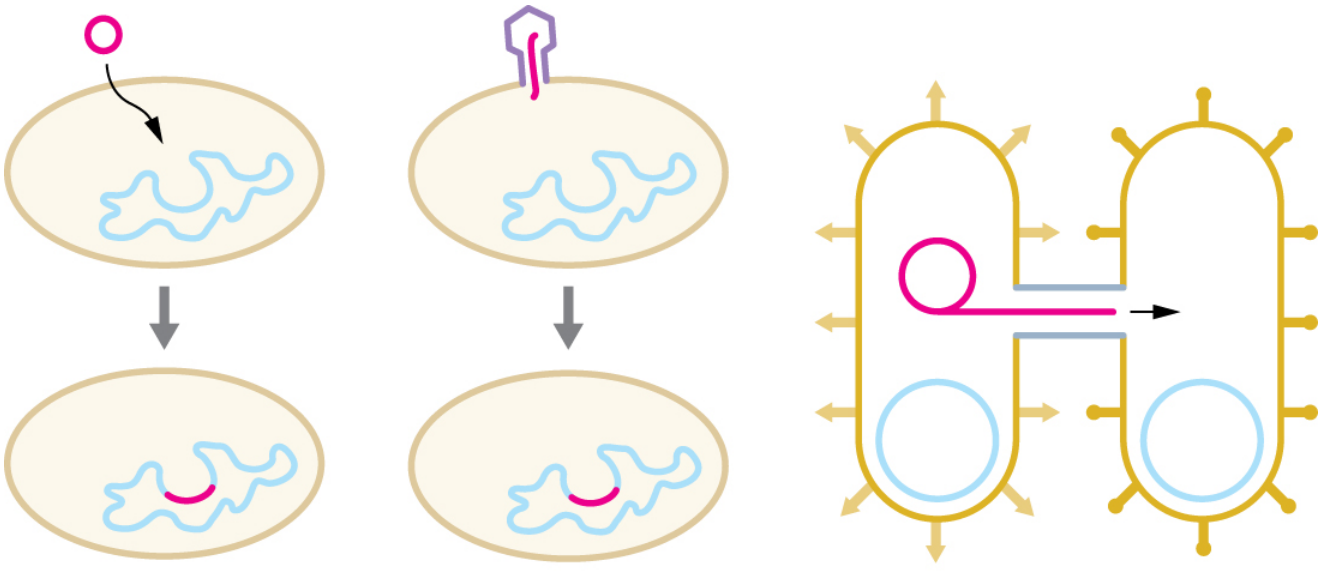
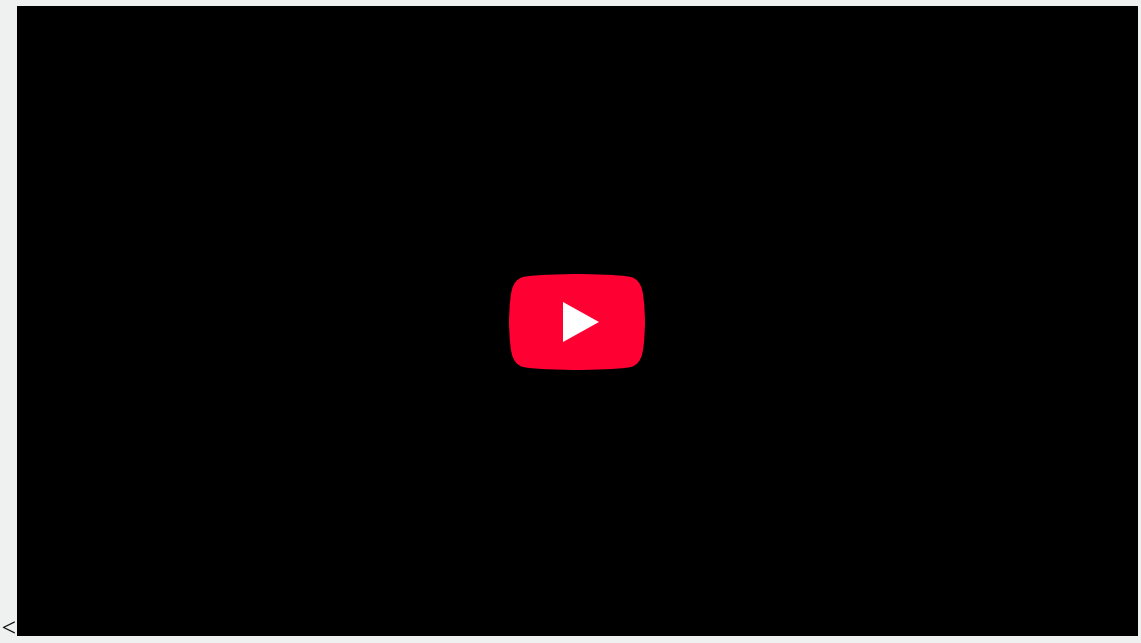
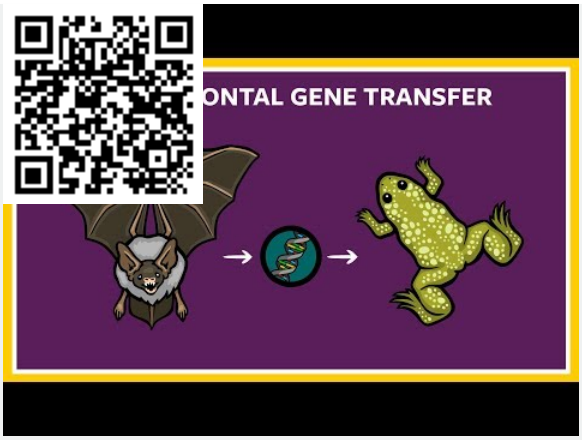


Figure **14.2.1**: There are three prokaryote-specific mechanisms leading to horizontal gene transfer in prokaryotes. a) In transformation, the cell takes up DNA directly from the environment. The DNA may remain separate as a plasmid or be incorporated into the host genome. b) In transduction, a bacteriophage injects DNA that is a hybrid of viral DNA and DNA from a previously infected bacterial cell. c) In conjugation, DNA is transferred between cells through a cytoplasmic bridge after a conjugation pilus draws the two cells close enough to form the bridge.

What is Horizontal Gene Transfer?





>

A Horizontal Gene Transfer (sometimes called lateral gene transfer or sideways gene transfer) is the passing of one or more genes through routes other than parent-to-offspring. In this animation you will learn what that looks like and how it effects evolution.

Transformation

Frederick Griffith was the first to demonstrate the process of transformation. In 1928, he showed that live, nonpathogenic *Streptococcus pneumoniae* bacteria could be transformed into pathogenic bacteria through exposure to a heat-killed pathogenic strain. He concluded that some sort of agent, which he called the "transforming principle," had been passed from the dead pathogenic bacteria to the live, nonpathogenic bacteria. In 1944, Oswald Avery (1877–1955), Colin MacLeod (1909–1972), and Maclyn McCarty (1911–2005) demonstrated that the transforming principle was DNA (see Using Microbiology to Discover the Secrets of Life).

In **transformation**, the prokaryote takes up naked DNA found in its environment and that is derived from other cells that have lysed on death and released their contents, including their genome, into the environment. Many bacteria are naturally competent, meaning that they actively bind to environmental DNA, transport it across their cell envelopes into their cytoplasm, and make it single stranded. Typically, double-stranded foreign DNA within cells is destroyed by nucleases as a defense against viral infection. However, these nucleases are usually ineffective against single-stranded DNA, so this single-stranded DNA within the cell has the opportunity to recombine into the bacterial genome. A molecule of DNA that contains fragments of DNA from different organisms is called recombinant DNA. (Recombinant DNA will be discussed in more detail in Microbes and the Tools of Genetic Engineering.) If the bacterium incorporates the new DNA into its own genome through recombination, the bacterial cell may gain new phenotypic properties. For example, if a nonpathogenic bacterium takes up DNA for a toxin gene from a pathogen and then incorporates it into its chromosome, it, too, may become pathogenic. Plasmid DNA may also be taken up by competent bacteria and confer new properties to the cell. Overall, transformation in nature is a relatively inefficient process because environmental DNA levels are low because of the activity of nucleases that are also released during cellular lysis. Additionally, genetic recombination is inefficient at incorporating new DNA sequences into the genome.

In nature, bacterial transformation is an important mechanism for the acquisition of genetic elements encoding virulence factors and antibiotic resistance. Genes encoding resistance to antimicrobial compounds have been shown to be widespread in nature, even in environments not influenced by humans. These genes, which allow microbes living in mixed communities to compete for limited resources, can be transferred within a population by transformation, as well as by the other processes of HGT. In the laboratory, we can exploit the natural process of bacterial transformation for genetic engineering to make a wide variety of medicinal products, as discussed in Microbes and the Tools of Genetic Engineering.

Transduction

Viruses that infect bacteria (bacteriophages) may also move short pieces of chromosomal DNA from one bacterium to another in a process called **transduction** (see Figure 6.2.3). Recall that in **generalized transduction**, any piece of chromosomal DNA may be transferred to a new host cell by accidental packaging of chromosomal DNA into a phage head during phage assembly. By contrast, **specialized transduction** results from the imprecise excision of a lysogenic prophage from the bacterial chromosome such that it carries with it a piece of the bacterial chromosome from either side of the phage's integration site to a new host cell. As a result, the host may acquire new properties. This process is called **lysogenic conversion**. Of medical significance, a lysogenic phage may carry with it a virulence gene to its new host. Once inserted into the new host's chromosome, the new host may gain pathogenicity. Several pathogenic bacteria, including *Corynebacterium diphtheriae* (the causative agent of diphtheria) and *Clostridium botulinum* (the causative agent of botulism), are virulent because of the introduction of toxin-encoding genes by lysogenic bacteriophages, affirming the clinical relevance of transduction in the exchange of genes involved in infectious disease. Archaea have their own viruses that translocate genetic material from one individual to another.

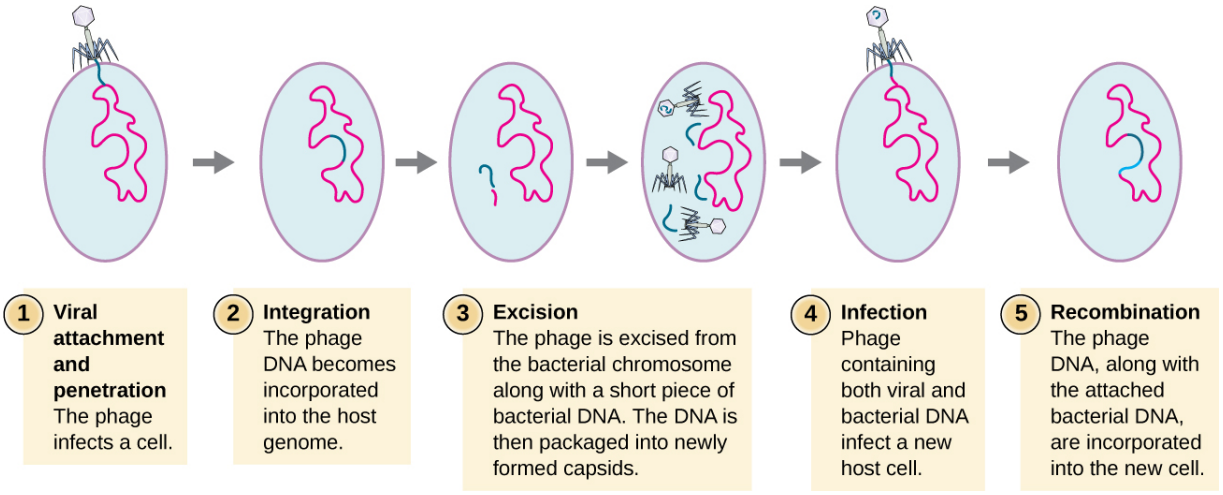


Figure **14.2.1**: This flowchart illustrates the mechanism of specialized transduction. An integrated phage excises, bringing with it a piece of the DNA adjacent to its insertion point. On reinfection of a new bacterium, the phage DNA integrates along with the genetic material acquired from the previous host.

The Clinical Consequences of Transduction

Paul, a 23-year-old relief worker from Atlanta, traveled to Haiti in 2011 to provide aid following the 2010 earthquake. After working there for several weeks, he suddenly began experiencing abdominal distress, including severe cramping, nausea, vomiting, and watery diarrhea. He also began to experience intense muscle cramping. At a local clinic, the physician suspected that Paul's symptoms were caused by cholera because there had been a cholera outbreak after the earthquake. Because cholera is transmitted by the fecal-oral route, breaches in sanitation infrastructure, such as often occur following natural disasters, may precipitate outbreaks. The physician confirmed the presumptive diagnosis using a cholera dipstick test. He then prescribed Paul a single dose of doxycycline, as well as oral rehydration salts, instructing him to drink significant amounts of clean water.

Cholera is caused by the gram-negative curved rod *Vibrio cholerae* (Figure **14.2.2**). Its symptoms largely result from the production of the cholera toxin (CT), which ultimately activates a chloride transporter to pump chloride ions out of the epithelial cells into the gut lumen. Water then follows the chloride ions, causing the prolific watery diarrhea characteristic of cholera. The gene encoding the cholera toxin is incorporated into the bacterial chromosome of *V. cholerae* through infection of the bacterium with the lysogenic filamentous CTX phage, which carries the CT gene and introduces it into the chromosome on integration of the prophage. Thus, pathogenic strains of *V. cholerae* result from horizontal gene transfer by specialized transduction.

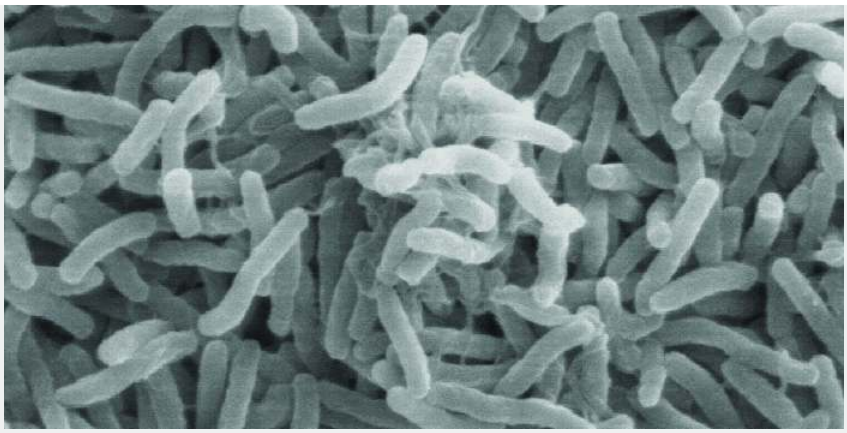


Figure 14.2.2: A scanning electron micrograph of *Vibrio cholerae* shows its characteristic curved rod shape.

Key Concepts and Summary

- **Horizontal gene transfer** is an important way for asexually reproducing organisms like prokaryotes to acquire new traits.
- There are three mechanisms of horizontal gene transfer typically used by bacteria: **transformation**, **transduction**, and **conjugation**.
- Transformation allows for competent cells to take up naked DNA, released from other cells on their death, into their cytoplasm, where it may recombine with the host genome.
- In **generalized transduction**, any piece of chromosomal DNA may be transferred by accidental packaging of the degraded host chromosome into a phage head. In **specialized transduction**, only chromosomal DNA adjacent to the integration site of a lysogenic phage may be transferred as a result of imprecise excision of the prophage.

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14.3: Gene Regulation - Operon Theory

Learning Objectives

- Distinguish structural genes and regulatory genes and their different roles in cells
- Define operon and identify its components
- Explain how repressible operon works using Trp operon as an example

Each nucleated cell in a multicellular organism contains copies of the same DNA. Similarly, all cells in two pure bacterial cultures inoculated from the same starting colony contain the same DNA, with the exception of changes that arise from spontaneous mutations. If each cell in a multicellular organism has the same DNA, then how is it that cells in different parts of the organism's body exhibit different characteristics? Similarly, how is it that the same bacterial cells within two pure cultures exposed to different environmental conditions can exhibit different phenotypes? In both cases, each genetically identical cell does not turn on, or express, the same set of genes. Only a subset of proteins in a cell at a given time is expressed.

Genomic DNA contains both **structural genes**, which encode products that serve as cellular structures or enzymes, and **regulatory genes**, which encode products that regulate gene expression. The expression of a gene is a highly regulated process. Whereas regulating gene expression in multicellular organisms allows for cellular differentiation, in single-celled organisms like prokaryotes, it primarily ensures that a cell's resources are not wasted making proteins that the cell does not need at that time.

Elucidating the mechanisms controlling gene expression is important to the understanding of human health. Malfunctions in this process in humans lead to the development of cancer and other diseases. Understanding the interaction between the gene expression of a pathogen and that of its human host is important for the understanding of a particular infectious disease. Gene regulation involves a complex web of interactions within a given cell among signals from the cell's environment, signaling molecules within the cell, and the cell's DNA. These interactions lead to the expression of some genes and the suppression of others, depending on circumstances.

Prokaryotes and eukaryotes share some similarities in their mechanisms to regulate gene expression; however, gene expression in eukaryotes is more complicated because of the temporal and spatial separation between the processes of transcription and translation. Thus, although most regulation of gene expression occurs through transcriptional control in prokaryotes, regulation of gene expression in eukaryotes occurs at the transcriptional level and post-transcriptionally (after the primary transcript has been made).

Prokaryotic Gene Regulation

In bacteria and archaea, structural proteins with related functions are usually encoded together within the genome in a block called an **operon** and are transcribed together under the control of a single promoter, resulting in the formation of a polycistronic transcript (Figure **14.3.1**). In this way, regulation of the transcription of all of the structural genes encoding the enzymes that catalyze the many steps in a single biochemical pathway can be controlled simultaneously, because they will either all be needed at the same time, or none will be needed. For example, in *E. coli*, all of the structural genes that encode enzymes needed to use lactose as an energy source lie next to each other in the lactose (or *lac*) operon under the control of a single promoter, the *lac* promoter. French scientists François Jacob (1920–2013) and Jacques Monod at the Pasteur Institute were the first to show the organization of bacterial genes into operons, through their studies on the *lac* operon of *E. coli*. For this work, they won the Nobel Prize in Physiology or Medicine in 1965. Although eukaryotic genes are not organized into operons, prokaryotic operons are excellent models for learning about gene regulation generally. There are some gene clusters in eukaryotes that function similar to operons. Many of the principles can be applied to eukaryotic systems and contribute to our understanding of changes in gene expression in eukaryotes that can result pathological changes such as cancer.

Each operon includes DNA sequences that influence its own transcription; these are located in a region called the **regulatory region**. The regulatory region includes the **promoter** and the region surrounding the promoter, to which **transcription factors**, proteins encoded by regulatory genes, can bind. Transcription factors influence the binding of **RNA polymerase** to the promoter and allow its progression to transcribe structural genes. A **repressor** is a transcription factor that suppresses transcription of a gene in response to an external stimulus by binding to a DNA sequence within the regulatory region called the **operator**, which is

located between the RNA polymerase binding site of the promoter and the transcriptional start site of the first structural gene. Repressor binding physically blocks RNA polymerase from transcribing structural genes. Conversely, an **activator** is a transcription factor that increases the transcription of a gene in response to an external stimulus by facilitating RNA polymerase binding to the promoter. An **inducer**, a third type of regulatory molecule, is a small molecule that either activates or represses transcription by interacting with a repressor or an activator.

In prokaryotes, there are examples of operons whose gene products are required rather consistently and whose expression, therefore, is unregulated. Such operons are **constitutively expressed**, meaning they are transcribed and translated continuously to provide the cell with constant intermediate levels of the protein products. Such genes encode enzymes involved in housekeeping functions required for cellular maintenance, including DNA replication, repair, and expression, as well as enzymes involved in core metabolism. In contrast, there are other prokaryotic operons that are expressed only when needed and are regulated by repressors, activators, and inducers.

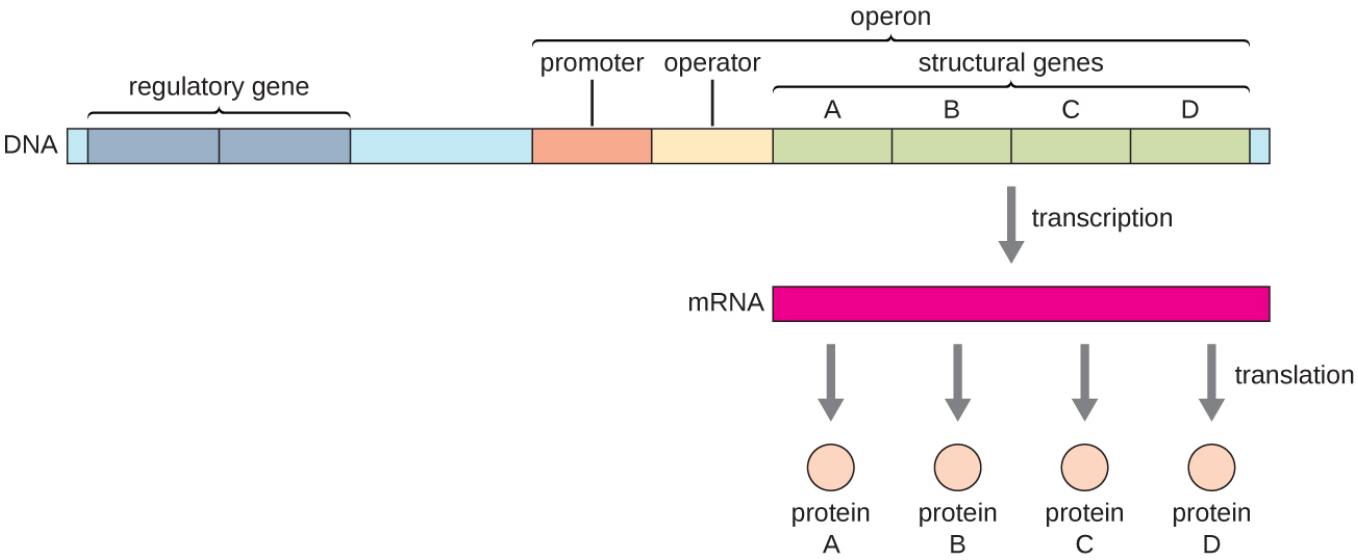


Figure **14.3.1**: In prokaryotes, structural genes of related function are often organized together on the genome and transcribed together under the control of a single promoter. The operon's regulatory region includes both the promoter and the operator. If a repressor binds to the operator, then the structural genes will not be transcribed. Alternatively, activators may bind to the regulatory region, enhancing transcription.

Gene Regulation and the Operon



Explore gene expression with the Amoeba Sisters, including the fascinating Lac Operon found in bacteria! Learn how genes can be turned "on" and "off" and why this is essential for cellular function.

Regulation by Repression

Prokaryotic operons are commonly controlled by the binding of repressors to operator regions, thereby preventing the transcription of the structural genes. Such operons are classified as either **repressible operons** or inducible operons. Repressible operons, like the tryptophan (*trp*) operon, typically contain genes encoding enzymes required for a biosynthetic pathway. As long as the product of the pathway, like tryptophan, continues to be required by the cell, a repressible operon will continue to be expressed. However, when the product of the biosynthetic pathway begins to accumulate in the cell, removing the need for the cell to continue to make more, the expression of the operon is repressed. Conversely, **inducible operons**, like the *lac* operon of *E. coli*, often contain genes encoding enzymes in a pathway involved in the metabolism of a specific substrate like lactose. These enzymes are only required when that substrate is available, thus expression of the operons is typically induced only in the presence of the substrate.

The trp Operon: A Repressible Operon

E. coli can synthesize tryptophan using enzymes that are encoded by five structural genes located next to each other in the *trp* operon (Figure **14.3.2**). When environmental tryptophan is low, the operon is turned on. This means that transcription is initiated, the genes are expressed, and tryptophan is synthesized. However, if tryptophan is present in the environment, the *trp* operon is turned off. Transcription does not occur and tryptophan is not synthesized.

When tryptophan is not present in the cell, the repressor by itself does not bind to the operator; therefore, the operon is active and tryptophan is synthesized. However, when tryptophan accumulates in the cell, two tryptophan molecules bind to the *trp* repressor molecule, which changes its shape, allowing it to bind to the *trp* operator. This binding of the active form of the *trp* repressor to the operator blocks RNA polymerase from transcribing the structural genes, stopping expression of the operon. Thus, the actual product of the biosynthetic pathway controlled by the operon regulates the expression of the operon.

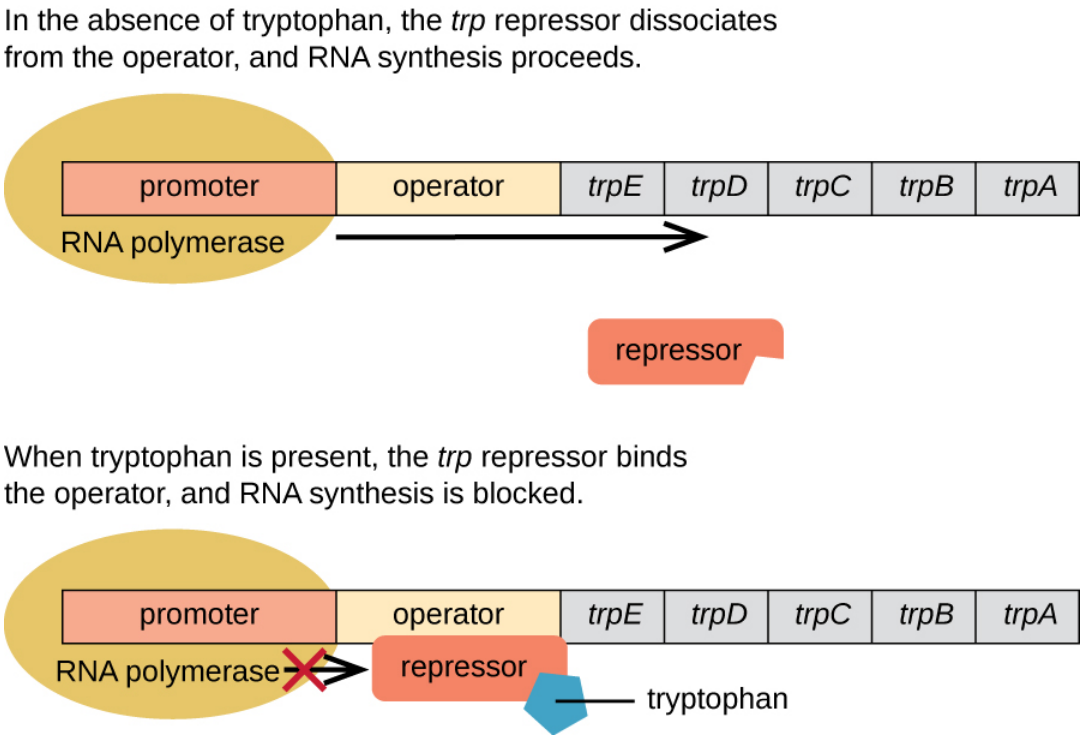


Figure **14.3.2**: The five structural genes needed to synthesize tryptophan in *E. coli* are located next to each other in the *trp* operon. When tryptophan is absent, the repressor protein does not bind to the operator, and the genes are transcribed. When tryptophan is plentiful, tryptophan binds the repressor protein at the operator sequence. This physically blocks the RNA polymerase from transcribing the tryptophan biosynthesis genes.

Link to Learning

Watch this video to learn more about the *trp* operon.

Key Concepts and Summary

- **Gene expression** is a tightly regulated process.
- Gene expression in prokaryotes is largely regulated at the point of transcription. Gene expression in eukaryotes is additionally regulated post-transcriptionally.
- Prokaryotic structural genes of related function are often organized into **operons**, all controlled by transcription from a single promoter. The regulatory region of an operon includes the promoter itself and the region surrounding the promoter to which transcription factors can bind to influence transcription.
- Although some operons are **constitutively expressed**, most are subject to regulation through the use of **transcription factors** (repressors and activators). A **repressor** binds to an **operator**, a DNA sequence within the regulatory region between the RNA polymerase binding site in the promoter and first structural gene, thereby physically blocking transcription of these operons. An **activator** binds within the regulatory region of an operon, helping RNA polymerase bind to the promoter, thereby enhancing the transcription of this operon. An **inducer**influences transcription through interacting with a repressor or activator.
- The *trp* operon is a classic example of a **repressible operon**. When tryptophan accumulates, tryptophan binds to a repressor, which then binds to the operator, preventing further transcription.

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14.E: Mechanisms of Microbial Genetics (Exercises)

13.1: Mutations

A mutation is a heritable change in the DNA sequence of an organism. The resulting organism, called a mutant, may have a recognizable change in phenotype compared to the wild type, which is the phenotype most commonly observed in nature. A change in the DNA sequence is conferred to mRNA through transcription, and may lead to an altered amino acid sequence in a protein on translation.

Multiple Choice

Which of the following is a change in the sequence that leads to formation of a stop codon?

A. missense mutation
B. nonsense mutation
C. silent mutation
D. deletion mutation

Answer

B

The formation of pyrimidine dimers results from which of the following?

A. spontaneous errors by DNA polymerase
B. exposure to gamma radiation
C. exposure to ultraviolet radiation
D. exposure to intercalating agents

Answer

C

Which of the following is an example of a frameshift mutation?

A. a deletion of a codon
B. missense mutation
C. silent mutation
D. deletion of one nucleotide

Answer

D

Which of the following is the type of DNA repair in which thymine dimers are directly broken down by the enzyme photolyase?

A. direct repair
B. nucleotide excision repair
C. mismatch repair
D. proofreading

Answer

A

Which of the following regarding the Ames test is true?

A. It is used to identify newly formed auxotrophic mutants.
B. It is used to identify mutants with restored biosynthetic activity.
C. It is used to identify spontaneous mutants.

D. It is used to identify mutants lacking photoreactivation activity.

Answer

B

Fill in the Blank

A chemical mutagen that is structurally similar to a nucleotide but has different base-pairing rules is called a _____.

Answer

nucleoside analog

The enzyme used in light repair to split thymine dimers is called _____.

Answer

photolyase

The phenotype of an organism that is most commonly observed in nature is called the _____.

Answer

wild type

True/False

Carcinogens are typically mutagenic.

Answer

True

Short Answer

Why is it more likely that insertions or deletions will be more detrimental to a cell than point mutations?

Critical Thinking

Below are several DNA sequences that are mutated compared with the wild-type sequence: 3’-T A C T G A C T G A C G A T C-5’. Envision that each is a section of a DNA molecule that has separated in preparation for transcription, so you are only seeing the template strand. Construct the complementary DNA sequences (indicating 5’ and 3’ ends) for each mutated DNA sequence, then

transcribe (indicating 5’ and 3’ ends) the template strands, and translate the mRNA molecules using the genetic code, recording the resulting amino acid sequence (indicating the N and C termini). What type of mutation is each?

Mutated DNA Template Strand #1: 3’-T A C T G T C T G A C G A T C-5’
Complementary DNA sequence:
mRNA sequence transcribed from template:
Amino acid sequence of peptide:
Type of mutation:

Mutated DNA Template Strand #2: 3’-T A C G G A C T G A C G A T C-5’
Complementary DNA sequence:
mRNA sequence transcribed from template:
Amino acid sequence of peptide:
Type of mutation:

Mutated DNA Template Strand #3: 3’-T A C T G A C T G A C T A T C-5’
Complementary DNA sequence:
mRNA sequence transcribed from template:
Amino acid sequence of peptide:
Type of mutation:

Mutated DNA Template Strand #4: 3’-T A C G A C T G A C T A T C-5’
Complementary DNA sequence:
mRNA sequence transcribed from template:
Amino acid sequence of peptide:
Type of mutation:

Why do you think the Ames test is preferable to the use of animal models to screen chemical compounds for mutagenicity?

13.2: How Asexual Prokaryotes Achieve Genetic Diversity

How do organisms whose dominant reproductive mode is asexual create genetic diversity? In prokaryotes, horizontal gene transfer (HGT), the introduction of genetic material from one organism to another organism within the same generation, is an important way to introduce genetic diversity. HGT allows even distantly related species to share genes, influencing their phenotypes.

Multiple Choice

Which is the mechanism by which improper excision of a prophage from a bacterial chromosome results in packaging of bacterial genes near the integration site into a phage head?

A. conjugation
B. generalized transduction
C. specialized transduction
D. transformation

Answer

C

Which of the following refers to the uptake of naked DNA from the surrounding environment?

A. conjugation
B. generalized transduction
C. specialized transduction
D. transformation

Answer

D

The F plasmid is involved in which of the following processes?

A. conjugation
B. transduction
C. transposition
D. transformation

Answer

A

Which of the following refers to the mechanism of horizontal gene transfer naturally responsible for the spread of antibiotic resistance genes within a bacterial population?

A. conjugation
B. generalized transduction
C. specialized transduction
D. transformation

Answer

A

Fill in the Blank

A small DNA molecule that has the ability to independently excise from one location in a larger DNA molecule and integrate into the DNA elsewhere is called a _____.

Answer

transposon or transposable element

_____ is a group of mechanisms that allow for the introduction of genetic material from one organism to another organism within the same generation.

Answer

Horizontal gene transfer

True/False

Asexually reproducing organisms lack mechanisms for generating genetic diversity within a population.

Answer

False

Short Answer

Briefly describe two ways in which chromosomal DNA from a donor cell may be transferred to a recipient cell during the process of conjugation.

Describe what happens when a nonsense mutation is introduced into the gene encoding transposase within a transposon.

13.3: Gene Regulation - Operon Theory

Genomic DNA contains both structural genes, which encode products that serve as cellular structures or enzymes, and regulatory genes, which encode products that regulate gene expression. The expression of a gene is a highly regulated process. Whereas regulating gene expression in multicellular organisms allows for cellular differentiation, in single-celled organisms like prokaryotes, it ensures that a cell's resources are not wasted making proteins that the cell does not need at that time.

Multiple Choice

An operon of genes encoding enzymes in a biosynthetic pathway is likely to be which of the following?

A. inducible
B. repressible
C. constitutive
D. monocistronic

Answer

B

An operon encoding genes that are transcribed and translated continuously to provide the cell with constant intermediate levels of the protein products is said to be which of the following?

A. repressible
B. inducible
C. constitutive
D. activated

Answer

C

Which of the following conditions leads to maximal expression of the *lac* operon?

A. lactose present, glucose absent
B. lactose present, glucose present
C. lactose absent, glucose absent
D. lactose absent, glucose present

Answer

A

Which of the following is a type of regulation of gene expression unique to eukaryotes?

A. attenuation
B. use of alternate σ factor
C. chemical modification of histones
D. alarmones

Answer

C

Fill in the Blank

The DNA sequence, to which repressors may bind, that lies between the promoter and the first structural gene is called the _____.

Answer

operator

The prevention of expression of operons encoding substrate use pathways for substrates other than glucose when glucose is present is called _____.

Answer

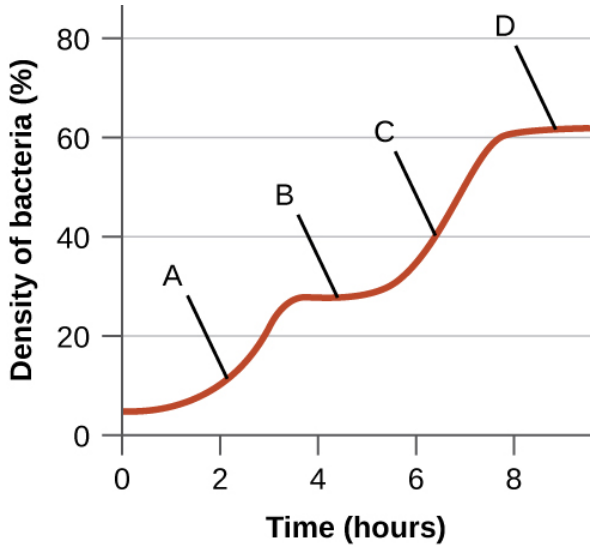
catabolite repression

Short Answer

What are two ways that bacteria can influence the transcription of multiple different operons simultaneously in response to a particular environmental condition?

Critical Thinking

The following figure is from Monod's original work on diauxic growth showing the growth of *E. coli* in the simultaneous presence of xylose and glucose as the only carbon sources. Explain what is happening at points A–D with respect to the carbon source being used for growth, and explain whether the xylose-use operon is being expressed (and why). Note that expression of the enzymes required for xylose use is regulated in a manner similar to the expression of the enzymes required for lactose use.



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CHAPTER OVERVIEW

15: Antibiotics

In nature, some microbes produce substances that inhibit or kill other microbes that might otherwise compete for the same resources. Humans have successfully exploited these abilities, using microbes to mass-produce substances that can be used as antimicrobial drugs. Since their discovery, antimicrobial drugs have saved countless lives, and they remain an essential tool for treating and controlling infectious disease. But their widespread and often unnecessary use has had an unintended side effect: the rise of multidrug-resistant microbial strains. In this chapter, we will discuss how antimicrobial drugs work, why microbes develop resistance, and what health professionals can do to encourage responsible use of antimicrobials.



Figure **15.1**: First mass produced in the 1940s, penicillin was instrumental in saving millions of lives during World War II and was considered a wonder drug.[1] Today, overprescription of antibiotics (especially for childhood illnesses) has contributed to the evolution of drug-resistant pathogens. (credit left: modification of work by Chemical Heritage Foundation; Credit right: DFID / Flickr; CC-BY)

15.1: Discovering Antimicrobial Drugs
15.2: Antibacterial Drugs
15.3: Antibacterial Drugs
15.4: Drugs Targeting Other Microorganisms
15.5: Clinical Considerations
15.6: Testing the Effectiveness of Antimicrobials
15.E: Antimicrobial Drugs (Exercises)

Footnotes

1. 1 "Treatment of War Wounds: A Historical Review." *Clinical Orthopaedics and Related Research* 467 no. 8 (2009):2168–2191.

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15.1: Discovering Antimicrobial Drugs

Learning Objectives

- Compare and contrast natural, semisynthetic, and synthetic antimicrobial drugs
- Describe the chemotherapeutic approaches of ancient societies
- Describe the historically important individuals and events that led to the development of antimicrobial drugs

Most people associate the term chemotherapy with treatments for cancer. However, chemotherapy is actually a broader term that refers to any use of chemicals or drugs to treat disease. Chemotherapy may involve drugs that target cancerous cells or tissues, or it may involve **antimicrobial drugs** that target infectious microorganisms. Antimicrobial drugs typically work by destroying or interfering with microbial structures and enzymes, either killing microbial cells or inhibiting of their growth. But before we examine how these drugs work, we will briefly explore the history of humans' use of antimicrobials for the purpose of chemotherapy.

Use of Antimicrobials in Ancient Societies

Although the discovery of antimicrobials and their subsequent widespread use is commonly associated with modern medicine, there is evidence that humans have been exposed to antimicrobial compounds for millennia. Chemical analyses of the skeletal remains of people from Nubia[1] (now found in present-day Sudan) dating from between 350 and 550 AD have shown residue of the antimicrobial agent tetracycline in high enough quantities to suggest the purposeful fermentation of tetracycline-producing *Streptomyces* during the beer-making process. The resulting beer, which was thick and gruel-like, was used to treat a variety of ailments in both adults and children, including gum disease and wounds. The antimicrobial properties of certain plants may also have been recognized by various cultures around the world, including Indian and Chinese herbalists (Figure **15.1.1**) who have long used plants for a wide variety of medical purposes. Healers of many cultures understood the antimicrobial properties of fungi and their use of moldy bread or other mold-containing products to treat wounds has been well documented for centuries.[2] Today, while about 80% of the world's population still relies on plant-derived medicines,[3] scientists are now discovering the active compounds conferring the medicinal benefits contained in many of these traditionally used plants.



Figure **15.1.1**: For millennia, Chinese herbalists have used many different species of plants for the treatment of a wide variety of human ailments.

The First Antimicrobial Drugs

Societies relied on traditional medicine for thousands of years; however, the first half of the 20th century brought an era of strategic drug discovery. In the early 1900s, the German physician and scientist Paul Ehrlich (1854–1915) set out to discover or synthesize chemical compounds capable of killing infectious microbes without harming the patient. In 1909, after screening more than 600 arsenic-containing compounds, Ehrlich's assistant Sahachiro Hata (1873–1938) found one such "magic bullet." Compound 606 targeted the bacterium *Treponema pallidum*, the causative agent of syphilis. Compound 606 was found to successfully cure syphilis

in rabbits and soon after was marketed under the name Salvarsan as a remedy for the disease in humans (Figure **15.1.2**). Ehrlich's innovative approach of systematically screening a wide variety of compounds remains a common strategy for the discovery of new antimicrobial agents even today.

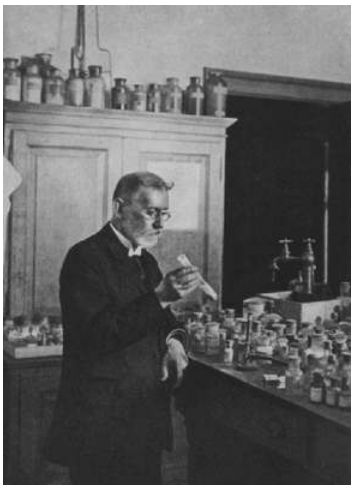
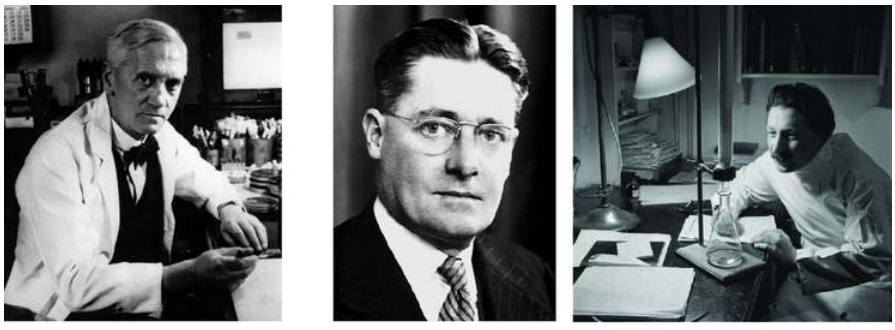


Figure **15.1.2**: Paul Ehrlich was influential in the discovery of Compound 606, an antimicrobial agent that proved to be an effective treatment for syphilis.

A few decades later, German scientists Josef Klarer, Fritz Mietzsch, and Gerhard Domagk discovered the antibacterial activity of a synthetic dye, prontosil, that could treat streptococcal and staphylococcal infections in mice. Domagk's own daughter was one of the first human recipients of the drug, which completely cured her of a severe streptococcal infection that had resulted from a poke with an embroidery needle. Gerhard Domagk (1895–1964) was awarded the Nobel Prize in Medicine in 1939 for his work with prontosil and sulfanilamide, the active breakdown product of prontosil in the body. Sulfanilamide, the first synthetic antimicrobial created, served as the foundation for the chemical development of a family of sulfa drugs. A **synthetic antimicrobial** is a drug that is developed from a chemical not found in nature. The success of the sulfa drugs led to the discovery and production of additional important classes of synthetic antimicrobials, including the quinolines and oxazolidinones.

A few years before the discovery of prontosil, scientist Alexander Fleming (1881–1955) made his own accidental discovery that turned out to be monumental. In 1928, Fleming returned from holiday and examined some old plates of staphylococci in his research laboratory at St. Mary's Hospital in London. He observed that contaminating mold growth (subsequently identified as a strain of *Penicillium notatum*) inhibited staphylococcal growth on one plate. Fleming, therefore, is credited with the discovery of **penicillin**, the first **natural antibiotic**, (Figure **15.1.3**). Further experimentation showed that penicillin from the mold was antibacterial against streptococci, meningococci, and *Corynebacterium diphtheriae*, the causative agent of diphtheria.



(a) (b)

Figure **15.1.3**: (a) Alexander Fleming was the first to discover a naturally produced antimicrobial, penicillin, in 1928. (b) Howard Florey and Ernst Chain discovered how to scale up penicillin production. Then they figured out how to purify it and showed its efficacy as an antimicrobial in animal and human trials in the early 1940s.

Fleming and his colleagues were credited with discovering and identifying penicillin, but its isolation and mass production were accomplished by a team of researchers at Oxford University under the direction of Howard Florey(1898–1968) and Ernst Chain (1906–1979) (Figure **15.1.3**). In 1940, the research team purified penicillin and reported its success as an antimicrobial agent

against streptococcal infections in mice. Their subsequent work with human subjects also showed penicillin to be very effective. Because of their important work, Fleming, Florey, and Chain were awarded the Nobel Prize in Physiology and Medicine in 1945.

In the early 1940s, scientist Dorothy Hodgkin (1910–1994), who studied crystallography at Oxford University, used X-rays to analyze the structure of a variety of natural products. In 1946, she determined the structure of penicillin, for which she was awarded the Nobel Prize in Chemistry in 1964. Once the structure was understood, scientists could modify it to produce a variety of **semisynthetic penicillins**. A semisynthetic antimicrobial is a chemically modified derivative of a natural antibiotic. The chemical modifications are generally designed to increase the range of bacteria targeted, increase stability, decrease toxicity, or confer other properties beneficial for treating infections.

Penicillin is only one example of a natural antibiotic. Also in the 1940s, Selman Waksman (1888–1973) (Figure **15.1.4**), a prominent soil microbiologist at Rutgers University, led a research team that discovered several antimicrobials, including actinomycin, streptomycin, and neomycin. The discoveries of these antimicrobials stemmed from Waksman's study of fungi and the Actinobacteria, including soil bacteria in the genus *Streptomyces*, known for their natural production of a wide variety of antimicrobials. His work earned him the Nobel Prize in Physiology and Medicine in 1952. The actinomycetes are the source of more than half of all natural antibiotics[4] and continue to serve as an excellent reservoir for the discovery of novel antimicrobial agents. Some researchers argue that we have not yet come close to tapping the full antimicrobial potential of this group.[5]



Figure **15.1.4**: Selman Waksman was the first to show the vast antimicrobial production capabilities of a group of soil bacteria, the actinomycetes.

Key Concepts and Summary

- **Antimicrobial drugs** produced by purposeful fermentation and/or contained in plants have been used as traditional medicines in many cultures for millennia.
- The purposeful and systematic search for a chemical "magic bullet" that specifically target infectious microbes was initiated by Paul Ehrlich in the early 20th century.
- The discovery of the **natural antibiotic**, penicillin, by Alexander Fleming in 1928 started the modern age of antimicrobial discovery and research.
- Sulfanilamide, the first **synthetic antimicrobial**, was discovered by Gerhard Domagk and colleagues and is a breakdown product of the synthetic dye, prontosil.

Footnotes

1. 1 M.L. Nelson et al. "Brief Communication: Mass Spectroscopic Characterization of Tetracycline in the Skeletal Remains of an Ancient Population from Sudanese Nubia 350–550 CE." *American Journal of Physical Anthropology* 143 no. 1 (2010):151–154.
2. 2 M. Wainwright. "Moulds in Ancient and More Recent Medicine." *Mycologist* 3 no. 1 (1989):21–23.
3. 3 S. Verma, S.P. Singh. "Current and Future Status of Herbal Medicines." *Veterinary World* 1 no. 11 (2008):347–350.
4. 4 J. Berdy. "Bioactive Microbial Metabolites." *The Journal of Antibiotics* 58 no. 1 (2005):1–26.

5. 5 M. Baltz. “Antimicrobials from Actinomycetes: Back to the Future.” *Microbe* 2 no. 3 (2007):125–131.

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15.2: Antibacterial Drugs

Learning Objectives

- Contrast bacteriostatic versus bactericidal antibacterial activities
- Contrast broad-spectrum drugs versus narrow-spectrum drugs
- Explain the significance of superinfections
- Discuss the significance of dosage and the route of administration of a drug
- Identify factors and variables that can influence the side effects of a drug
- Describe the significance of positive and negative interactions between drugs

Several factors are important in choosing the most appropriate antimicrobial drug therapy, including bacteriostatic versus bactericidal mechanisms, spectrum of activity, dosage and route of administration, the potential for side effects, and the potential interactions between drugs. The following discussion will focus primarily on antibacterial drugs, but the concepts translate to other antimicrobial classes.

Bacteriostatic Versus Bactericidal

Antibacterial drugs can be either bacteriostatic or bactericidal in their interactions with target bacteria. **Bacteriostatic** drugs cause a reversible inhibition of growth, with bacterial growth restarting after elimination of the drug. By contrast, **bactericidal** drugs kill their target bacteria. The decision of whether to use a bacteriostatic or bactericidal drugs depends on the type of infection and the immune status of the patient. In a patient with strong immune defenses, bacteriostatic and bactericidal drugs can be effective in achieving clinical cure. However, when a patient is immunocompromised, a bactericidal drug is essential for the successful treatment of infections. Regardless of the immune status of the patient, life-threatening infections such as acute endocarditis require the use of a bactericidal drug.

Spectrum of Activity

The spectrum of activity of an antibacterial drug relates to diversity of targeted bacteria. A **narrow-spectrum antimicrobial** targets only specific subsets of bacterial pathogens. For example, some narrow-spectrum drugs only target gram-positive bacteria, whereas others target only gram-negative bacteria. If the pathogen causing an infection has been identified, it is best to use a narrow-spectrum antimicrobial and minimize collateral damage to the normal microbiota. A **broad-spectrum antimicrobial** targets a wide variety of bacterial pathogens, including both gram-positive and gram-negative species, and is frequently used as empiric therapy to cover a wide range of potential pathogens while waiting on the laboratory identification of the infecting pathogen. Broad-spectrum antimicrobials are also used for polymicrobic infections (mixed infection with multiple bacterial species), or as prophylactic prevention of infections with surgery/invasive procedures. Finally, broad-spectrum antimicrobials may be selected to treat an infection when a narrow-spectrum drug fails because of development of drug resistance by the target pathogen.

The risk associated with using broad-spectrum antimicrobials is that they will also target a broad spectrum of the normal microbiota, increasing the risk of a **superinfection**, a secondary infection in a patient having a preexisting infection. A superinfection develops when the antibacterial intended for the preexisting infection kills the protective microbiota, allowing another pathogen resistant to the antibacterial to proliferate and cause a secondary infection (Figure **15.2.1**). Common examples of superinfections that develop as a result of antimicrobial usage include yeast infections (candidiasis) and pseudomembranous colitis caused by *Clostridium difficile*, which can be fatal.

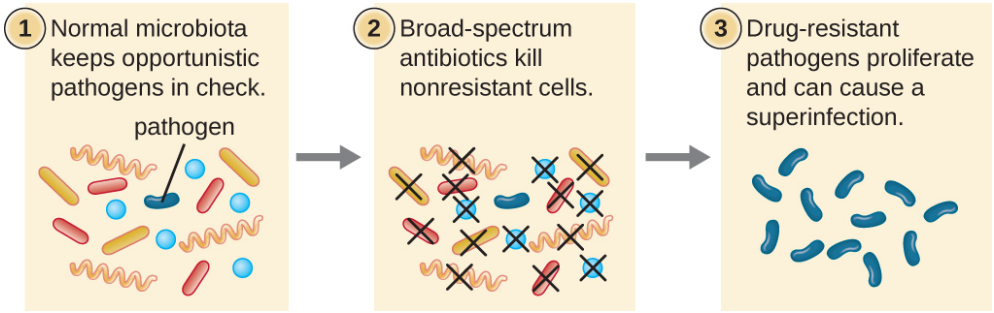


Figure 15.2.1: Broad-spectrum antimicrobial use may lead to the development of a superinfection. (credit: modification of work by Centers for Disease Control and Prevention)

Dosage and Route of Administration

The amount of medication given during a certain time interval is the **dosage**, and it must be determined carefully to ensure that optimum therapeutic drug levels are achieved at the site of infection without causing significant toxicity(side effects) to the patient. Each drug class is associated with a variety of potential side effects, and some of these are described for specific drugs later in this chapter. Despite best efforts to optimize dosing, allergic reactions and other potentially serious side effects do occur. Therefore, the goal is to select the optimum dosage that will minimize the risk of side effects while still achieving clinical cure, and there are important factors to consider when selecting the best dose and dosage interval. For example, in children, dose is based upon the patient's mass. However, the same is not true for adults and children 12 years of age and older, for which there is typically a single standard dose regardless of the patient's mass. With the great variability in adult body mass, some experts have argued that mass should be considered for all patients when determining appropriate dosage.[1] An additional consideration is how drugs are metabolized and eliminated from the body. In general, patients with a history of liver or kidney dysfunction may experience reduced drug metabolism or clearance from the body, resulting in increased drug levels that may lead to toxicity and make them more prone to side effects.

There are also some factors specific to the drugs themselves that influence appropriate dose and time interval between doses. For example, the half-life, or rate at which 50% of a drug is eliminated from the plasma, can vary significantly between drugs. Some drugs have a short half-life of only 1 hour and must be given multiple times a day, whereas other drugs have half-lives exceeding 12 hours and can be given as a single dose every 24 hours. Although a longer half-life can be considered an advantage for an antibacterial when it comes to convenient dosing intervals, the longer half-life can also be a concern for a drug that has serious side effects because drug levels may remain toxic for a longer time. Last, some drugs are dose dependent, meaning they are more effective when administered in large doses to provide high levels for a short time at the site of infection. Others are time dependent, meaning they are more effective when lower optimum levels are maintained over a longer period of time.

The **route of administration**, the method used to introduce a drug into the body, is also an important consideration for drug therapy. Drugs that can be administered orally are generally preferred because patients can more conveniently take these drugs at home. However, some drugs are not absorbed easily from the gastrointestinal (GI) tract into the bloodstream. These drugs are often useful for treating diseases of the intestinal tract, such as tapeworms treated with niclosamide, or for decontaminating the bowel, as with colistin. Some drugs that are not absorbed easily, such as bacitracin, polymyxin, and several antifungals, are available as topical preparations for treatment of superficial skin infections. Sometimes, patients may not initially be able to take oral medications because of their illness (e.g., vomiting, intubation for respirator). When this occurs, and when a chosen drug is not absorbed in the GI tract, administration of the drug by a parenteral route (intravenous or intramuscular injection) is preferred and typically is performed in health-care settings. For most drugs, the plasma levels achieved by intravenous administration is substantially higher than levels achieved by oral or intramuscular administration, and this can also be an important consideration when choosing the route of administration for treating an infection (Figure 15.2.2).

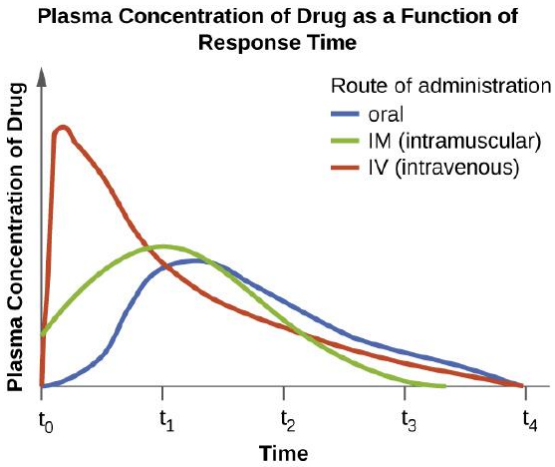


Figure **15.2.2**: On this graph, t_0 represents the time at which a drug dose is administered. The curves illustrate how plasma concentration of the drug changes over specific intervals of time (t_1 through t_4). As the graph shows, when a drug is administered intravenously, the concentration peaks very quickly and then gradually decreases. When drugs are administered orally or intramuscularly, it takes longer for the concentration to reach its peak.

Drug Interactions

For the optimum treatment of some infections, two antibacterial drugs may be administered together to provide a **synergistic interaction** that is better than the efficacy of either drug alone. A classic example of synergistic combinations is trimethoprim and sulfamethoxazole (Bactrim). Individually, these two drugs provide only bacteriostatic inhibition of bacterial growth, but combined, the drugs are bactericidal.

Whereas synergistic drug interactions provide a benefit to the patient, **antagonistic interactions** produce harmful effects. Antagonism can occur between two antimicrobials or between antimicrobials and nonantimicrobials being used to treat other conditions. The effects vary depending on the drugs involved, but antagonistic interactions may cause loss of drug activity, decreased therapeutic levels due to increased metabolism and elimination, or increased potential for toxicitydue to decreased metabolism and elimination. As an example, some antibacterials are absorbed most effectively from the acidic environment of the stomach. If a patient takes antacids, however, this increases the pH of the stomach and negatively impacts the absorption of these antimicrobials, decreasing their effectiveness in treating an infection. Studies have also shown an association between use of some antimicrobials and failure of oral contraceptives.[2]

Resistance Police

In the United States and many other countries, most antimicrobial drugs are self-administered by patients at home. Unfortunately, many patients stop taking antimicrobials once their symptoms dissipate and they feel better. If a 10-day course of treatment is prescribed, many patients only take the drug for 5 or 6 days, unaware of the negative consequences of not completing the full course of treatment. A shorter course of treatment not only fails to kill the target organisms to expected levels, it also selects for drug-resistant variants within the target population and within the patient's microbiota.

Patients' nonadherence especially amplifies drug resistance when the recommended course of treatment is long. Treatment for tuberculosis (TB) is a case in point, with the recommended treatment lasting from 6 months to a year. The CDC estimates that about one-third of the world's population is infected with TB, most living in underdeveloped or underserved regions where antimicrobial drugs are available over the counter. In such countries, there may be even lower rates of adherence than in developed areas. Nonadherence leads to antibiotic resistance and more difficulty in controlling pathogens. As a direct result, the emergence of multidrug-resistant and extensively drug-resistant strains of TB is becoming a huge problem.

Overprescription of antimicrobials also contributes to antibiotic resistance. Patients often demand antibiotics for diseases that do not require them, like viral colds and ear infections. Pharmaceutical companies aggressively market drugs to physicians and clinics, making it easy for them to give free samples to patients, and some pharmacies even offer certain antibiotics free to low-income patients with a prescription.

In recent years, various initiatives have aimed to educate parents and clinicians about the judicious use of antibiotics. However, a recent study showed that, between 2000 and 2013, the parental expectation for antimicrobial prescriptions for children actually increased (Figure **15.2.3**).

One possible solution is a regimen called directly observed therapy (DOT), which involves the supervised administration of medications to patients. Patients are either required to visit a health-care facility to receive their medications, or health-care providers must administer medication in patients' homes or another designated location. DOT has been implemented in many cases for the treatment of TB and has been shown to be effective; indeed, DOT is an integral part of WHO's global strategy for eradicating TB.[3],[4] But is this a practical strategy for all antibiotics? Would patients taking penicillin, for example, be more or less likely to adhere to the full course of treatment if they had to travel to a health-care facility for each dose? And who would pay for the increased cost associated with DOT? When it comes to overprescription, should someone be policing physicians or drug companies to enforce best practices? What group should assume this responsibility, and what penalties would be effective in discouraging overprescription?

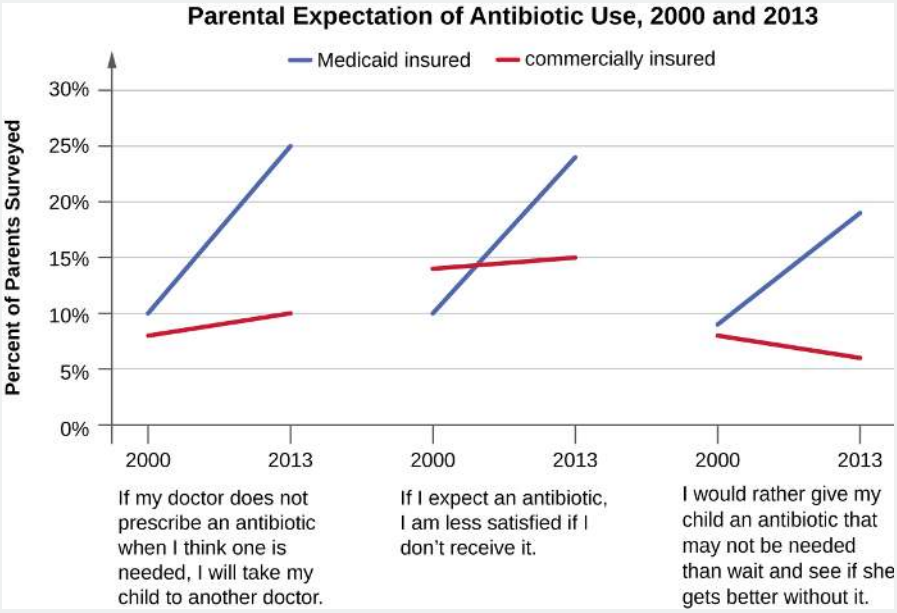


Figure **15.2.3**: This graph indicates trends in parental expectations related to prescription of antibiotics based on a recent study.[5] Among parents of Medicaid-insured children, there was a clear upward trend in parental expectations for prescription antibiotics. Expectations were relatively stable (and lesser) among parents whose children were commercially insured, suggesting that these parents were somewhat better informed than those with Medicaid-insured children.

Key Concepts and Summary

- Antimicrobial drugs can be **bacteriostatic** or **bactericidal**, and these characteristics are important considerations when selecting the most appropriate drug.
- The use of **narrow-spectrum** antimicrobial drugs is preferred in many cases to avoid **superinfection** and the development of antimicrobial resistance.
- **Broad-spectrum** antimicrobial use is warranted for serious systemic infections when there is no time to determine the causative agent, when narrow-spectrum antimicrobials fail, or for the treatment or prevention of infections with multiple types of microbes.
- The **dosage** and **route of administration** are important considerations when selecting an antimicrobial to treat and infection. Other considerations include the patient's age, mass, ability to take oral medications, liver and kidney function, and possible interactions with other drugs the patient may be taking.

Footnotes

1. 1 M.E. Falagas, D.E. Karageorgopoulos. "Adjustment of Dosing of Antimicrobial Agents for Bodyweight in Adults." *The Lancet* 375 no. 9710 (2010):248–251.

2. 2 B.D. Dickinson et al. “Drug Interactions between Oral Contraceptives and Antibiotics.” *Obstetrics & Gynecology* 98, no. 5 (2001):853–860.
3. 3 Centers for Disease Control and Prevention. “Tuberculosis (TB).” www.cdc.gov/tb/education/ssmo...s9reading2.htm. Accessed June 2, 2016.
4. 4 World Health Organization. “Tuberculosis (TB): The Five Elements of DOTS.” http://www.who.int/tb/dots/whatisdots/en/. Accessed June 2, 2016.
5. 5 Vaz, L.E., et al. “Prevalence of Parental Misconceptions About Antibiotic Use.” *Pediatrics* 136 no.2 (August 2015). DOI: 10.1542/peds.2015-0883.

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15.3: Antibacterial Drugs

15.03: Antibacterial Drugs

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15.4: Drugs Targeting Other Microorganisms

Learning Objectives

- Describe the mechanisms of action associated with drugs that inhibit cell wall biosynthesis.
- Explain the mode of action of β-lactams.
- Describe why β-lactamase (penicillinase) offers drug resistance for bacteria.
- Explain the mode of action of non-β-lactam cell wall inhibitors.

An important quality for an antimicrobial drug is **selective toxicity**, meaning that it selectively kills or inhibits the growth of microbial targets while causing minimal or no harm to the host. Most antimicrobial drugs currently in clinical use are antibacterial because the prokaryotic cell provides a greater variety of unique targets for selective toxicity, in comparison to fungi, parasites, and viruses. Each class of antibacterial drugs has a unique mode of action (the way in which a drug affects microbes at the cellular level), and these are summarized in Figure 15.4.1 and Table 15.4.1.

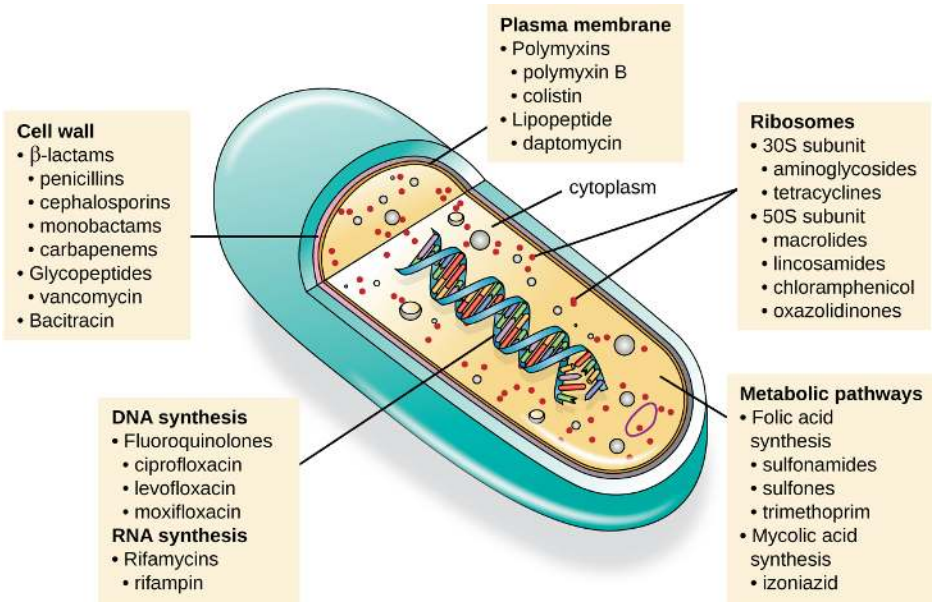


Figure 15.4.1: There are several classes of antibacterial compounds that are typically classified based on their bacterial target.

Table 15.4.1: Common Antibacterial Drugs by Mode of Action

Mode of Action	Target	Drug Class
Inhibit cell wall biosynthesis	Penicillin-binding proteins	β-lactams: penicillins, cephalosporins, monobactams, carbapenems
	Peptidoglycan subunits	Glycopeptides
	Peptidoglycan subunit transport	Bacitracin
Inhibit biosynthesis of proteins	30S ribosomal subunit	Aminoglycosides, tetracyclines
	50S ribosomal subunit	Macrolides, lincosamides, chloramphenicol, oxazolidinones
Disrupt membranes	Lipopolysaccharide, inner and outer membranes	Polymyxin B, colistin, daptomycin
Inhibit nucleic acid synthesis	RNA	Rifamycin
	DNA	Fluoroquinolones
Antimetabolites	Folic acid synthesis enzyme	Sulfonamides, trimethoprim
	Mycolic acid synthesis enzyme	Isonicotinic acid hydrazide
Mycobacterial adenosine triphosphate (ATP) synthase inhibitor	Mycobacterial ATP synthase	Diarylquinoline

Inhibitors of Cell Wall Biosynthesis

Several different classes of antibacterials block steps in the biosynthesis of peptidoglycan, making cells more susceptible to osmotic lysis (Table **15.4.2**). Therefore, antibacterials that target cell wall biosynthesis are bactericidal in their action. Because human cells do not make peptidoglycan, this mode of action is an excellent example of selective toxicity.

Penicillin, the first antibiotic discovered, is one of several antibacterials within a class called **β-lactams**. This group of compounds includes the penicillins, cephalosporins, monobactams, and carbapenems, and is characterized by the presence of a β-lactam ring found within the central structure of the drug molecule (Figure **15.4.2**). The β-lactam antibacterials block the crosslinking of peptide chains during the biosynthesis of new peptidoglycan in the bacterial cell wall. They are able to block this process because the β-lactam structure is similar to the structure of the peptidoglycan subunit component that is recognized by the crosslinking transpeptidase enzyme, also known as a penicillin-binding protein (PBP). Although the β-lactam ring must remain unchanged for these drugs to retain their antibacterial activity, strategic chemical changes to the R groups have allowed for development of a wide variety of semisynthetic β-lactam drugs with increased potency, expanded spectrum of activity, and longer half-lives for better dosing, among other characteristics.

Penicillin G and penicillin V are natural antibiotics from fungi and are primarily active against gram-positive bacterial pathogens, and a few gram-negative bacterial pathogens such as *Pasteurella multocida*. Figure **15.4.2** summarizes the semisynthetic development of some of the penicillins. Adding an amino group ($-NH_2$) to penicillin G created the aminopenicillins (i.e., ampicillin and amoxicillin) that have increased spectrum of activity against more gram-negative pathogens. Furthermore, the addition of a hydroxyl group (-OH) to amoxicillin increased acid stability, which allows for improved oral absorption. Methicillin is a semisynthetic penicillin that was developed to address the spread of enzymes (penicillinases) that were inactivating the other penicillins. Changing the R group of penicillin G to the more bulky dimethoxyphenyl group provided protection of the β-lactam ring from enzymatic destruction by penicillinases, giving us the first penicillinase-resistant penicillin.

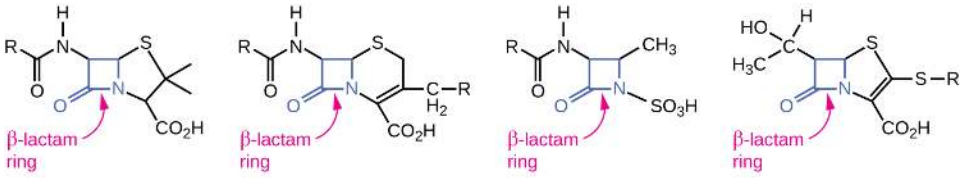
Similar to the penicillins, **cephalosporins** contain a β-lactam ring (Figure **15.4.2**) and block the transpeptidase activity of penicillin-binding proteins. However, the β-lactam ring of cephalosporins is fused to a six-member ring, rather than the five-member ring found in penicillins. This chemical difference provides cephalosporins with an increased resistance to enzymatic inactivation by **β-lactamases**. The drug cephalosporin C was originally isolated from the fungus *Cephalosporium acremonium* in the 1950s and has a

similar spectrum of activity to that of penicillin against gram-positive bacteria but is active against more gram-negative bacteria than penicillin. Another important structural difference is that cephalosporin C possesses two R groups, compared with just one R group for penicillin, and this provides for greater diversity in chemical alterations and development of semisynthetic cephalosporins. The family of semisynthetic cephalosporins is much larger than the penicillins, and these drugs have been classified into generations based primarily on their spectrum of activity, increasing in spectrum from the narrow-spectrum, first-generation cephalosporins to the broad-spectrum, fourth-generation cephalosporins. A new fifth-generation cephalosporin has been developed that is active against **methicillin-resistant *Staphylococcus aureus* (MRSA)**.

The carbapenems and monobactams also have a β-lactam ring as part of their core structure, and they inhibit the transpeptidase activity of penicillin-binding proteins. The only monobactam used clinically is aztreonam. It is a narrow-spectrum antibacterial with activity only against gram-negative bacteria. In contrast, the carbapenem family includes a variety of semisynthetic drugs (imipenem, meropenem, and doripenem) that provide very broad-spectrum activity against gram-positive and gram-negative bacterial pathogens.

The drug **vancomycin**, a member of a class of compounds called the **glycopeptides**, was discovered in the 1950s as a natural antibiotic from the actinomycete *Amycolatopsis orientalis*. Similar to the β-lactams, vancomycin inhibits cell wall biosynthesis and is bactericidal. However, in contrast to the β-lactams, the structure of vancomycin is not similar to that of cell-wall peptidoglycan subunits and does not directly inactivate penicillin-binding proteins. Rather, vancomycin is a very large, complex molecule that binds to the end of the peptide chain of cell wall precursors, creating a structural blockage that prevents the cell wall subunits from being incorporated into the growing N-acetylglucosamine and N-acetylmuramic acid (NAM-NAG) backbone of the peptidoglycan structure (transglycosylation). Vancomycin also structurally blocks transpeptidation. Vancomycin is bactericidal against gram-positive bacterial pathogens, but it is not active against gram-negative bacteria because of its inability to penetrate the protective outer membrane.

The drug **bacitracin** consists of a group of structurally similar peptide antibiotics originally isolated from *Bacillus subtilis*. Bacitracin blocks the activity of a specific cell-membrane molecule that is responsible for the movement of peptidoglycan precursors from the cytoplasm to the exterior of the cell, ultimately preventing their incorporation into the cell wall. Bacitracin is effective against a wide range of bacteria, including gram-positive organisms found on the skin, such as *Staphylococcus* and *Streptococcus*. Although it may be administered orally or intramuscularly in some circumstances, bacitracin has been shown to be nephrotoxic (damaging to the kidneys). Therefore, it is more commonly combined with neomycin and polymyxin in topical ointments such as Neosporin.



penicillin | cephalosporin | monobactam | carbapenem

R group	—CH_2—	CH_2—O—	—CH(—NH_2)—	—CH(—NH_2)—OH	CH_3O, CH_3O
Drug name	penicillin G	penicillin V	ampicillin	amoxicillin	methicillin
Spectrum of activity	G+ and a few G−	similar to penicillin G	G+ and more G− than penicillin	similar to ampicillin	G+ only, including β-lactamase producers
Route of administration	parenteral	oral	parenteral and oral	oral (better than ampicillin)	parenteral

Figure **15.4.2**: Penicillins, cephalosporins, monobactams, and carbapenems all contain a β-lactam ring, the site of attack by inactivating β-lactamase enzymes. Although they all share the same nucleus, various penicillins differ from each other in the structure of their R groups. Chemical changes to the R groups provided increased spectrum of activity, acid stability, and resistance to β-lactamase degradation.

Table **15.4.2**: Drugs that Inhibit Bacterial Cell Wall Synthesis

Mechanism of Action	Drug Class	Specific Drugs	Natural or Semisynthetic	Spectrum of Activity
Interact directly with PBPs and inhibit transpeptidase activity	Penicillins	Penicillin G, penicillin V	Natural	Narrow-spectrum against gram-positive and a few gram-negative bacteria
		Ampicillin, amoxicillin	Semisynthetic	Narrow-spectrum against gram-positive bacteria but with increased gram-negative spectrum
		Methicillin	Semisynthetic	Narrow-spectrum against gram-positive bacteria only, including strains producing penicillinase
	Cephalosporins	Cephalosporin C	Natural	Narrow-spectrum similar to penicillin but with increased gram-negative spectrum
		First-generation cephalosporins	Semisynthetic	Narrow-spectrum similar to cephalosporin C
		Second-generation cephalosporins	Semisynthetic	Narrow-spectrum but with increased gram-negative spectrum compared with first generation
		Third- and fourth-generation cephalosporins	Semisynthetic	Broad-spectrum against gram-positive and gram-negative bacteria, including some β-lactamase producers
		Fifth-generation cephalosporins	Semisynthetic	Broad-spectrum against gram-positive and gram-negative bacteria, including MRSA
	Monobactams	Aztreonam	Semisynthetic	Narrow-spectrum against gram-negative bacteria, including some β-lactamase producers
	Carbapenems	Imipenem, meropenem, doripenem	Semisynthetic	Broadest spectrum of the β-lactams against gram-positive and gram-negative bacteria, including many β-lactamase producers
Large molecules that bind to the peptide chain of peptidoglycan subunits, blocking	Glycopeptides	Vancomycin	Natural	Narrow spectrum against gram-positive bacteria only, including multidrug-resistant strains

Mechanism of Action	Drug Class	Specific Drugs	Natural or Semisynthetic	Spectrum of Activity
transglycosylation and transpeptidation				
Block transport of peptidoglycan subunits across cytoplasmic membrane	Bacitracin	Bacitracin	Natural	Broad-spectrum against gram-positive and gram-negative bacteria

Key Concepts and Summary

- Antibacterial compounds exhibit **selective toxicity**, largely due to differences between prokaryotic and eukaryotic cell structure.
- Cell wall synthesis inhibitors, including the **β-lactams**, the **glycopeptides**, and **bacitracin**, interfere with peptidoglycan synthesis, making bacterial cells more prone to osmotic lysis.

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15.5: Clinical Considerations

Learning Objectives

- Explain how disruption of ergosterol biosynthesis is an effective mode of action for antifungals
- Describe how certain nucleoside analogs act as effective antiviral drugs
- Explain antiretroviral therapy (ART) for treating HIV infections

Because fungi, protozoa, and helminths are eukaryotic, their cells are very similar to human cells, making it more difficult to develop drugs with selective toxicity. Additionally, viruses replicate within human host cells, making it difficult to develop drugs that are selectively toxic to viruses or virus-infected cells. Despite these challenges, there are antimicrobial drugs that target fungi, protozoa, helminths, and viruses, and some even target more than one type of microbe. Table 15.5.1, Table 15.5.2, Table 15.5.3, and Table 15.5.4 provide examples for antimicrobial drugs in these various classes.

Antifungal Drugs

The most common mode of action for antifungal drugs is the disruption of the cell membrane. Antifungals take advantage of small differences between fungi and humans in the biochemical pathways that synthesize sterols. The sterols are important in maintaining proper membrane fluidity and, hence, proper function of the cell membrane. For most fungi, the predominant membrane sterol is **ergosterol**. Because human cell membranes use **cholesterol**, instead of ergosterol, antifungal drugs that target ergosterol synthesis are selectively toxic (Figure 15.5.1).

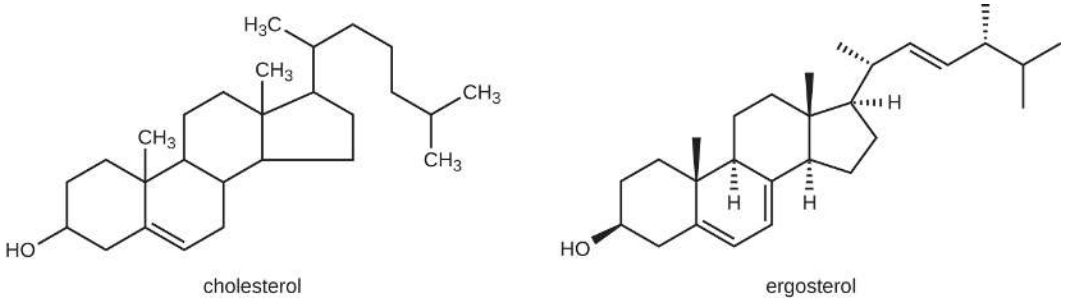


Figure 15.5.1: The predominant sterol found in human cells is cholesterol, whereas the predominant sterol found in fungi is ergosterol, making ergosterol a good target for antifungal drug development.

The **imidazoles** are synthetic fungicides that disrupt ergosterol biosynthesis; they are commonly used in medical applications and also in agriculture to keep seeds and harvested crops from molding. Examples include miconazole, ketoconazole, and clotrimazole, which are used to treat fungal skin infections such as ringworm, specifically tinea pedis (athlete's foot), tinea cruris (jock itch), and tinea corporis. These infections are commonly caused by dermatophytes of the genera *Trichophyton*, *Epidermophyton*, and *Microsporum*. Miconazole is also used predominantly for the treatment of vaginal yeast infections caused by the fungus *Candida*, and ketoconazole is used for the treatment of tinea versicolor and dandruff, which both can be caused by the fungus *Malassezia*.

The **triazole** drugs, including **fluconazole**, also inhibit ergosterol biosynthesis. However, they can be administered orally or intravenously for the treatment of several types of systemic yeast infections, including oral thrush and cryptococcal meningitis, both of which are prevalent in patients with AIDS. The triazoles also exhibit more selective toxicity, compared with the imidazoles, and are associated with fewer side effects.

The **allylamines**, a structurally different class of synthetic antifungal drugs, inhibit an earlier step in ergosterol biosynthesis. The most commonly used allylamine is **terbinafine** (marketed under the brand name Lamisil), which is used topically for the treatment of dermatophytic skin infections like athlete's foot, ringworm, and jock itch. Oral treatment with terbinafine is also used for the treatment of fingernail and toenail fungus, but it can be associated with the rare side effect of hepatotoxicity.

The **polyenes** are a class of antifungal agents naturally produced by certain actinomycete soil bacteria and are structurally related to macrolides. These large, lipophilic molecules bind to ergosterol in fungal cytoplasmic membranes, thus creating pores. Common examples include nystatin and **amphotericin B**. Nystatin is typically used as a topical treatment for yeast infections of the skin, mouth, and vagina, but may also be used for intestinal fungal infections. The drug amphotericin B is used for systemic fungal

infections like aspergillosis, cryptococcal meningitis, histoplasmosis, blastomycosis, and candidiasis. Amphotericin B was the only antifungal drug available for several decades, but its use is associated with some serious side effects, including nephrotoxicity (kidney toxicity).

Amphotericin B is often used in combination with flucytosine, a fluorinated pyrimidine analog that is converted by a fungal-specific enzyme into a toxic product that interferes with both DNA replication and protein synthesis in fungi. Flucytosine is also associated with hepatotoxicity (liver toxicity) and bone marrow depression.

Beyond targeting ergosterol in fungal cell membranes, there are a few antifungal drugs that target other fungal structures (Figure **15.5.2**). The echinocandins, including caspofungin, are a group of naturally produced antifungal compounds that block the synthesis of β(1→3) glucan found in fungal cell walls but not found in human cells. This drug class has the nickname "penicillin for fungi." Caspofungin is used for the treatment of aspergillosis as well as systemic yeast infections.

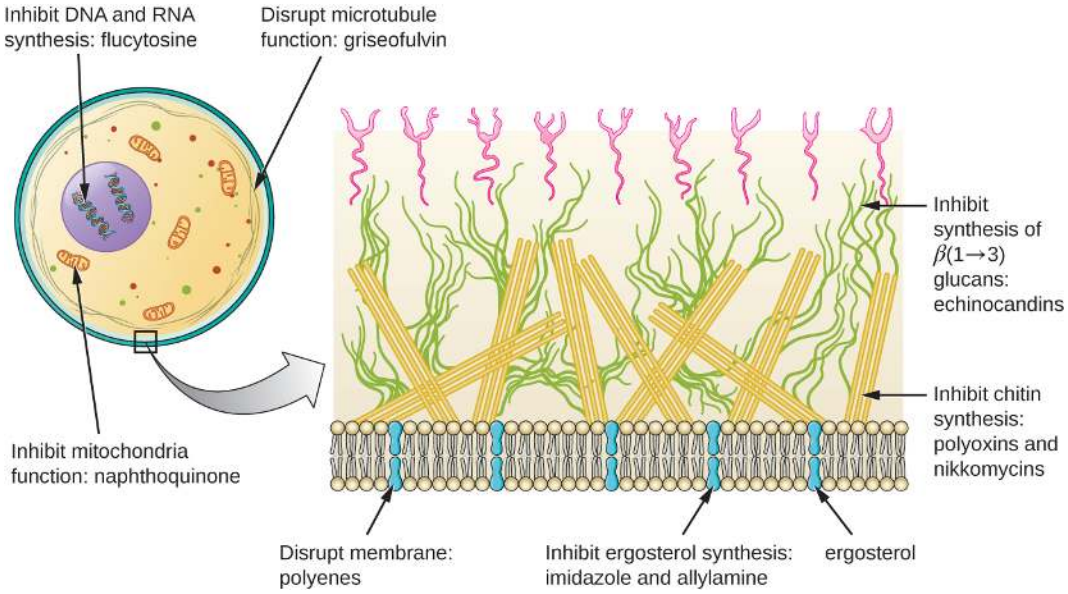


Figure **15.5.2**: Antifungal drugs target several different cell structures. (credit right: modification of work by "Maya and Rike"/Wikimedia Commons)

Although chitin is only a minor constituent of fungal cell walls, it is also absent in human cells, making it a selective target. The polyoxins and nikkomycins are naturally produced antifungals that target chitin synthesis. Polyoxins are used to control fungi for agricultural purposes, and nikkomycin Z is currently under development for use in humans to treat yeast infections and Valley fever (coccidioidomycosis), a fungal disease prevalent in the southwestern US.[1]

The naturally produced antifungal griseofulvin is thought to specifically disrupt fungal cell division by interfering with microtubules involved in spindle formation during mitosis. It was one of the first antifungals, but its use is associated with hepatotoxicity. It is typically administered orally to treat various types of dermatophytic skin infections when other topical antifungal treatments are ineffective.

There are a few drugs that act as antimetabolites against fungal processes. For example, atovaquone, a representative of the naphthoquinone drug class, is a semisynthetic antimetabolite for fungal and protozoal versions of a mitochondrial cytochrome important in electron transport. Structurally, it is an analog of coenzyme Q, with which it competes for electron binding. It is particularly useful for the treatment of *Pneumocystis* pneumonia caused by *Pneumocystis jirovecii*. The antibacterial sulfamethoxazole-trimethoprim combination also acts as an antimetabolite against *P. jirovecii*.

Table **15.5.1** shows the various therapeutic classes of antifungal drugs, categorized by mode of action, with examples of each.

Table **15.5.1**: Common Antifungal Drugs

Mechanism of Action	Drug Class	Specific Drugs	Clinical Uses
Inhibit ergosterol synthesis	Imidazoles	Miconazole, ketoconazole, clotrimazole	Fungal skin infections and vaginal yeast infections
	Triazoles	Fluconazole	Systemic yeast infections, oral thrush, and cryptococcal meningitis
	Allylamines	Terbinafine	Dermatophytic skin infections (athlete's foot, ring worm, jock itch), and infections of fingernails and toenails
Bind ergosterol in the cell membrane and create pores that disrupt the membrane	Polyenes	Nystatin	Used topically for yeast infections of skin, mouth, and vagina; also used for fungal infections of the intestine
		Amphotericin B	Variety systemic fungal infections
Inhibit cell wall synthesis	Echinocandins	Caspofungin	Aspergillosis and systemic yeast infections
	Not applicable	Nikkomycin Z	Coccidioidomycosis (Valley fever) and yeast infections
Inhibit microtubules and cell division	Not applicable	Griseofulvin	Dermatophytic skin infections

Treating a Fungal Infection of the Lungs

Jack, a 48-year-old engineer, is HIV positive but generally healthy thanks to antiretroviral therapy (ART). However, after a particularly intense week at work, he developed a fever and a dry cough. He assumed that he just had a cold or mild flu due to overexertion and didn't think much of it. However, after about a week, he began to experience fatigue, weight loss, and shortness of breath. He decided to visit his physician, who found that Jack had a low level of blood oxygenation. The physician ordered blood testing, a chest X-ray, and the collection of an induced sputum sample for analysis. His X-ray showed a fine cloudiness and several pneumatoceles (thin-walled pockets of air), which indicated *Pneumocystis* pneumonia (PCP), a type of pneumonia caused by the fungus *Pneumocystis jirovecii*. Jack's physician admitted him to the hospital and prescribed Bactrim, a combination of sulfamethoxazole and trimethoprim, to be administered intravenously.

P. jirovecii is a yeast-like fungus with a life cycle similar to that of protozoans. As such, it was classified as a protozoan until the 1980s. It lives only in the lung tissue of infected persons and is transmitted from person to person, with many people exposed as children. Typically, *P. jirovecii* only causes pneumonia in immunocompromised individuals. Healthy people may carry the fungus in their lungs with no symptoms of disease. PCP is particularly problematic among HIV patients with compromised immune systems.

PCP is usually treated with oral or intravenous Bactrim, but atovaquone or pentamidine(another antiparasitic drug) are alternatives. If not treated, PCP can progress, leading to a collapsed lung and nearly 100% mortality. Even with antimicrobial drug therapy, PCP still is responsible for 10% of HIV-related deaths.

The cytological examination, using direct immunofluorescence assay (DFA), of a smear from Jack's sputum sample confirmed the presence of *P. jirovecii* (Figure **15.5.3**). Additionally, the results of Jack's blood tests revealed that his white blood cell count had dipped, making him more susceptible to the fungus. His physician reviewed his ART regimen and made adjustments. After

a few days of hospitalization, Jack was released to continue his antimicrobial therapy at home. With the adjustments to his ART therapy, Jack's CD4 counts began to increase and he was able to go back to work.

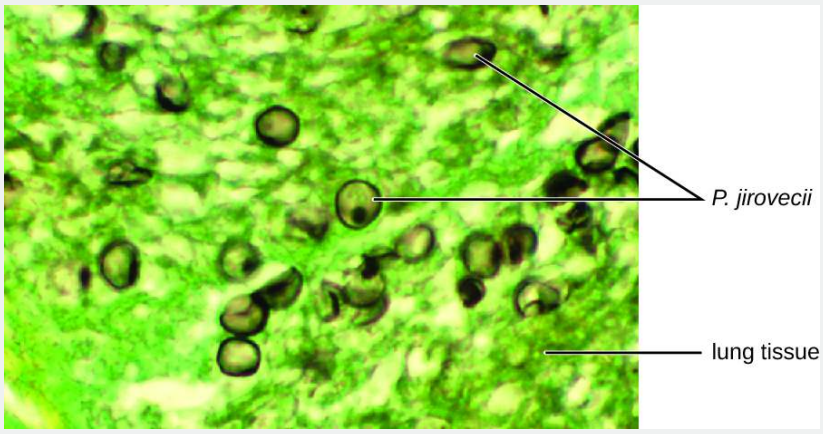


Figure 15.5.3: Microscopic examination of an induced sputum sample or bronchoaveolar lavage sample typically reveals the organism, as shown here. (credit: modification of work by the Centers for Disease Control and Prevention)

Antiviral Drugs

Unlike the complex structure of fungi, protozoa, and helminths, viral structure is simple, consisting of nucleic acid, a protein coat, viral enzymes, and, sometimes, a lipid envelope. Furthermore, viruses are obligate intracellular pathogens that use the host's cellular machinery to replicate. These characteristics make it difficult to develop drugs with selective toxicity against viruses.

Many antiviral drugs are **nucleoside analogs** and function by inhibiting nucleic acid biosynthesis. For example, **acyclovir**(marketed as Zovirax) is a synthetic analog of the nucleoside guanosine (Figure 15.5.4). It is activated by the herpes simplex viral enzyme thymidine kinase and, when added to a growing DNA strand during replication, causes chain termination. Its specificity for virus-infected cells comes from both the need for a viral enzyme to activate it and the increased affinity of the activated form for viral DNA polymerase compared to host cell DNA polymerase. Acyclovir and its derivatives are frequently used for the treatment of herpes virus infections, including genital herpes, chickenpox, shingles, Epstein-Barr virus infections, and cytomegalovirus infections. Acyclovir can be administered either topically or systemically, depending on the infection. One possible side effect of its use includes nephrotoxicity. The drug adenine-arabinoside, marketed as vidarabine, is a synthetic analog to deoxyadenosine that has a mechanism of action similar to that of acyclovir. It is also effective for the treatment of various human herpes viruses. However, because of possible side effects involving low white blood cell counts and neurotoxicity, treatment with acyclovir is now preferred.

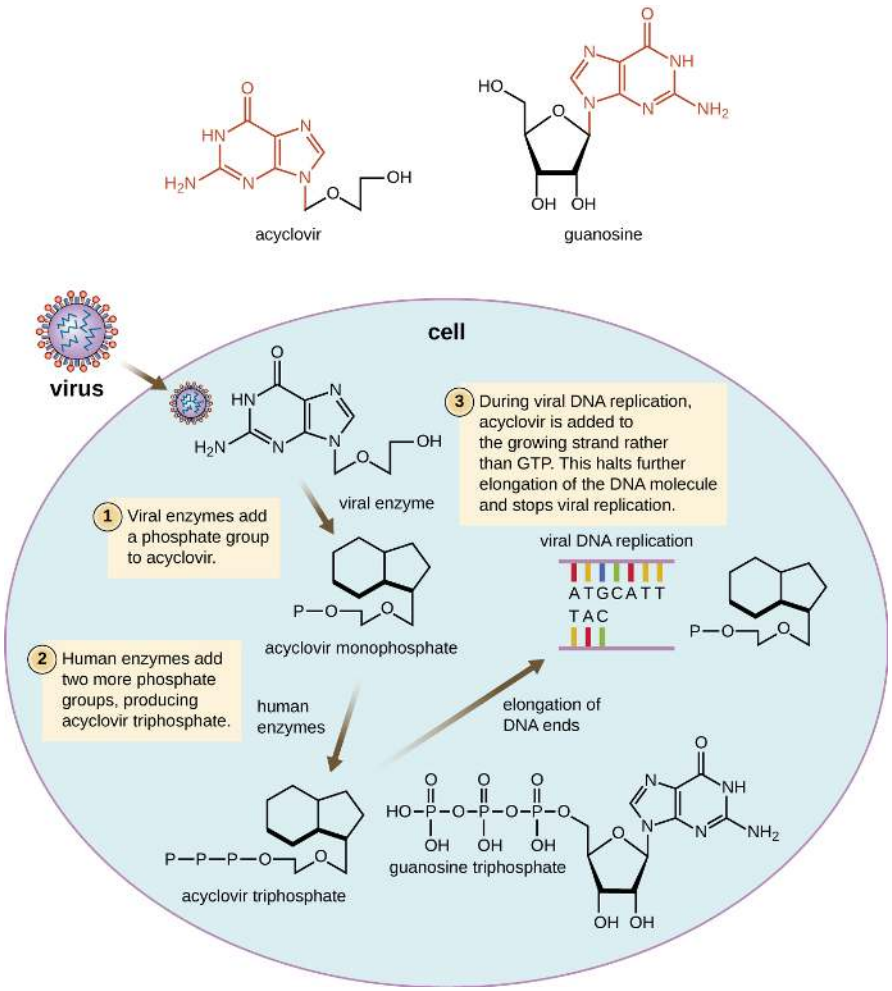


Figure 15.5.4: Acyclovir is a structural analog of guanosine. It is specifically activated by the viral enzyme thymidine kinase and then preferentially binds to viral DNA polymerase, leading to chain termination during DNA replication.

Ribavirin, another synthetic guanosine analog, works by a mechanism of action that is not entirely clear. It appears to interfere with both DNA and RNA synthesis, perhaps by reducing intracellular pools of guanosine triphosphate (GTP). Ribavarin also appears to inhibit the RNA polymerase of hepatitis C virus. It is primarily used for the treatment of the RNA viruses like hepatitis C (in combination therapy with interferon) and respiratory syncytial virus. Possible side effects of ribavirin use include anemia and developmental effects on unborn children in pregnant patients. In recent years, another nucleotide analog, sofosbuvir (Solvaldi), has also been developed for the treatment of hepatitis C. Sofosbuvir is a uridine analog that interferes with viral polymerase activity. It is commonly coadministered with ribavirin, with and without interferon.

Inhibition of nucleic acid synthesis is not the only target of synthetic antivirals. Although the mode of action of **amantadine** and its relative **rimantadine** are not entirely clear, these drugs appear to bind to a transmembrane protein that is involved in the escape of the influenza virus from endosomes. Blocking escape of the virus also prevents viral RNA release into host cells and subsequent viral replication. Increasing resistance has limited the use of amantadine and rimantadine in the treatment of influenza A. Use of amantadine can result in neurological side effects, but the side effects of rimantadine seem less severe. Interestingly, because of their effects on brain chemicals such as dopamine and NMDA (N-methyl D-aspartate), amantadine and rimantadine are also used for the treatment of Parkinson's disease.

Neuraminidase inhibitors, including olsetamivir (Tamiflu), zanamivir (Relenza), and peramivir (Rapivab), specifically target **influenza viruses** by blocking the activity of influenza virus neuraminidase, preventing the release of the virus from infected cells. These three antivirals can decrease flu symptoms and shorten the duration of illness, but they differ in their modes of administration: olsetamivir is administered orally, zanamivir is inhaled, and peramivir is administered intravenously. Resistance to these neuraminidase inhibitors still seems to be minimal.

Pleconaril is a synthetic antiviral under development that showed promise for the treatment of picornaviruses. Use of pleconaril for the treatment of the common cold caused by rhinoviruses was not approved by the FDA in 2002 because of lack of proven effectiveness, lack of stability, and association with irregular menstruation. Its further development for this purpose was halted in 2007. However, pleconaril is still being investigated for use in the treatment of life-threatening complications of enteroviruses, such as meningitis and sepsis. It is also being investigated for use in the global eradication of a specific enterovirus, polio.[6] Pleconaril seems to work by binding to the viral capsid and preventing the uncoating of viral particles inside host cells during viral infection.

Viruses with complex life cycles, such as HIV, can be more difficult to treat. First, HIV targets CD4-positive white blood cells, which are necessary for a normal immune response to infection. Second, HIV is a retrovirus, meaning that it converts its RNA genome into a DNA copy that integrates into the host cell's genome, thus hiding within host cell DNA. Third, the HIV reverse transcriptase lacks proofreading activity and introduces mutations that allow for rapid development of antiviral drug resistance. To help prevent the emergence of resistance, a combination of specific synthetic antiviral drugs is typically used in ART for HIV (Figure 15.5.5).

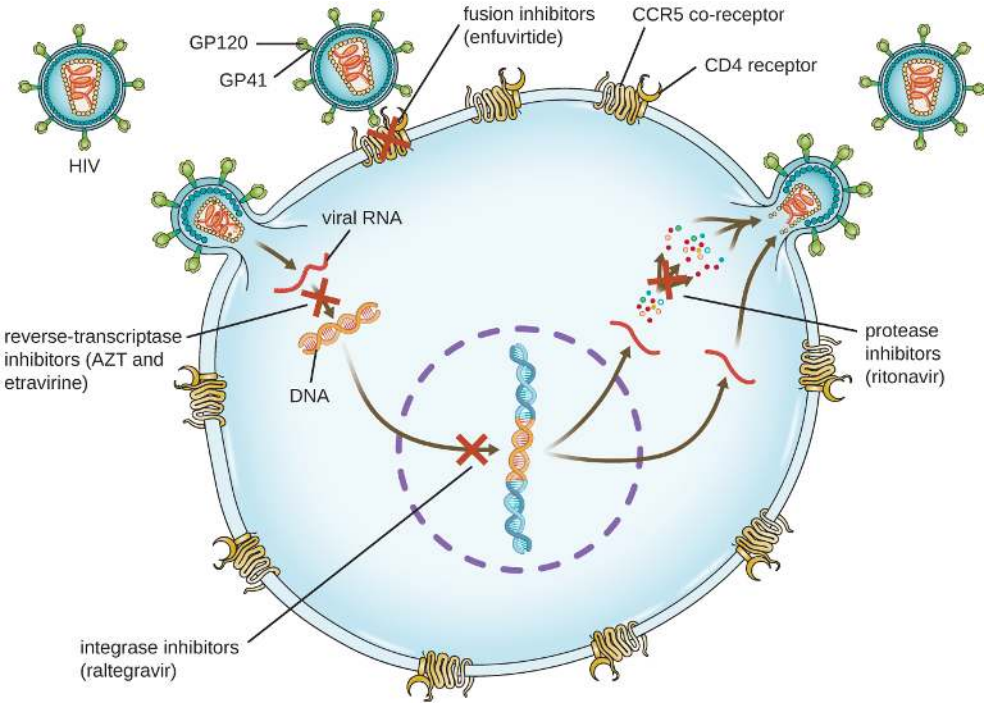


Figure 15.5.5: Antiretroviral therapy (ART) is typically used for the treatment of HIV. The targets of drug classes currently in use are shown here. (credit: modification of work by Thomas Splettstoesser)

The **reverse transcriptase inhibitors** block the early step of converting viral RNA genome into DNA, and can include competitive nucleoside analog inhibitors (e.g., azidothymidine/zidovudine, or AZT) and non-nucleoside noncompetitive inhibitors (e.g., etravirine) that bind reverse transcriptase and cause an inactivating conformational change. Drugs called **protease inhibitors** (e.g., ritonavir) block the processing of viral proteins and prevent viral maturation. Protease inhibitors are also being developed for the treatment of other viral types.[7] For example, simeprevir (Olysio) has been approved for the treatment of hepatitis C and is administered with ribavirin and interferon in combination therapy. The **integrase inhibitors** (e.g., raltegravir), block the activity of the HIV integrase responsible for the recombination of a DNA copy of the viral genome into the host cell chromosome. Additional drug classes for HIV treatment include the CCR5 antagonists and the **fusion inhibitors** (e.g., enfuviritide), which prevent the binding of HIV to the host cell coreceptor (chemokine receptor type 5 [CCR5]) and the merging of the viral envelope with the host cell membrane, respectively. Table 15.5.4 shows the various therapeutic classes of antiviral drugs, categorized by mode of action, with examples of each.

Table 15.5.4: Common Antiviral Drugs

Mechanism of Action	Drug	Clinical Uses
Nucleoside analog inhibition of nucleic acid synthesis	Acyclovir	Herpes virus infections
	Azidothymidine/zidovudine (AZT)	HIV infections
	Ribavirin	Hepatitis C virus and respiratory syncytial virus infections
	Vidarabine	Herpes virus infections
	Sofosbuvir	Hepatitis C virus infections
Non-nucleoside noncompetitive inhibition	Etravirine	HIV infections
Inhibit escape of virus from endosomes	Amantadine, rimantadine	Infections with influenza virus
Inhibit neuraminadase	Olsetamivir, zanamivir, peramivir	Infections with influenza virus
Inhibit viral uncoating	Pleconaril	Serious enterovirus infections
Inhibition of protease	Ritonavir	HIV infections
	Simeprevir	Hepatitis C virus infections
Inhibition of integrase	Raltegravir	HIV infections
Inhibition of membrane fusion	Enfuviritide	HIV infections

Link to Learning

To learn more about the various classes of antiretroviral drugs used in the ART of HIV infection, explore each of the drugs in the HIV drug classes provided by US Department of Health and Human Services at this website.

Key Concepts and Summary

- Because fungi, protozoans, and helminths are eukaryotic organisms like human cells, it is more challenging to develop antimicrobial drugs that specifically target them. Similarly, it is hard to target viruses because human viruses replicate inside of human cells.
- **Antifungal drugs** interfere with ergosterol synthesis, bind to ergosterol to disrupt fungal cell membrane integrity, or target cell wall-specific components or other cellular proteins.
- **Antiviral drugs** inhibit viral entry, inhibit viral uncoating, inhibit nucleic acid biosynthesis, prevent viral escape from endosomes in host cells, and prevent viral release from infected cells.
- Because it can easily mutate to become drug resistant, HIV is typically treated with a combination of several **antiretroviral drugs**, which may include **reverse transcriptase inhibitors, protease inhibitors, integrase inhibitors**, and drugs that interfere with viral binding and fusion to initiate infection.

Footnotes

1. 1 Centers for Disease Control and Prevention. “Valley Fever: Awareness Is Key.” www.cdc.gov/features/valleyfever/. Accessed June 1, 2016.
2. 2 B. Chu et al. “A Benzimidazole Derivative Exhibiting Antitumor Activity Blocks EGFR and HER2 Activity and Upregulates DR5 in Breast Cancer Cells.” *Cell Death and Disease* 6 (2015):e1686
3. 3 J.-X. Pan et al. “Niclosamide, An Old Antihelminthic Agent, Demonstrates Antitumor Activity by Blocking Multiple Signaling Pathways of Cancer Stem Cells.” *Chinese Journal of Cancer* 31 no. 4 (2012):178–184.
4. 4 F. Imperi et al. “New Life for an Old Drug: The Anthelmintic Drug Niclosamide Inhibits *Pseudomonas aeruginosa* Quorum Sensing.” *Antimicrobial Agents and Chemotherapy* 57 no. 2 (2013):996-1005.

5. 5 A. Jurgeit et al. “Niclosamide Is a Proton Carrier and Targets Acidic Endosomes with Broad Antiviral Effects.” *PLoS Pathogens* 8 no. 10 (2012):e1002976.
6. 6 M.J. Abzug. “The Enteroviruses: Problems in Need of Treatments.” *Journal of Infection* 68 no. S1 (2014):108–14.
7. 7 B.L. Pearlman. “Protease Inhibitors for the Treatment of Chronic Hepatitis C Genotype-1 Infection: The New Standard of Care.” *Lancet Infectious Diseases* 12 no. 9 (2012):717–728.

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15.6: Testing the Effectiveness of Antimicrobials

Learning Objectives

- Explain the concept of drug resistance
- Describe how microorganisms develop or acquire drug resistance
- Describe the different mechanisms of antimicrobial drug resistance

Antimicrobial resistance is not a new phenomenon. In nature, microbes are constantly evolving in order to overcome the antimicrobial compounds produced by other microorganisms. Human development of antimicrobial drugs and their widespread clinical use has simply provided another selective pressure that promotes further evolution. Several important factors can accelerate the evolution of **drug resistance**. These include the overuse and misuse of antimicrobials, inappropriate use of antimicrobials, subtherapeutic dosing, and patient noncompliance with the recommended course of treatment.

Exposure of a pathogen to an antimicrobial compound can select for chromosomal mutations conferring resistance, which can be transferred vertically to subsequent microbial generations and eventually become predominant in a microbial population that is repeatedly exposed to the antimicrobial. Alternatively, many genes responsible for drug resistance are found on plasmids or in transposons that can be transferred easily between microbes through horizontal gene transfer (see How Asexual Prokaryotes Achieve Genetic Diversity). Transposons also have the ability to move resistance genes between plasmids and chromosomes to further promote the spread of resistance.



Mechanisms for Drug Resistance

There are several common mechanisms for drug resistance, which are summarized in Figure **15.6.1**. These mechanisms include enzymatic modification of the drug, modification of the antimicrobial target, and prevention of drug penetration or accumulation.

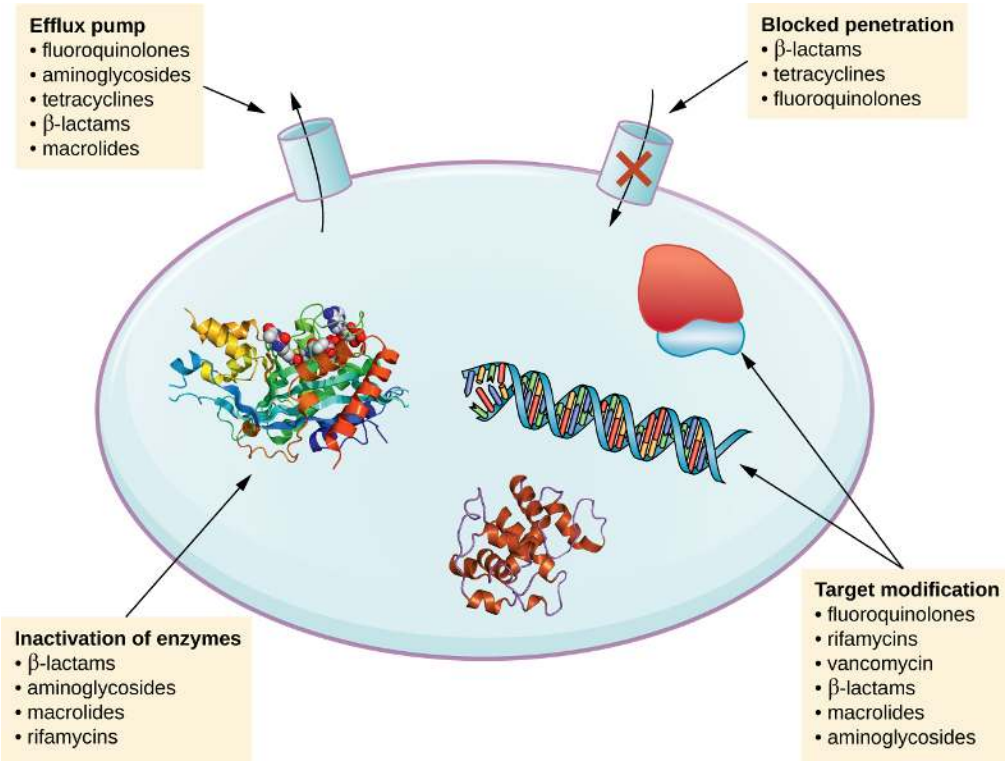


Figure **15.6.1**: There are multiple strategies that microbes use to develop resistance to antimicrobial drugs. (Not shown: target overproduction, target mimicry, and enzymatic bypass). (credit: modification of work by Gerard D Wright)

Drug Modification or Inactivation

Resistance genes may code for enzymes that chemically modify an antimicrobial, thereby inactivating it, or destroy an antimicrobial through hydrolysis. Resistance to many types of antimicrobials occurs through this mechanism. For example, aminoglycoside resistance can occur through enzymatic transfer of chemical groups to the drug molecule, impairing the binding of the drug to its bacterial target. For β-lactams, bacterial resistance can involve the enzymatic hydrolysis of the β-lactam bond within the β-lactam ring of the drug molecule. Once the β-lactam bond is broken, the drug loses its antibacterial activity. This mechanism of resistance is mediated by **β-lactamases**, which are the most common mechanism of β-lactam resistance. Inactivation of rifampin commonly occurs through glycosylation, phosphorylation, or adenosine diphosphate (ADP) ribosylation, and resistance to macrolides and lincosamides can also occur due to enzymatic inactivation of the drug or modification.

Prevention of Cellular Uptake or Efflux

Microbes may develop resistance mechanisms that involve inhibiting the accumulation of an antimicrobial drug, which then prevents the drug from reaching its cellular target. This strategy is common among gram-negative pathogens and can involve changes in outer membrane lipid composition, porin channel selectivity, and/or porin channel concentrations. For example, a common mechanism of carbapenem resistance among *Pseudomonas aeruginosa* is to decrease the amount of its OprD porin, which is the primary portal of entry for carbapenems through the outer membrane of this pathogen. Additionally, many gram-positive and gram-negative pathogenic bacteria produce **efflux pumps** that actively transport an antimicrobial drug out of the cell and prevent the accumulation of drug to a level that would be antibacterial. For example, resistance to β-lactams, tetracyclines, and fluoroquinolones commonly occurs through active efflux out of the cell, and it is rather common for a single efflux pump to have the ability to translocate multiple types of antimicrobials.

Target Modification

Because antimicrobial drugs have very specific targets, structural changes to those targets can prevent drug binding, rendering the drug ineffective. Through spontaneous mutations in the genes encoding antibacterial drug targets, bacteria have an evolutionary advantage that allows them to develop resistance to drugs. This mechanism of resistance development is quite common. Genetic changes impacting the active site of penicillin-binding proteins (PBPs) can inhibit the binding of β-lactam drugs and provide

resistance to multiple drugs within this class. This mechanism is very common among strains of *Streptococcus pneumoniae*, which alter their own PBPs through genetic mechanisms. In contrast, strains of *Staphylococcus aureus* develop resistance to methicillin (MRSA) through the acquisition of a new low-affinity PBP, rather than structurally alter their existing PBPs. Not only does this new low-affinity PBP provide resistance to methicillin but it provides resistance to virtually all β-lactam drugs, with the exception of the newer fifth-generation cephalosporins designed specifically to kill MRSA. Other examples of this resistance strategy include alterations in

- ribosome subunits, providing resistance to macrolides, tetracyclines, and aminoglycosides;
- lipopolysaccharide (LPS) structure, providing resistance to polymyxins;
- RNA polymerase, providing resistance to rifampin;
- DNA gyrase, providing resistance to fluoroquinolones;
- metabolic enzymes, providing resistance to sulfa drugs, sulfones, and trimethoprim; and
- peptidoglycan subunit peptide chains, providing resistance to glycopeptides.

Target Overproduction or Enzymatic Bypass

When an antimicrobial drug functions as an antimetabolite, targeting a specific enzyme to inhibit its activity, there are additional ways that microbial resistance may occur. First, the microbe may overproduce the target enzyme such that there is a sufficient amount of antimicrobial-free enzyme to carry out the proper enzymatic reaction. Second, the bacterial cell may develop a bypass that circumvents the need for the functional target enzyme. Both of these strategies have been found as mechanisms of sulfonamide resistance. Vancomycin resistance among *S. aureus* has been shown to involve the decreased cross-linkage of peptide chains in the bacterial cell wall, which provides an increase in targets for vancomycin to bind to in the outer cell wall. Increased binding of vancomycin in the outer cell wall provides a blockage that prevents free drug molecules from penetrating to where they can block new cell wall synthesis.

Target Mimicry

A recently discovered mechanism of resistance called target mimicry involves the production of proteins that bind and sequester drugs, preventing the drugs from binding to their target. For example, *Mycobacterium tuberculosis*produces a protein with regular pentapeptide repeats that appears to mimic the structure of DNA. This protein binds fluoroquinolones, sequestering them and keeping them from binding to DNA, providing *M. tuberculosis* resistance to fluoroquinolones. Proteins that mimic the A-site of the bacterial ribosome have been found to contribute to aminoglycoside resistance as well.[1]

Key Concepts and Summary

- **Antimicrobial resistance** is on the rise and is the result of selection of drug-resistant strains in clinical environments, the overuse and misuse of antibacterials, the use of subtherapeutic doses of antibacterial drugs, and poor patient compliance with antibacterial drug therapies.
- Drug resistance genes are often carried on plasmids or in transposons that can undergo vertical transfer easily and between microbes through horizontal gene transfer.
- Common modes of antimicrobial drug resistance include drug modification or inactivation, prevention of cellular uptake or efflux, target modification, target overproduction or enzymatic bypass, and target mimicry.

Footnotes

1. 1 D.H. Fong, A.M. Berghuis. “Substrate Promiscuity of an Aminoglycoside Antibiotic Resistance Enzyme Via Target Mimicry.” *EMBO Journal* 21 no. 10 (2002):2323–2331.
2. 2 Centers for Disease Control and Prevention. “Antibiotic/Antimicrobial Resistance.” http://www.cdc.gov/drugresistance/index.html. Accessed June 2, 2016.

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15.E: Antimicrobial Drugs (Exercises)

14.1: Discovering Antimicrobial Drugs

Antimicrobial drugs produced by purposeful fermentation and/or contained in plants have been used as traditional medicines in many cultures for millennia. The purposeful and systematic search for a chemical “magic bullet” that specifically target infectious microbes was initiated by Paul Ehrlich in the early 20th century. The discovery of the natural antibiotic, penicillin, by Alexander Fleming in 1928 started the modern age of antimicrobial discovery and research.

Multiple Choice

A scientist discovers that a soil bacterium he has been studying produces an antimicrobial that kills gram-negative bacteria. She isolates and purifies the antimicrobial compound, then chemically converts a chemical side chain to a hydroxyl group. When she tests the antimicrobial properties of this new version, she finds that this antimicrobial drug can now also kill gram-positive bacteria. The new antimicrobial drug with broad-spectrum activity is considered to be which of the following?

A. resistant
B. semisynthetic
C. synthetic
D. natural

Answer

B

Which of the following antimicrobial drugs is synthetic?

A. sulfanilamide
B. penicillin
C. actinomycin
D. neomycin

Answer

A

Fill in the Blank

The group of soil bacteria known for their ability to produce a wide variety of antimicrobials is called the _____.

Answer

actinomycetes

Short Answer

Where do antimicrobials come from naturally? Why?

Why was Salvarsan considered to be a “magic bullet” for the treatment of syphilis?

Critical Thinking

In nature, why do antimicrobial-producing microbes commonly also have antimicrobial resistance genes?

14.2: Antibacterial Drugs

Antimicrobial drugs can be bacteriostatic or bactericidal, and these characteristics are important considerations when selecting the most appropriate drug. The use of narrow-spectrum antimicrobial drugs is preferred in many cases to avoid superinfection and the development of antimicrobial resistance. Broad-spectrum antimicrobial use is warranted for serious systemic infections when there is no time to determine the causative agent or when narrow-spectrum antimicrobials fail.

Multiple Choice

Which of the following combinations would most likely contribute to the development of a superinfection?

A. long-term use of narrow-spectrum antimicrobials
B. long-term use of broad-spectrum antimicrobials
C. short-term use of narrow-spectrum antimicrobials
D. short-term use of broad-spectrum antimicrobials

Answer

B

Which of the following routes of administration would be appropriate and convenient for home administration of an antimicrobial to treat a systemic infection?

A. oral
B. intravenous
C. topical
D. parenteral

Answer

A

Which clinical situation would be appropriate for treatment with a narrow-spectrum antimicrobial drug?

A. treatment of a polymicrobic mixed infection in the intestine
B. prophylaxis against infection after a surgical procedure
C. treatment of strep throat caused by culture identified *Streptococcus pyogenes*
D. empiric therapy of pneumonia while waiting for culture results

Answer

C

Fill in the Blank

The bacterium known for causing pseudomembranous colitis, a potentially deadly superinfection, is _____.

Answer

Clostridium difficile

True/False

Narrow-spectrum antimicrobials are commonly used for prophylaxis following surgery.

Answer

False

Short Answer

When prescribing antibiotics, what aspects of the patient's health history should the clinician ask about and why?

When is using a broad-spectrum antimicrobial drug warranted?

Critical Thinking

Why are yeast infections a common type of superinfection that results from long-term use of broad-spectrum antimicrobials?

Too often patients will stop taking antimicrobial drugs before the prescription is finished. What are factors that cause a patient to stop too soon, and what negative impacts could this have?

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CHAPTER OVERVIEW

16: Antibiotic Resistance and Other Antimicrobials

In nature, some microbes produce substances that inhibit or kill other microbes that might otherwise compete for the same resources. Humans have successfully exploited these abilities, using microbes to mass-produce substances that can be used as antimicrobial drugs. Since their discovery, antimicrobial drugs have saved countless lives, and they remain an essential tool for treating and controlling infectious disease. But their widespread and often unnecessary use has had an unintended side effect: the rise of multidrug-resistant microbial strains. In this chapter, we will discuss how antimicrobial drugs work, why microbes develop resistance, and what health professionals can do to encourage responsible use of antimicrobials.



Figure **16.1**: First mass produced in the 1940s, penicillin was instrumental in saving millions of lives during World War II and was considered a wonder drug.[1] Today, overprescription of antibiotics (especially for childhood illnesses) has contributed to the evolution of drug-resistant pathogens. (credit left: modification of work by Chemical Heritage Foundation; Credit right: DFID / Flickr; CC-BY)

16.1: The Emergence of Drug Resistance
16.2: Current Strategies for Antimicrobial Discovery
16.E: Antimicrobial Drugs (Exercises)

Footnotes

1. 1 "Treatment of War Wounds: A Historical Review." *Clinical Orthopaedics and Related Research* 467 no. 8 (2009):2168–2191.

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16.1: The Emergence of Drug Resistance

16.02: The Emergence of Drug Resistance

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16.2: Current Strategies for Antimicrobial Discovery

Learning Objectives

- Describe the methods and strategies used for discovery of new antimicrobial agents.

With the continued evolution and spread of antimicrobial resistance, and now the identification of pan-resistant bacterial pathogens, the search for new antimicrobials is essential for preventing the postantibiotic era. Although development of more effective semisynthetic derivatives is one strategy, resistance to them develops rapidly because bacterial pathogens are already resistant to earlier-generation drugs in the family and can easily mutate and develop resistance to the new semisynthetic drugs. Today, scientists continue to hunt for new antimicrobial compounds and explore new avenues of antimicrobial discovery and synthesis. They check large numbers of soils and microbial products for antimicrobial activity by using high-throughput screening methods, which use automation to test large numbers of samples simultaneously. The recent development of the iChip[1] allows researchers to investigate the antimicrobial-producing capabilities of soil microbes that are difficult to grow by standard cultivation techniques in the laboratory. Rather than grow the microbes in the laboratory, they are grown in situ—right in the soil. Use of the iChip has resulted in the discovery of teixobactin, a novel antimicrobial from Mount Ararat, Turkey. Teixobactin targets two distinct steps in gram-positive cell wall synthesis and for which antimicrobial resistance appears not yet to have evolved.

Although soils have been widely examined, other environmental niches have not been tested as fully. Since 70% of the earth is covered with water, marine environments could be mined more fully for the presence of antimicrobial-producing microbes. In addition, researchers are using combinatorial chemistry, a method for making a very large number of related compounds from simple precursors, and testing them for antimicrobial activity. An additional strategy that needs to be explored further is the development of compounds that inhibit resistance mechanisms and restore the activity of older drugs, such as the strategy described earlier for β-lactamase inhibitors like clavulanic acid. Finally, developing inhibitors of virulence factor production and function could be a very important avenue. Although this strategy would not be directly antibacterial, drugs that slow the progression of an infection could provide an advantage for the immune system and could be used successfully in combination with antimicrobial drugs.

The (Free?) Market for New Antimicrobials

There used to be plenty of antimicrobial drugs on the market to treat infectious diseases. However, the spread of antimicrobial resistance has created a need for new antibiotics to replace those that are no longer as effective as they once were. Unfortunately, pharmaceutical companies are not particularly motivated to fill this need. As of 2009, all but five pharmaceutical companies had moved away from antimicrobial drug development.[2] As a result, the number of FDA approvals of new antimicrobials has fallen drastically in recent decades (Figure **16.2.1**).

Given that demand usually encourages supply, one might expect pharmaceutical companies to be rushing to get back in the business of developing new antibiotics. But developing new drugs is a lengthy process and requires large investments in research and development. Pharmaceutical companies can typically get a higher return on their investment by developing products for chronic, nonmicrobial diseases like diabetes; such drugs must be taken for life, and therefore generate more long-term revenue than an antibiotic that does its job in a week or two. But what will happen when drugs like vancomycin, a superantimicrobial reserved for use as a last resort, begin to lose their effectiveness against ever more drug-resistant superbugs? Will drug companies wait until all antibiotics have become useless before beginning to look for new ones?

Recently, it has been suggested that large pharmaceutical companies should be given financial incentives to pursue such research. In September 2014, the White House released an executive order entitled “Combating Antibiotic Resistant Bacteria,” calling upon various government agencies and the private sector to work together to “accelerate basic and applied research and development for new antimicrobials, other therapeutics, and vaccines.”[3] As a result, as of March 2015, President Obama’s proposed fiscal year 2016 budget doubled the amount of federal funding to $1.2 billion for “combating and preventing antibiotic resistance,” which includes money for antimicrobial research and development.[4] Similar suggestions have also been

made on a global scale. In December 2014, a report chaired by former Goldman Sachs economist Jim O'Neill was published in *The Review on Antimicrobial Resistance*.[5]

These developments reflect the growing belief that for-profit pharmaceutical companies must be subsidized to encourage development of new antimicrobials. But some ask whether pharmaceutical development should be motivated by profit at all. Given that millions of lives may hang in the balance, some might argue that drug companies have an ethical obligation to devote their research and development efforts to high-utility drugs, as opposed to highly profitable ones. Yet this obligation conflicts with the fundamental goals of a for-profit company. Are government subsidies enough to ensure that drug companies make the public interest a priority, or should government agencies assume responsibility for developing critical drugs that may have little or no return on investment?

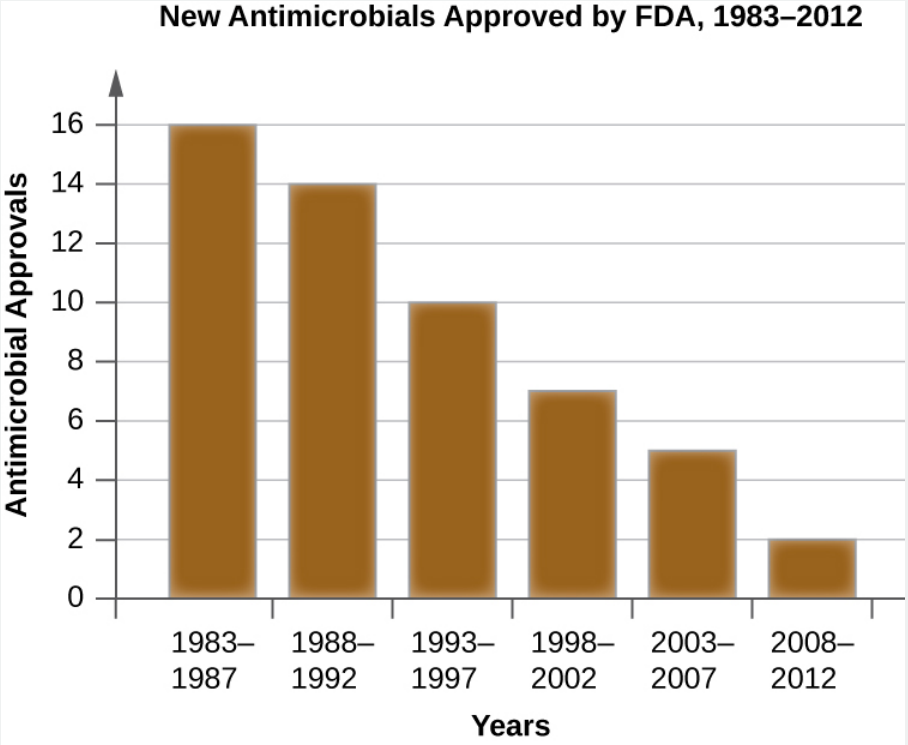


Figure **16.2.1**: In recent decades, approvals of new antimicrobials by the FDA have steadily fallen. In the five-year period from 1983–1987, 16 new antimicrobial drugs were approved, compared to just two from 2008–2012.

Link to Learning

To further examine the scope of the problem, view this video.

To learn more about the history of antimicrobial drug discovery, visit Michigan State University's Antimicrobial Resistance Learning Site.

Key Concepts and Summary

- Current research into the development of antimicrobial drugs involves the use of high-throughput screening and combinatorial chemistry technologies.
- New technologies are being developed to discover novel antibiotics from soil microorganisms that cannot be cultured by standard laboratory methods.
- Additional strategies include searching for antibiotics from sources other than soil, identifying new antibacterial targets, using combinatorial chemistry to develop novel drugs, developing drugs that inhibit resistance mechanisms, and developing drugs that target virulence factors and hold infections in check.

Footnotes

1. 1 L. Losee et al. “A New Antibiotic Kills Pathogens Without Detectable Resistance.” *Nature* 517 no. 7535 (2015):455–459.
2. 2 H.W. Boucher et al. “Bad Bugs, No Drugs: No ESKAPE! An Update from the Infectious Diseases Society of America.” *Clinical Infectious Diseases* 48 no. 1 (2009):1–12.
3. 3 The White House. *National Action Plan for Combating Antibiotic-Resistant Bacteria.* Washington, DC: The White House, 2015.
4. 4 White House Office of the Press Secretary. “Fact Sheet: Obama Administration Releases National Action Plan to Combat Antibiotic-Resistant Bacteria.” March 27, 2015. https://www.whitehouse.gov/the-press...lan-combat-ant
5. 5 Review on Antimicrobial Resistance. http://amr-review.org. Accessed June 1, 2016.

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16.E: Antimicrobial Drugs (Exercises)

15.1: Drugs Targeting Other Microorganisms

Antibacterial compounds exhibit selective toxicity, largely due to differences between prokaryotic and eukaryotic cell structure. Cell wall synthesis inhibitors, including the β-lactams, the glycopeptides, and bacitracin, interfere with peptidoglycan synthesis, making bacterial cells more prone to osmotic lysis. There are a variety of broad-spectrum, bacterial protein synthesis inhibitors that selectively target the prokaryotic 70S ribosome, including those that bind to the 30S and 50S subunits.

Multiple Choice

Which of the following terms refers to the ability of an antimicrobial drug to harm the target microbe without harming the host?

A. mode of action
B. therapeutic level
C. spectrum of activity
D. selective toxicity

Answer

D

Which of the following is not a type of β-lactam antimicrobial?

A. penicillins
B. glycopeptides
C. cephalosporins
D. monobactams

Answer

B

Which of the following does not bind to the 50S ribosomal subunit?

A. tetracyclines
B. lincosamides
C. macrolides
D. chloramphenicol

Answer

A

Which of the following antimicrobials inhibits the activity of DNA gyrase?

A. polymyxin B
B. clindamycin
C. nalidixic acid
D. rifampin

Answer

C

Fill in the Blank

Selective toxicity antimicrobials are easier to develop against bacteria because they are _____ cells, whereas human cells are eukaryotic.

Answer

prokaryotic

True/False

β-lactamases can degrade vancomycin.

Answer

false

Short Answer

If human cells and bacterial cells perform transcription, how are the rifamycins specific for bacterial infections?

What bacterial structural target would make an antibacterial drug selective for gram-negative bacteria? Provide one example of an antimicrobial compound that targets this structure.

Critical Thinking

In considering the cell structure of prokaryotes compared with that of eukaryotes, propose one possible reason for side effects in humans due to treatment of bacterial infections with protein synthesis inhibitors.

15.2: Clinical Considerations

Because fungi, protozoans, and helminths are eukaryotic organisms like human cells, it is more challenging to develop antimicrobial drugs that specifically target them. Similarly, it is hard to target viruses because human viruses replicate inside of human cells.

Multiple Choice

Which of the following is not an appropriate target for antifungal drugs?

A. ergosterol
B. chitin
C. cholesterol
D. β(1→3) glucan

Answer

C

Which of the following drug classes specifically inhibits neuronal transmission in helminths?

A. quinolines
B. avermectins
C. amantadines
D. imidazoles

Answer

B

Which of the following is a nucleoside analog commonly used as a reverse transcriptase inhibitor in the treatment of HIV?

A. acyclovir
B. ribavirin
C. adenine-arabinoside
D. azidothymidine

Answer

D

Which of the following is an antimalarial drug that is thought to increase ROS levels in target cells?

A. artemisinin
B. amphotericin b
C. praziquantel
D. pleconaril

Answer

A

Fill in the Blank

Antiviral drugs, like Tamiflu and Relenza, that are effective against the influenza virus by preventing viral escape from host cells are called _____.

Answer

neuraminidase inhibitors

True/False

Echinocandins, known as “penicillin for fungi,” target $\beta(1 \rightarrow 3)$ glucan in fungal cell walls.

Answer

true

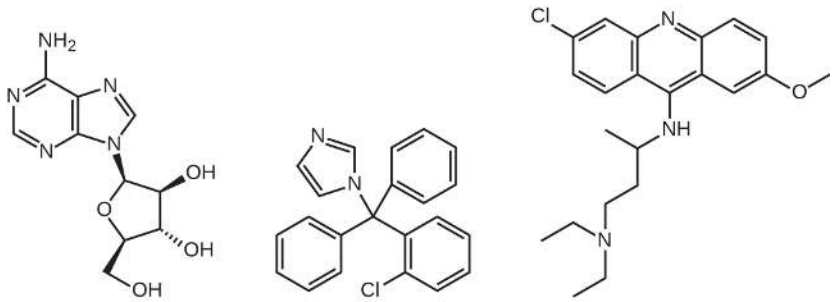
Short Answer

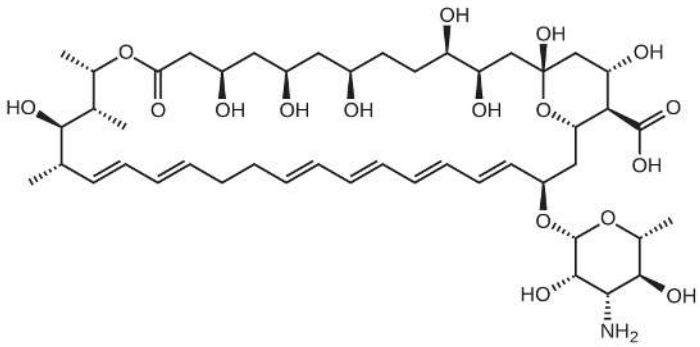
How does the biology of HIV necessitate the need to treat HIV infections with multiple drugs?

Niclosamide is insoluble and thus is not readily absorbed from the stomach into the bloodstream. How does the insolubility of niclosamide aid its effectiveness as a treatment for tapeworm infection?

Critical Thinking

Which of the following molecules is an example of a nucleoside analog?





Why can’t drugs used to treat influenza, like amantadines and neuraminidase inhibitors, be used to treat a wider variety of viral infections?

15.3: Testing the Effectiveness of Antimicrobials

Antimicrobial resistance is on the rise and is the result of selection of drug-resistant strains in clinical environments, the overuse and misuse of antibacterials, the use of subtherapeutic doses of antibacterial drugs, and poor patient compliance with antibacterial drug therapies. Drug resistance genes are often carried on plasmids or in transposons that can undergo vertical transfer easily and between microbes through horizontal gene transfer.

Multiple Choice

Which of the following resistance mechanisms describes the function of β-lactamase?

A. efflux pump
B. target mimicry
C. drug inactivation
D. target overproduction

Answer

C

Which of the following resistance mechanisms is commonly effective against a wide range of antimicrobials in multiple classes?

A. efflux pump
B. target mimicry
C. target modification
D. target overproduction

Answer

A

Which of the following resistance mechanisms is the most nonspecific to a particular class of antimicrobials?

A. drug modification
B. target mimicry
C. target modification
D. efflux pump

Answer

D

Which of the following types of drug-resistant bacteria do not typically persist in individuals as a member of their intestinal microbiota?

A. MRSA
B. VRE
C. CRE
D. ESBL-producing bacteria

Answer

A

Fill in the Blank

Staphylococcus aureus, including MRSA strains, may commonly be carried as a normal member of the _____ microbiota in some people.

Answer

nasal

Short Answer

Why does the length of time of antimicrobial treatment for tuberculosis contribute to the rise of resistant strains?

What is the difference between multidrug resistance and cross-resistance?

15.4: The Emergence of Drug Resistance

The Kirby-Bauer disk diffusion test helps determine the susceptibility of a microorganism to various antimicrobial drugs. However, the zones of inhibition measured must be correlated to known standards to determine susceptibility and resistance, and do not provide information on bactericidal versus bacteriostatic activity, or allow for direct comparison of drug potencies. Antibiograms are useful for monitoring local trends in antimicrobial resistance/susceptibility.

Multiple Choice

In the Kirby-Bauer disk diffusion test, the _____ of the zone of inhibition is measured and used for interpretation.

A. diameter
B. microbial population
C. circumference
D. depth

Answer

A

Which of the following techniques cannot be used to determine the minimum inhibitory concentration of an antimicrobial drug against a particular microbe?

A. Etest

B. microbroth dilution test
C. Kirby-Bauer disk diffusion test
D. macrobroth dilution test

Answer

C

The utility of an antibiogram is that it shows antimicrobial susceptibility trends

A. over a large geographic area.
B. for an individual patient.
C. in research laboratory strains.
D. in a localized population.

Answer

D

Fill in the Blank

The method that can determine the MICs of multiple antimicrobial drugs against a microbial strain using a single agar plate is called the _____.

Answer

Etest

True/False

If drug A produces a larger zone of inhibition than drug B on the Kirby-Bauer disk diffusion test, drug A should always be prescribed.

Answer

false

Short Answer

How is the information from a Kirby-Bauer disk diffusion test used for the recommendation of the clinical use of an antimicrobial drug?

What is the difference between MIC and MBC?

Critical Thinking

Can an Etest be used to find the MBC of a drug? Explain.

15.5: Current Strategies for Antimicrobial Discovery

With the continued evolution and spread of antimicrobial resistance, and now the identification of pan-resistant bacterial pathogens, the search for new antimicrobials is essential for preventing the postantibiotic era.

Multiple Choice

Which of the following has yielded compounds with the most antimicrobial activity?

A. water
B. air
C. volcanoes
D. soil

Answer

D

True/False

The rate of discovery of antimicrobial drugs has decreased significantly in recent decades.

Answer

true

Critical Thinking

Who should be responsible for discovering and developing new antibiotics? Support your answer with reasoning.

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CHAPTER OVERVIEW

17: Microbial Mechanisms of Pathogenicity

Jane woke up one spring morning feeling not quite herself. Her throat felt a bit dry and she was sniffling. She wondered why she felt so lousy. Was it because of a change in the weather? The pollen count? Was she coming down with something? Did she catch a bug from her coworker who sneezed on her in the elevator yesterday?

The signs and symptoms we associate with illness can have many different causes. Sometimes they are the direct result of a pathogenic infection, but in other cases they result from a response by our immune system to a pathogen or another perceived threat. For example, in response to certain pathogens, the immune system may release pyrogens, chemicals that cause the body temperature to rise, resulting in a fever. This response creates a less-than-favorable environment for the pathogen, but it also makes us feel sick.

Medical professionals rely heavily on analysis of signs and symptoms to determine the cause of an ailment and prescribe treatment. In some cases, signs and symptoms alone are enough to correctly identify the causative agent of a disease, but since few diseases produce truly unique symptoms, it is often necessary to confirm the identity of the infectious agent by other direct and indirect diagnostic methods.



Figure **17.1**: Although medical professionals rely heavily on signs and symptoms to diagnose disease and prescribe treatment, many diseases can produce similar signs and symptoms. (credit left: modification of work by U.S. Navy)

17.1: Characteristics of Infectious Diseases

17.2: How Pathogens Cause Disease

17.3: Virulence Factors

17.4: Aseptic Techniques

17.E: Microbial Mechanisms of Pathogenicity (Exercises)

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17.1: Characteristics of Infectious Diseases

Learning Objectives

- Distinguish between signs and symptoms of disease
- Explain the difference between a communicable disease and a noncommunicable disease
- Compare different types of infectious diseases, including iatrogenic, nosocomial, and zoonotic diseases

A **disease** is any condition in which the normal structure or functions of the body are damaged or impaired. Physical injuries or disabilities are not classified as disease, but there can be several causes for disease, including infection by a pathogen, genetics (as in many cancers or deficiencies), noninfectious environmental causes, or inappropriate immune responses. Our focus in this chapter will be on infectious diseases, although when diagnosing infectious diseases, it is always important to consider possible noninfectious causes.



Signs and Symptoms of Disease

An **infection** is the successful colonization of a host by a microorganism. Infections can lead to disease, which causes signs and symptoms resulting in a deviation from the normal structure or functioning of the host. Microorganisms that can cause disease are known as pathogens.

The **signs** of disease are objective and measurable, and can be directly observed by a clinician. Vital signs, which are used to measure the body's basic functions, include body temperature (normally 37°C [98.6°F]), heart rate (normally 60–100 beats per minute), breathing rate (normally 12–18 breaths per minute), and blood pressure (normally between 90/60 and 120/80 mm Hg). Changes in any of the body's vital signs may be indicative of disease. For example, having a fever (a body temperature significantly higher than 37°C or 98.6°F) is a sign of disease because it can be measured.

In addition to changes in vital signs, other observable conditions may be considered signs of disease. For example, the presence of antibodies in a patient's serum (the liquid portion of blood that lacks clotting factors) can be observed and measured through blood tests and, therefore, can be considered a sign. However, it is important to note that the presence of antibodies is not always a sign of an active disease. Antibodies can remain in the body long after an infection has resolved; also, they may develop in response to a pathogen that is in the body but not currently causing disease.

Unlike signs, **symptoms** of disease are subjective. Symptoms are felt or experienced by the patient, but they cannot be clinically confirmed or objectively measured. Examples of symptoms include nausea, loss of appetite, and pain. Such symptoms are important to consider when diagnosing disease, but they are subject to memory bias and are difficult to measure precisely. Some clinicians attempt to quantify symptoms by asking patients to assign a numerical value to their symptoms. For example, the Wong-Baker Faces pain-rating scale asks patients to rate their pain on a scale of 0–10. An alternative method of quantifying pain is

measuring skin conductance fluctuations. These fluctuations reflect sweating due to skin sympathetic nerve activity resulting from the stressor of pain.[1]

A specific group of signs and symptoms characteristic of a particular disease is called a **syndrome**. Many syndromes are named using a nomenclature based on signs and symptoms or the location of the disease. Table **17.1.1** lists some of the prefixes and suffixes commonly used in naming syndromes.

Table **17.1.1**: Nomenclature of Symptoms

Affix	Meaning	Example
cyto-	cell	cytopenia: reduction in the number of blood cells
hepat-	of the liver	hepatitis: inflammation of the liver
-pathy	disease	neuropathy: a disease affecting nerves
-emia	of the blood	bacteremia: presence of bacteria in blood
-itis	inflammation	colitis: inflammation of the colon
-lysis	destruction	hemolysis: destruction of red blood cells
-oma	tumor	lymphoma: cancer of the lymphatic system
-osis	diseased or abnormal condition	leukocytosis: abnormally high number of white blood cells
-derma	of the skin	keratoderma: a thickening of the skin

Clinicians must rely on signs and on asking questions about symptoms, medical history, and the patient's recent activities to identify a particular disease and the potential causative agent. Diagnosis is complicated by the fact that different microorganisms can cause similar signs and symptoms in a patient. For example, an individual presenting with symptoms of diarrhea may have been infected by one of a wide variety of pathogenic microorganisms. Bacterial pathogens associated with diarrheal disease include *Vibrio cholerae*, *Listeria monocytogenes*, *Campylobacter jejuni*, and enteropathogenic *Escherichia coli* (EPEC). Viral pathogens associated with diarrheal disease include norovirus and rotavirus. Parasitic pathogens associated with diarrhea include *Giardia lamblia* and *Cryptosporidium parvum*. Likewise, fever is indicative of many types of infection, from the common cold to the deadly Ebola hemorrhagic fever.

Finally, some diseases may be asymptomatic or subclinical, meaning they do not present any noticeable signs or symptoms. For example, most individual infected with herpes simplex virus remain asymptomatic and are unaware that they have been infected.

Classifications of Disease

The World Health Organization's (WHO) International Classification of Diseases (ICD) is used in clinical fields to classify diseases and monitor morbidity (the number of cases of a disease) and mortality (the number of deaths due to a disease). In this section, we will introduce terminology used by the ICD (and in health-care professions in general) to describe and categorize various types of disease.

An **infectious disease** is any disease caused by the direct effect of a pathogen. A pathogen may be cellular (bacteria, parasites, and fungi) or acellular (viruses, viroids, and prions). Some infectious diseases are also communicable, meaning they are capable of being spread from person to person through either direct or indirect mechanisms. Some infectious communicable diseases are also considered **contagious** diseases, meaning they are easily spread from person to person. Not all contagious diseases are equally so; the degree to which a disease is contagious usually depends on how the pathogen is transmitted. For example, measles is a highly contagious viral disease that can be transmitted when an infected person coughs or sneezes and an uninfected person breathes in droplets containing the virus. Gonorrhea is not as contagious as measles because transmission of the pathogen (*Neisseria gonorrhoeae*) requires close intimate contact (usually sexual) between an infected person and an uninfected person.

Diseases that are contracted as the result of a medical procedure are known as **iatrogenic diseases**. Iatrogenic diseases can occur after procedures involving wound treatments, catheterization, or surgery if the wound or surgical site becomes contaminated. For example, an individual treated for a skin wound might acquire necrotizing fasciitis (an aggressive, "flesh-eating" disease) if bandages or other dressings became contaminated by *Clostridium perfringens* or one of several other bacteria that can cause this condition.

Diseases acquired in hospital settings are known as **nosocomial diseases**. Several factors contribute to the prevalence and severity of nosocomial diseases. First, sick patients bring numerous pathogens into hospitals, and some of these pathogens can be transmitted easily via improperly sterilized medical equipment, bed sheets, call buttons, door handles, or by clinicians, nurses, or therapists who do not wash their hands before touching a patient. Second, many hospital patients have weakened immune systems, making them more susceptible to infections. Compounding this, the prevalence of antibiotics in hospital settings can select for drug-resistant bacteria that can cause very serious infections that are difficult to treat.

Certain infectious diseases are not transmitted between humans directly but can be transmitted from animals to humans. Such a disease is called **zoonotic disease** (or **zoonosis**). According to WHO, a zoonosis is a disease that occurs when a pathogen is transferred from a vertebrate animal to a human; however, sometimes the term is defined more broadly to include diseases transmitted by all animals (including invertebrates). For example, rabies is a viral zoonotic disease spread from animals to humans through bites and contact with infected saliva. Many other zoonotic diseases rely on insects or other arthropods for transmission. Examples include yellow fever (transmitted through the bite of mosquitoes infected with yellow fever virus) and Rocky Mountain spotted fever (transmitted through the bite of ticks infected with *Rickettsia rickettsii*).

In contrast to communicable infectious diseases, a **noncommunicable** infectious disease is not spread from one person to another. One example is tetanus, caused by *Clostridium tetani*, a bacterium that produces endospores that can survive in the soil for many years. This disease is typically only transmitted through contact with a skin wound; it cannot be passed from an infected person to another person. Similarly, Legionnaires disease is caused by *Legionella pneumophila*, a bacterium that lives within amoebae in moist locations like water-cooling towers. An individual may contract Legionnaires disease via contact with the contaminated water, but once infected, the individual cannot pass the pathogen to other individuals.

In addition to the wide variety of noncommunicable infectious diseases, noninfectious diseases (those not caused by pathogens) are an important cause of morbidity and mortality worldwide. Noninfectious diseases can be caused by a wide variety factors, including genetics, the environment, or immune system dysfunction, to name a few. For example, sickle cell anemia is an inherited disease caused by a genetic mutation that can be passed from parent to offspring (Figure **17.1.1**). Other types of noninfectious diseases are listed in Table **17.1.2**.

Table **17.1.2**: Types of Noninfectious Diseases

Type	Definition	Example
Inherited	A genetic disease	Sickle cell anemia
Congenital	Disease that is present at or before birth	Down syndrome
Degenerative	Progressive, irreversible loss of function	Parkinson disease (affecting central nervous system)
Nutritional deficiency	Impaired body function due to lack of nutrients	Scurvy (vitamin C deficiency)
Endocrine	Disease involving malfunction of glands that release hormones to regulate body functions	Hypothyroidism – thyroid does not produce enough thyroid hormone, which is important for metabolism
Neoplastic	Abnormal growth (benign or malignant)	Some forms of cancer
Idiopathic	Disease for which the cause is unknown	Idiopathic juxtafoveal retinal telangiectasia (dilated, twisted blood vessels in the retina of the eye)

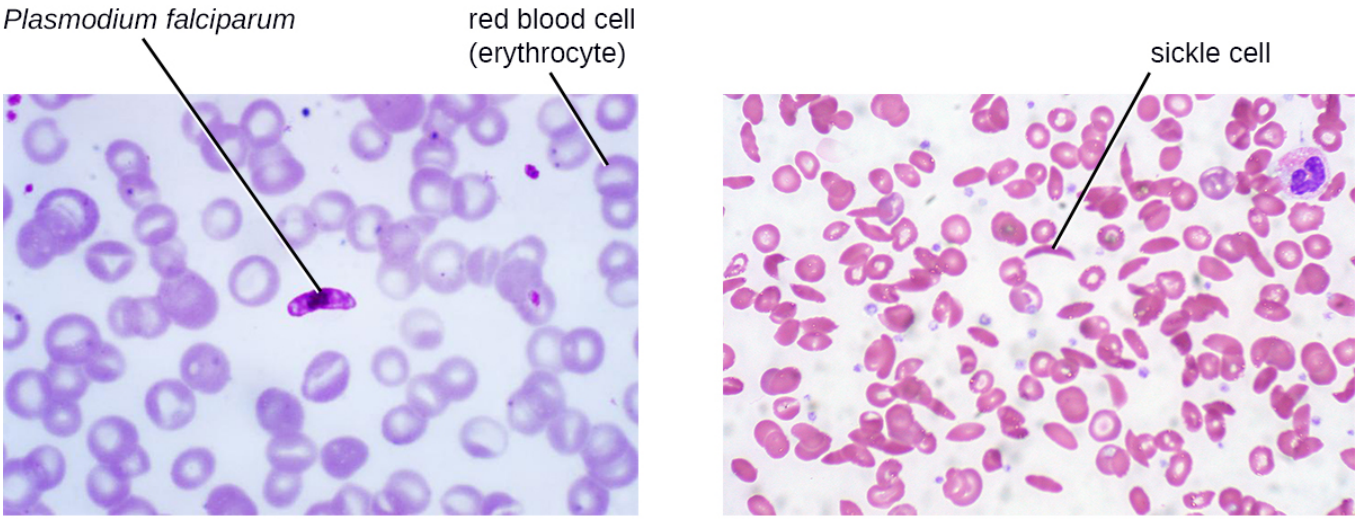


Figure **17.1.1**: Blood smears showing two diseases of the blood. (a) Malaria is an infectious, zoonotic disease caused by the protozoan pathogen *Plasmodium falciparum* (shown here) and several other species of the genus *Plasmodium*. It is transmitted by mosquitoes to humans. (b) Sickle cell disease is a noninfectious genetic disorder that results in abnormally shaped red blood cells, which can stick together and obstruct the flow of blood through the circulatory system. It is not caused by a pathogen, but rather a genetic mutation. (credit a: modification of work by Centers for Disease Control and Prevention; credit b: modification of work by Ed Uthman)

Key Concepts and Summary

- In an **infection**, a microorganism enters a host and begins to multiply. Some infections cause **disease**, which is any deviation from the normal function or structure of the host.
- **Signs** of a disease are objective and are measured. **Symptoms** of a disease are subjective and are reported by the patient.
- Diseases can either be **noninfectious** (due to genetics and environment) or **infectious** (due to pathogens). Some infectious diseases are **communicable** (transmissible between individuals) or **contagious** (easily transmissible between individuals); others are **noncommunicable**, but may be contracted via contact with environmental reservoirs or animals (**zoonoses**)
- **Nosocomial diseases** are contracted in hospital settings, whereas **iatrogenic disease** are the direct result of a medical procedure

Footnotes

1. 1 F. Savino et al. “Pain Assessment in Children Undergoing Venipuncture: The Wong–Baker Faces Scale Versus Skin Conductance Fluctuations.” *PeerJ* 1 (2013):e37; https://peerj.com/articles/37/
2. 2 J.G. Kusters et al. Pathogenesis of *Helicobacter pylori* Infection. *Clinical Microbiology Reviews* 19 no. 3 (2006):449–490.
3. 3 N.R. Salama et al. “Life in the Human Stomach: Persistence Strategies of the Bacterial Pathogen *Helicobacter pylori*.” *Nature Reviews Microbiology* 11 (2013):385–399.

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17.2: How Pathogens Cause Disease

Learning Objectives

- Explain the significance and limitations of Koch’s postulates
- Compare and contrast Koch's postulates and Molecular Koch's postulates
- Analyze case studies where Koch's postulates and Molecular Koch's postulates have been applied

For most infectious diseases, the ability to accurately identify the causative pathogen is a critical step in finding or prescribing effective treatments. Today’s physicians, patients, and researchers owe a sizable debt to the physician Robert Koch (1843–1910), who devised a systematic approach for confirming causative relationships between diseases and specific pathogens.

Koch’s Postulates

In 1884, Koch published four postulates that summarized his method for determining whether a particular microorganism was the cause of a particular disease. Each of Koch’s postulates represents a criterion that must be met before a disease can be positively linked with a pathogen. In order to determine whether the criteria are met, tests are performed on laboratory animals and cultures from healthy and diseased animals are compared (Figure **17.2.1**).

Koch’s Postulates

1. The suspected pathogen must be found in every case of disease and not be found in healthy individuals.
2. The suspected pathogen can be isolated and grown in pure culture.
3. A healthy test subject infected with the suspected pathogen must develop the same signs and symptoms of disease as seen in postulate
4. The pathogen must be re-isolated from the new host and must be identical to the pathogen from postulate 2.

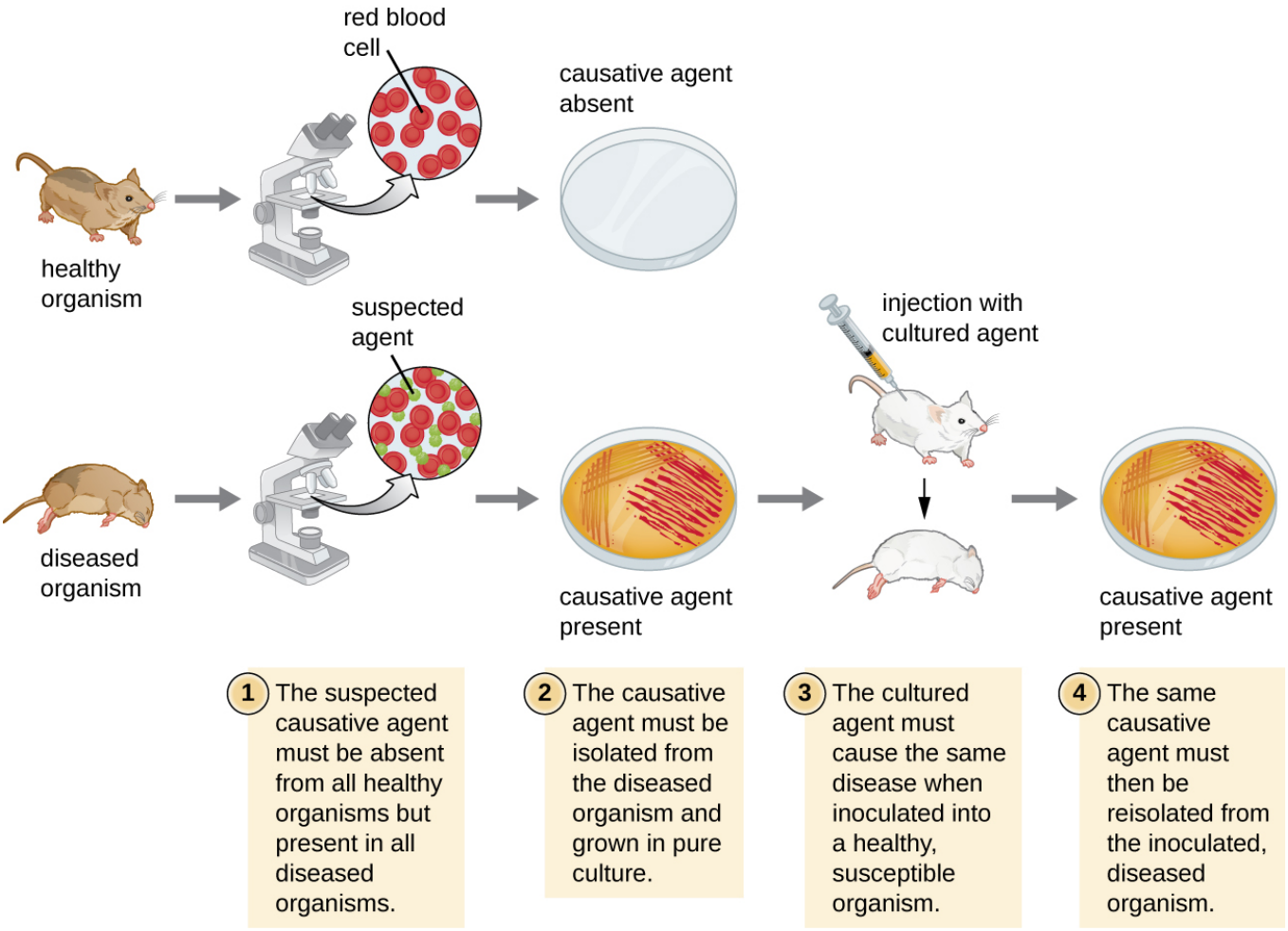


Figure **17.2.1**: The steps for confirming that a pathogen is the cause of a particular disease using Koch's postulates.

In many ways, Koch's postulates are still central to our current understanding of the causes of disease. However, advances in microbiology have revealed some important limitations in Koch's criteria. Koch made several assumptions that we now know are untrue in many cases. The first relates to postulate 1, which assumes that pathogens are only found in diseased, not healthy, individuals. This is not true for many pathogens. For example, *H. pylori*, described earlier in this chapter as a pathogen causing chronic gastritis, is also part of the normal microbiota of the stomach in many healthy humans who never develop gastritis. It is estimated that upwards of 50% of the human population acquires *H. pylori* early in life, with most maintaining it as part of the normal microbiota for the rest of their life without ever developing disease.

Koch's second faulty assumption was that all healthy test subjects are equally susceptible to disease. We now know that individuals are not equally susceptible to disease. Individuals are unique in terms of their microbiota and the state of their immune system at any given time. The makeup of the resident microbiota can influence an individual's susceptibility to an infection. Members of the normal microbiota play an important role in immunity by inhibiting the growth of transient pathogens. In some cases, the microbiota may prevent a pathogen from establishing an infection; in others, it may not prevent an infection altogether but may influence the severity or type of signs and symptoms. As a result, two individuals with the same disease may not always present with the same signs and symptoms. In addition, some individuals have stronger immune systems than others. Individuals with immune systems weakened by age or an unrelated illness are much more susceptible to certain infections than individuals with strong immune systems.

Koch also assumed that all pathogens are microorganisms that can be grown in pure culture (postulate 2) and that animals could serve as reliable models for human disease. However, we now know that not all pathogens can be grown in pure culture, and many human diseases cannot be reliably replicated in animal hosts. Viruses and certain bacteria, including *Rickettsia* and *Chlamydia*, are

obligate intracellular pathogens that can grow only when inside a host cell. If a microbe cannot be cultured, a researcher cannot move past postulate 2. Likewise, without a suitable nonhuman host, a researcher cannot evaluate postulate 2 without deliberately infecting humans, which presents obvious ethical concerns. AIDS is an example of such a disease because the human immunodeficiency virus (HIV) only causes disease in humans.

Molecular Koch's Postulates

In 1988, Stanley Falkow (1934–) proposed a revised form of Koch's postulates known as molecular Koch's postulates. These are listed in the left column of Table **17.2.1**. The premise for molecular Koch's postulates is not in the ability to isolate a particular pathogen but rather to identify a gene that may cause the organism to be pathogenic.

Falkow's modifications to Koch's original postulates explain not only infections caused by intracellular pathogens but also the existence of pathogenic strains of organisms that are usually nonpathogenic. For example, the predominant form of the bacterium *Escherichia coli* is a member of the normal microbiota of the human intestine and is generally considered harmless. However, there are pathogenic strains of *E. coli* such as enterotoxigenic *E. coli* (ETEC) and enterohemorrhagic *E. coli* (O157:H7) (EHEC). We now know ETEC and EHEC exist because of the acquisition of new genes by the once-harmless *E. coli*, which, in the form of these pathogenic strains, is now capable of producing toxins and causing illness. The pathogenic forms resulted from minor genetic changes. The right-side column of Table **17.2.1** illustrates how molecular Koch's postulates can be applied to identify EHEC as a pathogenic bacterium.

Table **17.2.1**: Molecular Koch's Postulates Applied to EHEC

Molecular Koch's Postulates	Application to EHEC
(1) The phenotype (sign or symptom of disease) should be associated only with pathogenic strains of a species.	EHEC causes intestinal inflammation and diarrhea, whereas nonpathogenic strains of *E. coli* do not.
(2) Inactivation of the suspected gene(s) associated with pathogenicity should result in a measurable loss of pathogenicity.	One of the genes in EHEC encodes for Shiga toxin, a bacterial toxin (poison) that inhibits protein synthesis. Inactivating this gene reduces the bacteria's ability to cause disease.
(3) Reversion of the inactive gene should restore the disease phenotype.	By adding the gene that encodes the toxin back into the genome (e.g., with a phage or plasmid), EHEC's ability to cause disease is restored.

As with Koch's original postulates, the molecular Koch's postulates have limitations. For example, genetic manipulation of some pathogens is not possible using current methods of molecular genetics. In a similar vein, some diseases do not have suitable animal models, which limits the utility of both the original and molecular postulates.

Key Concepts and Summary

- **Koch's postulates** are used to determine whether a particular microorganism is a pathogen. **Molecular Koch's postulates** are used to determine what genes contribute to a pathogen's ability to cause disease.

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17.3: Virulence Factors

17.03: Virulence Factors

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17.4: Aseptic Techniques

17.04: Aseptic Techniques

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17.E: Microbial Mechanisms of Pathogenicity (Exercises)

16.1: Characteristics of Infectious Diseases

In an infection, a microorganism enters a host and begins to multiply. Some infections cause disease, which is any deviation from the normal function or structure of the host. Signs of a disease are objective and are measured. Symptoms of a disease are subjective and are reported by the patient. Diseases can either be noninfectious (due to genetics and environment) or infectious (due to pathogens).

Multiple Choice

Which of the following would be a sign of an infection?

A. muscle aches
B. headache
C. fever
D. nausea

Answer

C

Which of the following is an example of a noncommunicable infectious disease?

A. infection with a respiratory virus
B. food poisoning due to a preformed bacterial toxin in food
C. skin infection acquired from a dog bite
D. infection acquired from the stick of a contaminated needle

Answer

B

During an oral surgery, the surgeon nicked the patient's gum with a sharp instrument. This allowed *Streptococcus*, a bacterium normally present in the mouth, to gain access to the blood. As a result, the patient developed bacterial endocarditis (an infection of the heart). Which type of disease is this?

A. iatrogenic
B. nosocomial
C. vectors
D. zoonotic

Answer

A

Which period is the stage of disease during which the patient begins to present general signs and symptoms?

A. convalescence
B. incubation
C. illness
D. prodromal

Answer

D

A communicable disease that can be easily transmitted from person to person is which type of disease?

A. contagious

B. iatrogenic
C. acute
D. nosocomial

Answer

A

Fill in the Blank

A difference between an acute disease and chronic disease is that chronic diseases have an extended period of _____.

Answer

illness

A person steps on a rusty nail and develops tetanus. In this case, the person has acquired a(n) _____ disease.

Answer

noncommunicable

Short Answer

Brian goes to the hospital after not feeling well for a week. He has a fever of 38 °C (100.4 °F) and complains of nausea and a constant migraine. Distinguish between the signs and symptoms of disease in Brian's case.

Critical Thinking

Two periods of acute disease are the periods of illness and period of decline. (a) In what way are both of these periods similar? (b) In terms of quantity of pathogen, in what way are these periods different? (c) What initiates the period of decline?

In July 2015, a report[1] was released indicating the gram-negative bacterium *Pseudomonas aeruginosa* was found on hospital sinks 10 years after the initial outbreak in a neonatal intensive care unit. *P. aeruginosa* usually causes localized ear and eye infections but can cause pneumonia or septicemia in vulnerable individuals like newborn babies. Explain how the current discovery of the presence of this reported *P. aeruginosa* could lead to a recurrence of nosocomial disease.

16.2: How Pathogens Cause Disease

Koch's postulates are used to determine whether a particular microorganism is a pathogen. Molecular Koch's postulates are used to determine what genes contribute to a pathogen's ability to cause disease. Virulence, the degree to which a pathogen can cause disease, can be quantified by calculating either the ID50 or LD50 of a pathogen on a given population. Primary pathogens are capable of causing pathological changes associated with disease in a healthy individual.

Multiple Choice

Which of the following is a pathogen that could not be identified by the original Koch's postulates?

A. *Staphylococcus aureus*
B. *Pseudomonas aeruginosa*
C. Human immunodeficiency virus
D. *Salmonella enterica* serovar Typhimurium

Answer

C

Pathogen A has an ID_{50} of 50 particles, pathogen B has an ID_{50} of 1,000 particles, and pathogen C has an $ID_{50}$of 1×10^6 particles. Which pathogen is most virulent?

A. pathogen A
B. pathogen B

C. pathogen C

Answer

A

Fill in the Blank

A(n) _____ pathogen causes disease only when conditions are favorable for the microorganism because of transfer to an inappropriate body site or weakened immunity in an individual.

Answer

opportunistic

The concentration of pathogen needed to kill 50% of an infected group of test animals is the _____.

Answer

LD_{50}

A(n) _____ infection is a small region of infection from which a pathogen may move to another part of the body to establish a second infection.

Answer

focal

Cilia, fimbriae, and pili are all examples of structures used by microbes for _____.

Answer

adhesion

Critical Thinking

Diseases that involve biofilm-producing bacteria are of serious concern. They are not as easily treated compared with those involving free-floating (or planktonic) bacteria. Explain three reasons why biofilm formers are more pathogenic.

A microbiologist has identified a new gram-negative pathogen that causes liver disease in rats. She suspects that the bacterium's fimbriae are a virulence factor. Describe how molecular Koch's postulates could be used to test this hypothesis.

Acupuncture is a form of alternative medicine that is used for pain relief. Explain how acupuncture could facilitate exposure to pathogens.



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CHAPTER OVERVIEW

18: Disease and Epidemiology

In the United States and other developed nations, public health is a key function of government. A healthy citizenry is more productive, content, and prosperous; high rates of death and disease, on the other hand, can severely hamper economic productivity and foster social and political instability. The burden of disease makes it difficult for citizens to work consistently, maintain employment, and accumulate wealth to better their lives and support a growing economy.

In this chapter, we will explore the intersections between microbiology and epidemiology, the science that underlies public health. Epidemiology studies how disease originates and spreads throughout a population, with the goal of preventing outbreaks and containing them when they do occur. Over the past two centuries, discoveries in epidemiology have led to public health policies that have transformed life in developed nations, leading to the eradication (or near eradication) of many diseases that were once causes of great human suffering and premature death. However, the work of epidemiologists is far from finished. Numerous diseases continue to plague humanity, and new diseases are always emerging. Moreover, in the developing world, lack of infrastructure continues to pose many challenges to efforts to contain disease.

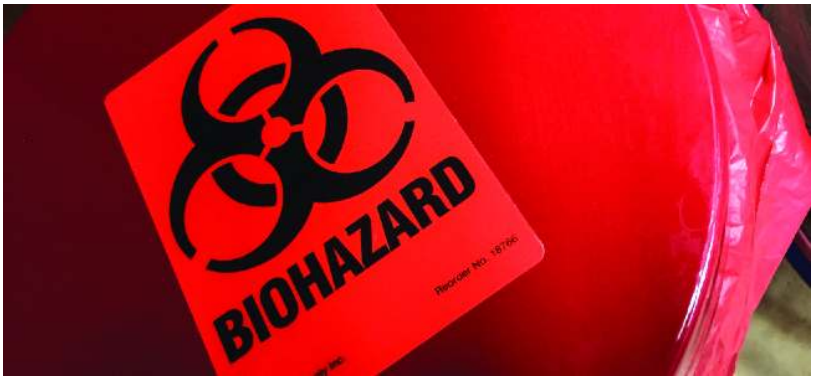


Figure 18.1: Signs like this may seem self-explanatory today, but a few short centuries ago, people lacked a basic understanding of how diseases spread. Microbiology has greatly contributed to the field of epidemiology, which focuses on containing the spread of disease. (credit: modification of work by Tony Webster)

18.1: The Language of Epidemiologists
18.2: Tracking Infectious Diseases
18.3: How Diseases Spread
18.4: Global Public Health
18.E: Disease and Epidemiology (Exercises)

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18.1: The Language of Epidemiologists

Learning Objectives

- Explain the difference between prevalence and incidence of disease
- Describe how morbidity and mortality rates are expressed
- Distinguish the characteristics of sporadic, endemic, epidemic, and pandemic diseases
- Explain the use of Koch's postulates and their modifications to determine the etiology of disease
- Explain the relationship between epidemiology and public health

The field of **epidemiology** concerns the geographical distribution and timing of infectious disease occurrences and how they are transmitted and maintained in nature, with the goal of recognizing and controlling outbreaks. The science of epidemiology includes **etiology** (the study of the causes of disease) and investigation of disease transmission (mechanisms by which a disease is spread).

Analyzing Disease in a Population

Epidemiological analyses are always carried out with reference to a population, which is the group of individuals that are at risk for the disease or condition. The population can be defined geographically, but if only a portion of the individuals in that area are susceptible, additional criteria may be required. Susceptible individuals may be defined by particular behaviors, such as intravenous drug use, owning particular pets, or membership in an institution, such as a college. Being able to define the population is important because most measures of interest in epidemiology are made with reference to the size of the population.

The state of being diseased is called **morbidity**. Morbidity in a population can be expressed in a few different ways. Morbidity or total morbidity is expressed in numbers of individuals without reference to the size of the population. The **morbidity rate** can be expressed as the number of diseased individuals out of a standard number of individuals in the population, such as 100,000, or as a percent of the population.

There are two aspects of morbidity that are relevant to an epidemiologist: a disease's **prevalence** and its **incidence**. Prevalence is the number, or proportion, of individuals with a particular illness in a given population at a point in time. For example, the Centers for Disease Control and Prevention (CDC) estimated that in 2012, there were about 1.2 million people 13 years and older with an active human immunodeficiency virus (HIV) infection. Expressed as a proportion, or rate, this is a prevalence of 467 infected persons per 100,000 in the population.[1] On the other hand, incidence is the number or proportion of *new* cases in a period of time. For the same year and population, the CDC estimates that there were 43,165 newly diagnosed cases of HIV infection, which is an incidence of 13.7 new cases per 100,000 in the population.[2] The relationship between incidence and prevalence can be seen in Figure **18.1.1**. For a chronic disease like HIV infection, prevalence will generally be higher than incidence because it represents the cumulative number of new cases over many years minus the number of cases that are no longer active (e.g., because the patient died or was cured).

In addition to morbidity rates, the incidence and prevalence of **mortality** (death) may also be reported. A mortality ratecan be expressed as the percentage of the population that has died from a disease or as the number of deaths per 100,000 persons (or other suitable standard number).

Drag the words into the correct boxes

In epidemiology, the state of being diseased is called _____ and the state of being dead is called _____. The number of diseased individuals in a population at a given time is the _____, whereas the number of new cases in a period of time is the _____.

morbidity
prevalence
mortality
incidence

Check

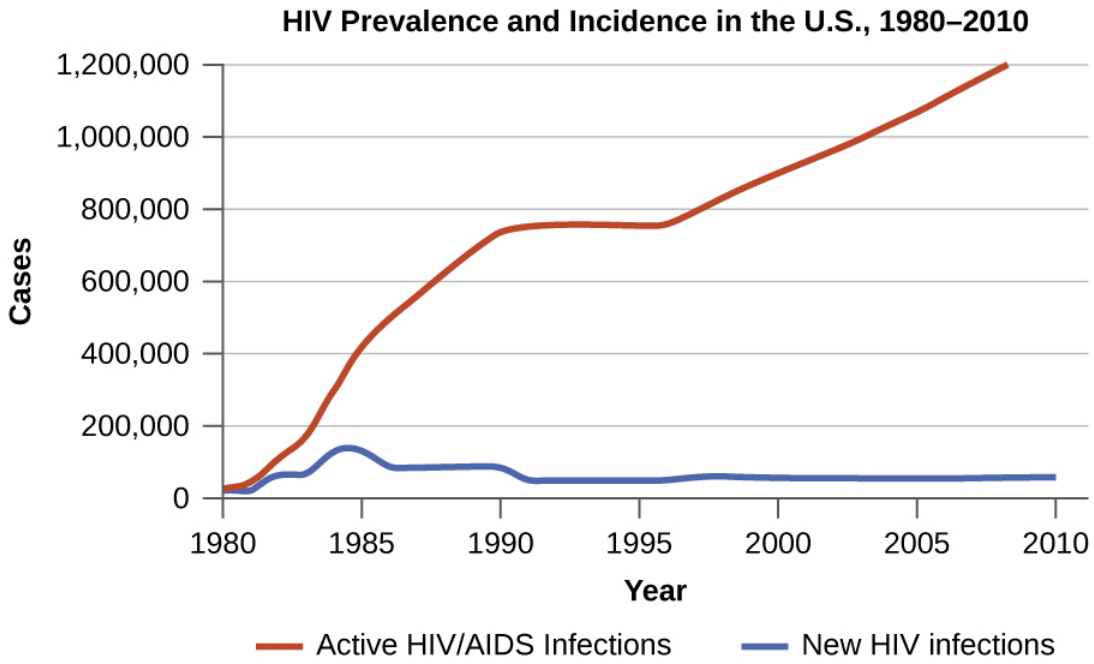


Figure **18.1.1**: This graph compares the incidence of HIV (the number of new cases reported each year) with the prevalence (the total number of cases each year). Prevalence and incidence can also be expressed as a rate or proportion for a given population.

Incidence rate and prevalence rate are often calculated as the number of people per 100,000 population, or as a percentage. For example, in 2003, it was reported that there were 44,232 new cases of AIDS in the US. The estimated mid-year population of the US in 2003 was 290 million (290,809,777). The incidence rate is calculated as: (44,232 / 290,809,777) X 100,000 = 15.21 new cases of AIDS per 100,000 population. In another example, a survey of 1,150 women who gave birth in Maine in 2000, a total of 468 reported taking a multivitamin at least 4 times a week during the month before becoming pregnant. The prevalence rate is calculated as (468 / 1,150) X 100 = 40.7%.

Here is an opportunity to practice the calculations as depicted in the HIV figure above.

Patterns of Incidence

Diseases that are seen only occasionally, and usually without geographic concentration, are called **sporadic diseases**. Examples of sporadic diseases include tetanus, rabies, and plague. In the United States, *Clostridium tetani*, the bacterium that causes tetanus, is ubiquitous in the soil environment, but incidences of infection occur only rarely and in scattered locations because most individuals

are vaccinated, clean wounds appropriately, or are only rarely in a situation that would cause infection.[3] Likewise in the United States there are a few scattered cases of plague each year, usually contracted from rodents in rural areas in the western states.[4]

Diseases that are constantly present (often at a low level) in a population within a particular geographic region are called **endemic diseases**. For example, malaria is endemic to some regions of Brazil, but is not endemic to the United States.

Diseases for which a larger than expected number of cases occurs in a short time within a geographic region are called **epidemic diseases**. Influenza is a good example of a commonly epidemic disease. Incidence patterns of influenza tend to rise each winter in the northern hemisphere. These seasonal increases are expected, so it would not be accurate to say that influenza is epidemic every winter; however, some winters have an usually large number of seasonal influenza cases in particular regions, and such situations would qualify as epidemics (Figure **18.1.2** and Figure **18.1.3**).

An epidemic disease signals the breakdown of an equilibrium in disease frequency, often resulting from some change in environmental conditions or in the population. In the case of influenza, the disruption can be due to antigenic shift or drift (see Virulence Factors of Bacterial and Viral Pathogens), which allows influenza virus strains to circumvent the acquired immunity of their human hosts.

An epidemic that occurs on a worldwide scale is called a **pandemic disease**. For example, HIV/AIDS is a pandemic disease and novel influenza virus strains often become pandemic.

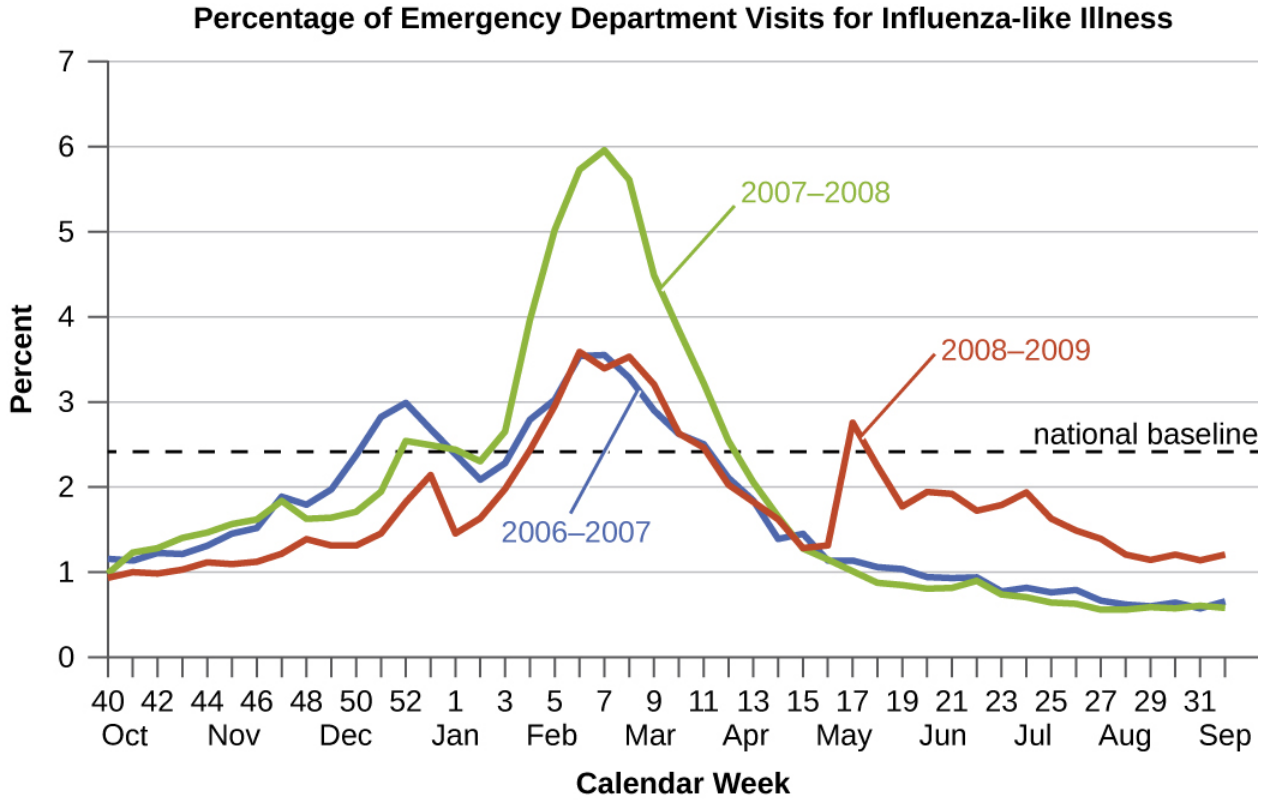


Figure **18.1.2**: The 2007–2008 influenza season in the United States saw much higher than normal numbers of visits to emergency departments for influenza-like symptoms as compared to the previous and the following years. (credit: modification of work by Centers for Disease Control and Prevention)

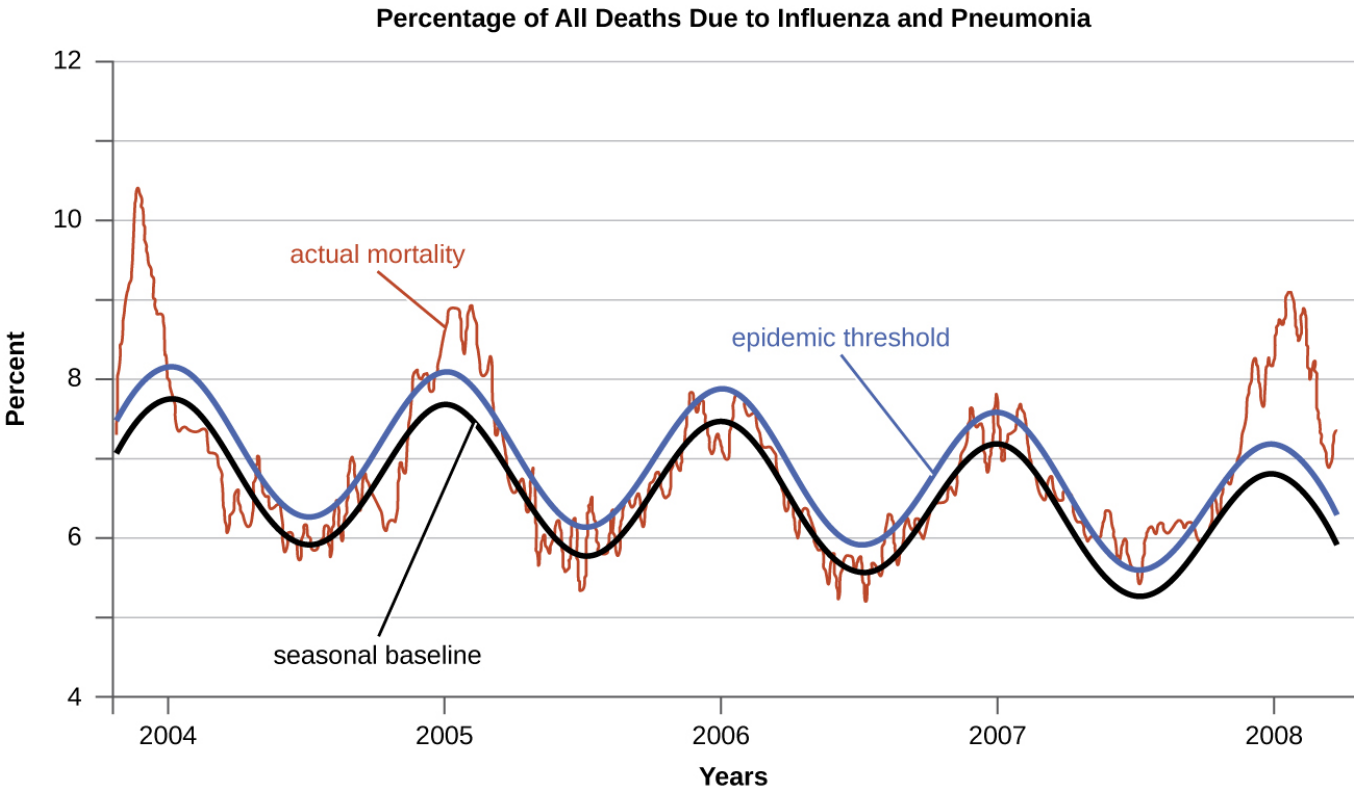


Figure **18.1.3**: The seasonal epidemic threshold (blue curve) is set by the CDC-based data from the previous five years. When actual mortality rates exceed this threshold, a disease is considered to be epidemic. As this graph shows, pneumonia- and influenza-related mortality saw pronounced epidemics during the winters of 2003–2004, 2005, and 2008. (credit: modification of work by Centers for Disease Control and Prevention)

Etiology

When studying an epidemic, an epidemiologist's first task is to determinate the cause of the disease, called the **etiologic agent** or **causative agent**. Connecting a disease to a specific pathogen can be challenging because of the extra effort typically required to demonstrate direct causation as opposed to a simple association. It is not enough to observe an association between a disease and a suspected pathogen; controlled experiments are needed to eliminate other possible causes. In addition, pathogens are typically difficult to detect when there is no immediate clue as to what is causing the outbreak. Signs and symptoms of disease are also commonly nonspecific, meaning that many different agents can give rise to the same set of signs and symptoms. This complicates diagnosis even when a causative agent is familiar to scientists.

Robert Koch was the first scientist to specifically demonstrate the causative agent of a disease (anthrax) in the late 1800s. Koch developed four criteria, now known as **Koch's postulates**, which had to be met in order to positively link a disease with a pathogenic microbe. Without Koch's postulates, the Golden Age of Microbiology would not have occurred. Between 1876 and 1905, many common diseases were linked with their etiologic agents, including cholera, diphtheria, gonorrhea, meningitis, plague, syphilis, tetanus, and tuberculosis. Today, we use the molecular Koch's postulates, a variation of Koch's original postulates that can be used to establish a link between the disease state and virulence traits unique to a pathogenic strain of a microbe. Koch's original postulates and molecular Koch's postulates were described in more detail in How Pathogens Cause Disease.

The Role of Public Health Organizations

The main national public health agency in the United States is the **Centers for Disease Control and Prevention (CDC)**, an agency of the Department of Health and Human Services. The CDC is charged with protecting the public from disease and injury. One way that the CDC carries out this mission is by overseeing the National Notifiable Disease Surveillance System (NNDSS) in cooperation with regional, state, and territorial public health departments. The NNDSS monitors diseases considered to be of public health importance on a national scale. Such diseases are called **notifiable diseases** or **reportable diseases** because all cases must

be reported to the CDC. A physician treating a patient with a notifiable disease is legally required to submit a report on the case. Notifiable diseases include HIV infection, measles, West Nile virus infections, and many others. Some states have their own lists of notifiable diseases that include diseases beyond those on the CDC's list.

Notifiable diseases are tracked by epidemiological studies and the data is used to inform health-care providers and the public about possible risks. The CDC publishes the ***Morbidity and Mortality Weekly Report* (*MMWR*)**, which provides physicians and health-care workers with updates on public health issues and the latest data pertaining to notifiable diseases. Table **18.1.1** is an example of the kind of data contained in the *MMWR*.

Table **18.1.1**: Incidence of Four Notifiable Diseases in the United States, Week Ending January 2, 2016

Disease	Current Week (Jan 2, 2016)	Median of Previous 52 Weeks	Maximum of Previous 52 Weeks	Cumulative Cases 2015
Campylobacteriosis	406	869	1,385	46,618
Chlamydia trachomatis infection	11,024	28,562	31,089	1,425,303
Giardiasis	115	230	335	11,870
Gonorrhea	3,207	7,155	8,283	369,926

Link to Learning

The current *Morbidity and Mortality Weekly Report* is available online.

Key Concepts and Summary

- **Epidemiology** is the science underlying public health.
- **Morbidity** means being in a state of illness, whereas **mortality** refers to death; both **morbidity rates** and **mortality rates** are of interest to epidemiologists.
- **Incidence** is the number of new cases (morbidity or mortality), usually expressed as a proportion, during a specified time period; **prevalence** is the total number affected in the population, again usually expressed as a proportion.
- **Sporadic diseases** only occur rarely and largely without a geographic focus. **Endemic diseases** occur at a constant (and often low) level within a population. **Epidemic diseases** and **pandemic diseases** occur when an outbreak occurs on a significantly larger than expected level, either locally or globally, respectively.
- **Koch's postulates** specify the procedure for confirming a particular pathogen as the etiologic agent of a particular disease. Koch's postulates have limitations in application if the microbe cannot be isolated and cultured or if there is no animal host for the microbe. In this case, molecular Koch's postulates would be utilized.
- In the United States, the **Centers for Disease Control and Prevention** monitors **notifiable diseases** and publishes weekly updates in the *Morbidity and Mortality Weekly Report*.

Footnotes

1. 1 H. Irene Hall, Qian An, Tian Tang, Ruiguang Song, Mi Chen, Timothy Green, and Jian Kang. "Prevalence of Diagnosed and Undiagnosed HIV Infection—United States, 2008–2012." *Morbidity and Mortality Weekly Report* 64, no. 24 (2015): 657–662.
2. 2 Centers for Disease Control and Prevention. "Diagnoses of HIV Infection in the United States and Dependent Areas, 2014." *HIV Surveillance Report* 26 (2015).
3. 3 Centers for Disease Control and Prevention. "Tetanus Surveillance—United States, 2001–2008." *Morbidity and Mortality Weekly Report* 60, no. 12 (2011): 365.
4. 4 Centers for Disease Control and Prevention. "Plague in the United States." 2015. http://www.cdc.gov/plague/maps. Accessed June 1, 2016.

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18.2: Tracking Infectious Diseases

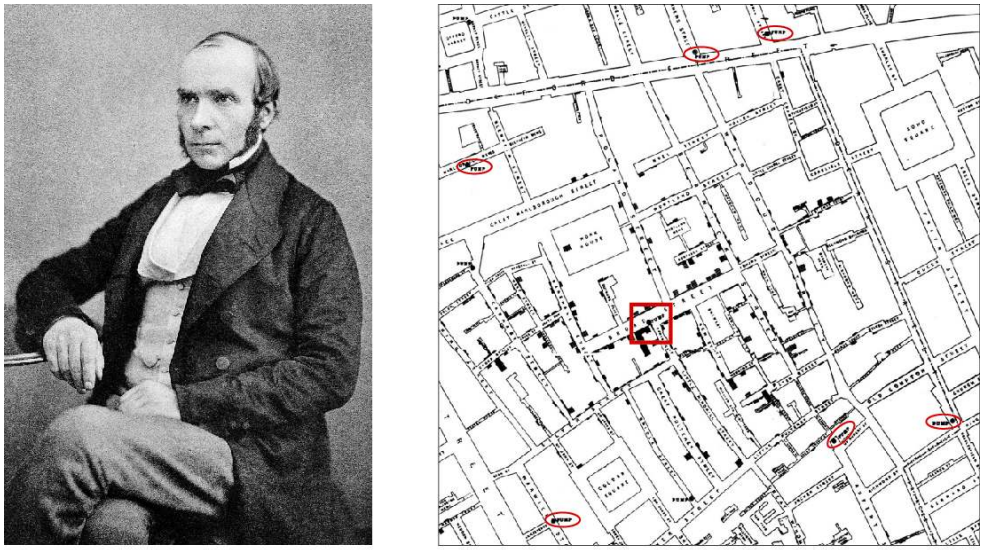
Learning Objectives

- Describe the contributions of John Snow to the field of epidemiology
- Explain how Florence Nightingale used statistical analysis and sanitation reforms to reduce mortality rate in military hospitals
- Analyze the impact of Joseph Lister's introduction to aseptic techniques in surgery

Epidemiology has its roots in the work of physicians who looked for patterns in disease occurrence as a way to understand how to prevent it. The idea that disease could be transmitted was an important precursor to making sense of some of the patterns. In 1546, Girolamo Fracastoro first proposed the germ theory of disease in his essay *De Contagione et Contagiosis Morbis*, but this theory remained in competition with other theories, such as the miasma hypothesis, for many years (see What Our Ancestors Knew). Uncertainty about the cause of disease was not an absolute barrier to obtaining useful knowledge from patterns of disease. Some important researchers, such as Florence Nightingale, subscribed to the miasma hypothesis. The transition to acceptance of the germ theory during the 19th century provided a solid mechanistic grounding to the study of disease patterns. The studies of 19th century physicians and researchers such as John Snow, Florence Nightingale, Ignaz Semmelweis, Joseph Lister, Robert Koch, Louis Pasteur, and others sowed the seeds of modern epidemiology.

Pioneers of Epidemiology

John Snow (Figure 18.2.1) was a British physician known as the father of epidemiology for determining the source of the 1854 Broad Street cholera epidemic in London. Based on observations he had made during an earlier cholera outbreak (1848–1849), Snow proposed that cholera was spread through a fecal-oral route of transmission and that a microbe was the infectious agent. He investigated the 1854 cholera epidemic in two ways. First, suspecting that contaminated water was the source of the epidemic, Snow identified the source of water for those infected. He found a high frequency of cholera cases among individuals who obtained their water from the River Thames downstream from London. This water contained the refuse and sewage from London and settlements upstream. He also noted that brewery workers did not contract cholera and on investigation found the owners provided the workers with beer to drink and stated that they likely did not drink water.[1] Second, he also painstakingly mapped the incidence of cholera and found a high frequency among those individuals using a particular water pump located on Broad Street. In response to Snow's advice, local officials removed the pump's handle,[2] resulting in the containment of the Broad Street cholera epidemic.



(a) (b)

Figure 18.2.1: (a) John Snow (1813–1858), British physician and father of epidemiology. (b) Snow's detailed mapping of cholera incidence led to the discovery of the contaminated water pump on Broad street (red square) responsible for the 1854 cholera epidemic. (credit a: modification of work by "Rsabbatini"/Wikimedia Commons)

Snow's work represents an early epidemiological study and it resulted in the first known public health response to an **epidemic**. Snow's meticulous case-tracking methods are now common practice in studying disease outbreaks and in associating new diseases with their causes. His work further shed light on unsanitary sewage practices and the effects of waste dumping in the Thames. Additionally, his work supported the germ theory of disease, which argued disease could be transmitted through contaminated items, including water contaminated with fecal matter.

Snow's work illustrated what is referred to today as a **common source spread** of infectious disease, in which there is a single source for all of the individuals infected. In this case, the single source was the contaminated well below the Broad Street pump. Types of common source spread include point source spread, continuous common source spread, and intermittent common source spread. In **point source spread** of infectious disease, the common source operates for a short time period—less than the incubation period of the pathogen. An example of point source spread is a single contaminated potato salad at a group picnic. In **continuous common source spread**, the infection occurs for an extended period of time, longer than the incubation period. An example of continuous common source spread would be the source of London water taken downstream of the city, which was continuously contaminated with sewage from upstream. Finally, with **intermittent common source spread**, infections occur for a period, stop, and then begin again. This might be seen in infections from a well that was contaminated only after large rainfalls and that cleared itself of contamination after a short period.

In contrast to common source spread, propagated spread occurs through direct or indirect person-to-person contact. With **propagated spread**, there is no single source for infection; each infected individual becomes a source for one or more subsequent infections. With propagated spread, unless the spread is stopped immediately, infections occur for longer than the incubation period. Although point sources often lead to large-scale but localized outbreaks of short duration, propagated spread typically results in longer duration outbreaks that can vary from small to large, depending on the population and the disease (Figure 18.2.2). In addition, because of person-to-person transmission, propagated spread cannot be easily stopped at a single source like point source spread.

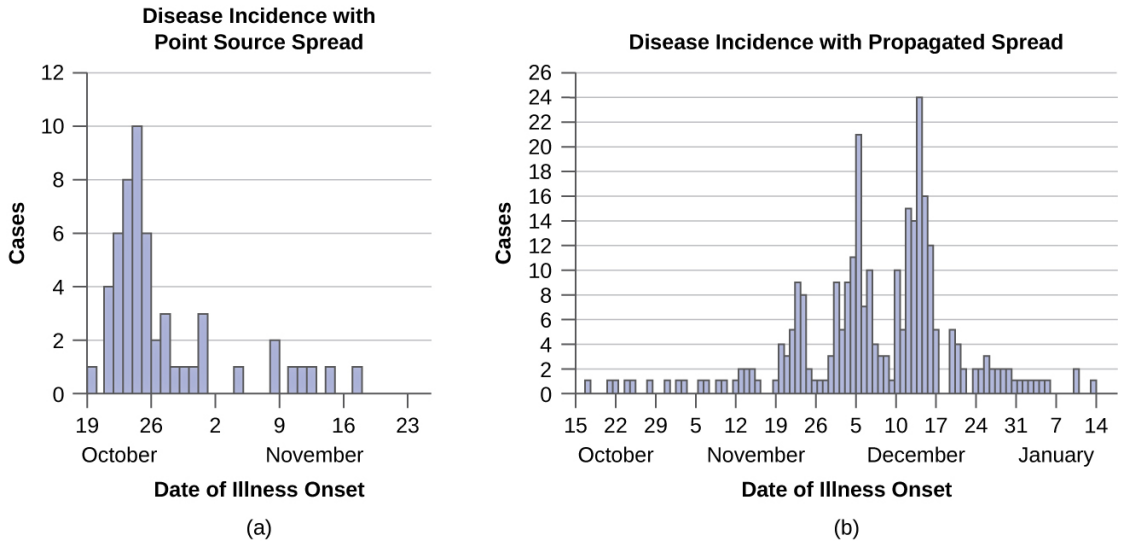
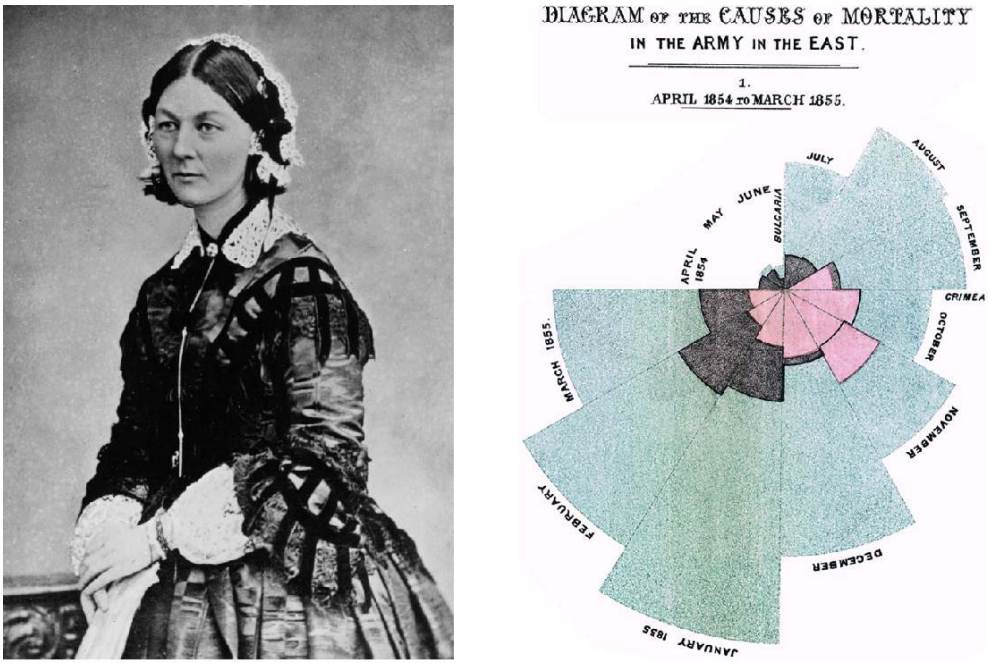


Figure 18.2.2: (a) Outbreaks that can be attributed to point source spread often have a short duration. (b) Outbreaks attributed to propagated spread can have a more extended duration. (credit a, b: modification of work by Centers for Disease Control and Prevention)

Florence Nightingale's work is another example of an early epidemiological study. In 1854, Nightingale was part of a contingent of nurses dispatched by the British military to care for wounded soldiers during the Crimean War. Nightingale kept meticulous records regarding the causes of illness and death during the war. Her recordkeeping was a fundamental task of what would later become the science of epidemiology. Her analysis of the data she collected was published in 1858. In this book, she presented monthly frequency data on causes of death in a wedge chart histogram (Figure 18.2.3). This graphical presentation of data, unusual at the time, powerfully illustrated that the vast majority of casualties during the war occurred not due to wounds sustained in action

but to what Nightingale deemed preventable infectious diseases. Often these diseases occurred because of poor sanitation and lack of access to hospital facilities. Nightingale's findings led to many reforms in the British military's system of medical care.

Joseph Lister provided early epidemiological evidence leading to good public health practices in clinics and hospitals. These settings were notorious in the mid-1800s for fatal infections of surgical wounds at a time when the germ theory of disease was not yet widely accepted (see Foundations of Modern Cell Theory). Most physicians did not wash their hands between patient visits or clean and sterilize their surgical tools. Lister, however, discovered the disinfecting properties of carbolic acid, also known as phenol (see Using Chemicals to Control Microorganisms). He introduced several disinfection protocols that dramatically lowered post-surgical infection rates.[3] He demanded that surgeons who worked for him use a 5% carbolic acid solution to clean their surgical tools between patients, and even went so far as to spray the solution onto bandages and over the surgical site during operations (Figure **18.2.4**). He also took precautions not to introduce sources of infection from his skin or clothing by removing his coat, rolling up his sleeves, and washing his hands in a dilute solution of carbolic acid before and during the surgery.



(a) (b)

Figure **18.2.3**: (a) Florence Nightingale reported on the data she collected as a nurse in the Crimean War. (b) Nightingale's diagram shows the number of fatalities in soldiers by month of the conflict from various causes. The total number dead in a particular month is equal to the area of the wedge for that month. The colored sections of the wedge represent different causes of death: wounds (pink), preventable infectious diseases (gray), and all other causes (brown).

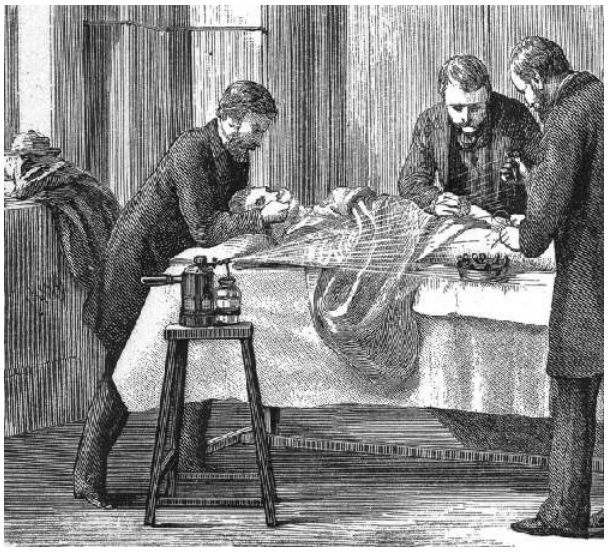


Figure 18.2.4: Joseph Lister initiated the use of a carbolic acid (phenol) during surgeries. This illustration of a surgery shows a pressurized canister of carbolic acid being sprayed over the surgical site.

Link to Learning

This CDC resource further breaks down the pattern expected from a point-source outbreak.

Learn more about Nightingale's wedge chart here.

Key Concepts and Summary

- Early pioneers of epidemiology such as John Snow, Florence Nightingale, and Joseph Lister, studied disease at the population level and used data to disrupt disease transmission.

Footnotes

1. John Snow. *On the Mode of Communication of Cholera. Second edition, Much Enlarged*. John Churchill, 1855.
2. John Snow. "The Cholera near Golden-Wquare, and at Deptford." *Medical Times and Gazette* 9 (1854): 321–322. http://www.ph.ucla.edu/epi/snow/chol...densquare.html.
3. O.M. Lidwell. "Joseph Lister and Infection from the Air." *Epidemiology and Infection* 99 (1987): 569–578. www.ncbi.nlm.nih.gov/pmc/arti...00006-0004.pdf.

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18.3: How Diseases Spread

Learning Objectives

- Describe the different types of disease reservoirs
- Compare contact, vector, and vehicle modes of transmission
- Identify important disease vectors
- Explain the prevalence of nosocomial infections

Understanding how infectious pathogens spread is critical to preventing infectious disease. Many pathogens require a living host to survive, while others may be able to persist in a dormant state outside of a living host. But having infected one host, all pathogens must also have a mechanism of transfer from one host to another or they will die when their host dies. Pathogens often have elaborate adaptations to exploit host biology, behavior, and ecology to live in and move between hosts. Hosts have evolved defenses against pathogens, but because their rates of evolution are typically slower than their pathogens (because their generation times are longer), hosts are usually at an evolutionary disadvantage. This section will explore where pathogens survive—both inside and outside hosts—and some of the many ways they move from one host to another.

Reservoirs and Carriers

For pathogens to persist over long periods of time they require **reservoirs** where they normally reside. Reservoirs can be living organisms or nonliving sites. Nonliving reservoirs can include soil and water in the environment. These may naturally harbor the organism because it may grow in that environment. These environments may also become contaminated with pathogens in human feces, pathogens shed by intermediate hosts, or pathogens contained in the remains of intermediate hosts.

Pathogens may have mechanisms of dormancy or resilience that allow them to survive (but typically not to reproduce) for varying periods of time in nonliving environments. For example, *Clostridium tetani* survives in the soil and in the presence of oxygen as a resistant endospore. Although many viruses are soon destroyed once in contact with air, water, or other non-physiological conditions, certain types are capable of persisting outside of a living cell for varying amounts of time. For example, a study that looked at the ability of influenza viruses to infect a cell culture after varying amounts of time on a banknote showed survival times from 48 hours to 17 days, depending on how they were deposited on the banknote.[1] On the other hand, cold-causing rhinoviruses are somewhat fragile, typically surviving less than a day outside of physiological fluids.

A human acting as a reservoir of a pathogen may or may not be capable of transmitting the pathogen, depending on the stage of infection and the pathogen. To help prevent the spread of disease among school children, the CDC has developed guidelines based on the risk of transmission during the course of the disease. For example, children with chickenpox are considered contagious for five days from the start of the rash, whereas children with most gastrointestinal illnesses should be kept home for 24 hours after the symptoms disappear.

An individual capable of transmitting a pathogen without displaying symptoms is referred to as a **carrier**. A **passive carrier** is contaminated with the pathogen and can mechanically transmit it to another host; however, a passive carrier is not infected. For example, a health-care professional who fails to wash his hands after seeing a patient harboring an infectious agent could become a passive carrier, transmitting the pathogen to another patient who becomes infected.

By contrast, an **active carrier** is an infected individual who can transmit the disease to others. An active carrier may or may not exhibit signs or symptoms of infection. For example, active carriers may transmit the disease during the incubation period (before they show signs and symptoms) or the period of convalescence (after symptoms have subsided). Active carriers who do not present signs or symptoms of disease despite infection are called **asymptomatic carriers**. Pathogens such as hepatitis B virus, herpes simplex virus, and HIV are frequently transmitted by asymptomatic carriers. Mary Mallon, better known as Typhoid Mary, is a famous historical example of an asymptomatic carrier. An Irish immigrant, Mallon worked as a cook for households in and around New York City between 1900 and 1915. In each household, the residents developed typhoid fever (caused by *Salmonella typhi*) a few weeks after Mallon started working. Later investigations determined that Mallon was responsible for at least 122 cases of typhoid fever, five of which were fatal.[2] See Eye on Ethics: Typhoid Mary for more about the Mallon case.

A pathogen may have more than one living reservoir. In zoonotic diseases, animals act as reservoirs of human disease and transmit the infectious agent to humans through direct or indirect contact. In some cases, the disease also affects the animal, but in other cases the animal is asymptomatic.

In parasitic infections, the parasite's preferred host is called the definitive host. In parasites with complex life cycles, the definitive host is the host in which the parasite reaches sexual maturity. Some parasites may also infect one or more intermediate hosts in which the parasite goes through several immature life cycle stages or reproduces asexually.

Link to Learning

George Soper, the sanitary engineer who traced the typhoid outbreak to Mary Mallon, gives an account of his investigation, an example of descriptive epidemiology, in "The Curious Career of Typhoid Mary."

Transmission

Regardless of the reservoir, transmission must occur for an infection to spread. First, transmission from the reservoir to the individual must occur. Then, the individual must transmit the infectious agent to other susceptible individuals, either directly or indirectly. Pathogenic microorganisms employ diverse transmission mechanisms.

Contact Transmission

Contact transmission includes direct contact or indirect contact. Person-to-person transmission is a form of **direct contact transmission**. Here the agent is transmitted by physical contact between two individuals (Figure 18.3.1) through actions such as touching, kissing, sexual intercourse, or droplet sprays. Direct contact can be categorized as vertical, horizontal, or droplet transmission. **Vertical direct contact transmission** occurs when pathogens are transmitted from mother to child during pregnancy, birth, or breastfeeding. Other kinds of direct contact transmission are called **horizontal direct contact transmission**. Often, contact between mucous membranes is required for entry of the pathogen into the new host, although skin-to-skin contact can lead to mucous membrane contact if the new host subsequently touches a mucous membrane. Contact transmission may also be site-specific; for example, some diseases can be transmitted by sexual contact but not by other forms of contact.

When an individual coughs or sneezes, small droplets of mucus that may contain pathogens are ejected. This leads to **direct droplet transmission**, which refers to droplet transmission of a pathogen to a new host over distances of one meter or less. A wide variety of diseases are transmitted by droplets, including influenza and many forms of pneumonia. Transmission over distances greater than one meter is called airborne transmission.

Indirect contact transmission involves inanimate objects called fomites that become contaminated by pathogens from an infected individual or reservoir (Figure 18.3.2). For example, an individual with the common cold may sneeze, causing droplets to land on a fomite such as a tablecloth or carpet, or the individual may wipe her nose and then transfer mucus to a fomite such as a doorknob or towel. Transmission occurs indirectly when a new susceptible host later touches the fomite and transfers the contaminated material to a susceptible portal of entry. Fomites can also include objects used in clinical settings that are not properly sterilized, such as syringes, needles, catheters, and surgical equipment. Pathogens transmitted indirectly via such fomites are a major cause of healthcare-associated infections (see Controlling Microbial Growth).




Figure 18.3.1: Direct contact transmission of pathogens can occur through physical contact. Many pathogens require contact with a mucous membrane to enter the body, but the host may transfer the pathogen from another point of contact (e.g., hand) to a mucous membrane (e.g., mouth or eye). (credit left: modification of work by Lisa Doehnert)






Figure 18.3.2: Fomites are nonliving objects that facilitate the indirect transmission of pathogens. Contaminated doorknobs, towels, and syringes are all common examples of fomites. (credit left: modification of work by Kate Ter Haar; credit middle: modification of work by Vernon Swanepoel; credit right: modification of work by "Zaldylmg"/Flickr)

Vehicle Transmission

The term **vehicle transmission** refers to the transmission of pathogens through vehicles such as water, food, and air. Water contamination through poor sanitation methods leads to waterborne transmission of disease. Waterborne disease remains a serious problem in many regions throughout the world. The World Health Organization (WHO) estimates that contaminated drinking water is responsible for more than 500,000 deaths each year.[3] Similarly, food contaminated through poor handling or storage can lead to foodborne transmission of disease (Figure 18.3.3).

Dust and fine particles known as aerosols, which can float in the air, can carry pathogens and facilitate the airborne transmission of disease. For example, dust particles are the dominant mode of transmission of hantavirus to humans. Hantavirus is found in mouse feces, urine, and saliva, but when these substances dry, they can disintegrate into fine particles that can become airborne when disturbed; inhalation of these particles can lead to a serious and sometimes fatal respiratory infection.

Although droplet transmission over short distances is considered contact transmission as discussed above, longer distance transmission of droplets through the air is considered vehicle transmission. Unlike larger particles that drop quickly out of the air column, fine mucus droplets produced by coughs or sneezes can remain suspended for long periods of time, traveling considerable distances. In certain conditions, droplets desiccate quickly to produce a droplet nucleus that is capable of transmitting pathogens; air temperature and humidity can have an impact on effectiveness of airborne transmission.

Tuberculosis is often transmitted via airborne transmission when the causative agent, *Mycobacterium tuberculosis*, is released in small particles with coughs. Because tuberculosis requires as few as 10 microbes to initiate a new infection, patients with tuberculosis must be treated in rooms equipped with special ventilation, and anyone entering the room should wear a mask.



Figure 18.3.3: Food is an important vehicle of transmission for pathogens, especially of the gastrointestinal and upper respiratory systems. Notice the glass shield above the food trays, designed to prevent pathogens ejected in coughs and sneezes from entering the food. (credit: Fort George G. Meade Public Affairs Office)

Vector Transmission

Diseases can also be transmitted by a mechanical or biological vector, an animal (typically an arthropod) that carries the disease from one host to another. **Mechanical transmission** is facilitated by a **mechanical vector**, an animal that carries a pathogen from one host to another without being infected itself. For example, a fly may land on fecal matter and later transmit bacteria from the feces to food that it lands on; a human eating the food may then become infected by the bacteria, resulting in a case of diarrhea or dysentery (Figure 18.3.4).

Biological transmission occurs when the pathogen reproduces within a **biological vector** that transmits the pathogen from one host to another (Figure 18.3.4). Arthropods are the main vectors responsible for biological transmission (Figure 18.3.5). Most arthropod vectors transmit the pathogen by biting the host, creating a wound that serves as a portal of entry. The pathogen may go through part of its reproductive cycle in the gut or salivary glands of the arthropod to facilitate its transmission through the bite. For example, hemipterans (called “kissing bugs” or “assassin bugs”) transmit Chagas disease to humans by defecating when they bite, after which the human scratches or rubs the infected feces into a mucous membrane or break in the skin.

Biological insect vectors include mosquitoes, which transmit malaria and other diseases, and lice, which transmit typhus. Other arthropod vectors can include arachnids, primarily ticks, which transmit Lyme disease and other diseases, and mites, which transmit scrub typhus and rickettsial pox. Biological transmission, because it involves survival and reproduction within a parasitized vector, complicates the biology of the pathogen and its transmission. There are also important non-arthropod vectors of disease, including mammals and birds. Various species of mammals can transmit rabies to humans, usually by means of a bite that transmits the rabies virus. Chickens and other domestic poultry can transmit avian influenza to humans through direct or indirect contact with avian influenza virus A shed in the birds’ saliva, mucous, and feces.

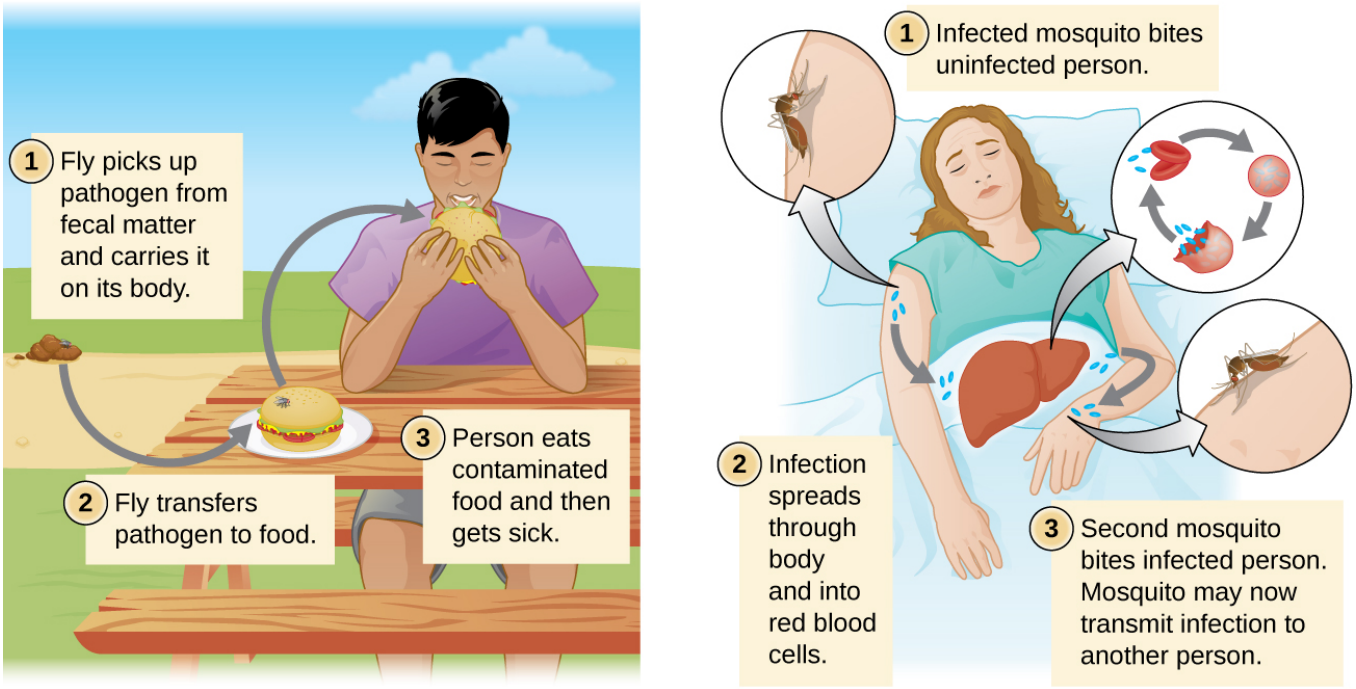


Figure 18.3.4: (a) A mechanical vector carries a pathogen on its body from one host to another, not as an infection. (b) A biological vector carries a pathogen from one host to another after becoming infected itself.

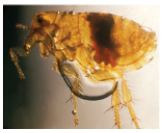
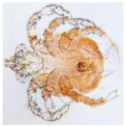
Common Arthropod Vectors and Select Pathogens			
Vector	**Species**	**Pathogen**	**Disease**
Black fly 	*Simulium* spp.	*Onchocerca volvulus*	Onchocerciasis (river blindness)
Flea 	*Xenopsylla cheopis*	*Rickettsia typhi*	Murine typhus
		Yersinia pestis	Plague
Kissing bug 	*Triatoma* spp.	*Trypanosoma cruzi*	Chagas disease
Louse 	*Pediculus humanus humanus*	*Bartonella quintana*	Trench fever
		Borrelia recurrentis	Relapsing fever
		Rickettsia prowazekii	Typhus
Mite (chigger) 	*Leptotrombidium* spp.	*Orientia tsutsugamushi*	Scrub typhus
	Liponyssoides sanguineus	*Rickettsia akari*	Rickettsialpox
Mosquito 	*Aedes* spp., *Haemagogus* spp.	*Yellow fever virus*	Yellow fever
	Anopheles spp.	*Plasmodium falciparum*	Malaria
	Culex pipiens	*West Nile virus*	West Nile disease
Sand fly 	*Phlebotomus* spp.	*Leishmania* spp.	Leishmaniasis
Tick 	*Ixodes* spp.	*Borrelia* spp.	Lyme disease
	Dermacentor spp. and others	*Rickettsia rickettsii*	Rocky Mountain spotted fever
Tsetse fly 	*Glossina* spp.	*Trypanosoma brucei*	African trypanosomiasis (sleeping sickness)

Figure 18.3.5: (credit “Black fly”, “Tick”, “Tsetse fly”: modification of work by USDA; credit: “Flea”: modification of work by Centers for Disease Control and Prevention; credit: “Louse”, “Mosquito”, “Sand fly”: modification of work by James Gathany, Centers for Disease Control and Prevention; credit “Kissing bug”: modification of work by Glenn Seplak; credit “Mite”: modification of work by Michael Wunderli)

Using GMOs to Stop the Spread of Zika

In 2016, an epidemic of the Zika virus was linked to a high incidence of birth defects in South America and Central America. As winter turned to spring in the northern hemisphere, health officials correctly predicted the virus would spread to North America, coinciding with the breeding season of its major vector, the *Aedes aegypti* mosquito.

The range of the *A. aegypti* mosquito extends well into the southern United States (Figure **18.3.6**). Because these same mosquitoes serve as vectors for other problematic diseases (dengue fever, yellow fever, and others), various methods of mosquito control have been proposed as solutions. Chemical pesticides have been used effectively in the past, and are likely to be used again; but because chemical pesticides can have negative impacts on the environment, some scientists have proposed an alternative that involves genetically engineering *A. aegypti* so that it cannot reproduce. This method, however, has been the subject of some controversy.

One method that has worked in the past to control pests, with little apparent downside, has been sterile male introductions. This method controlled the screw-worm fly pest in the southwest United States and fruit fly pests of fruit crops. In this method, males of the target species are reared in the lab, sterilized with radiation, and released into the environment where they mate with wild females, who subsequently bear no live offspring. Repeated releases shrink the pest population.

A similar method, taking advantage of recombinant DNA technology,[4] introduces a dominant lethal allele into male mosquitoes that is suppressed in the presence of tetracycline (an antibiotic) during laboratory rearing. The males are released into the environment and mate with female mosquitoes. Unlike the sterile male method, these matings produce offspring, but they die as larvae from the lethal gene in the absence of tetracycline in the environment. As of 2016, this method has yet to be implemented in the United States, but a UK company tested the method in Piracicaba, Brazil, and found an 82% reduction in wild *A. aegypti* larvae and a 91% reduction in dengue cases in the treated area.[5] In August 2016, amid news of Zika infections in several Florida communities, the FDA gave the UK company permission to test this same mosquito control method in Key West, Florida, pending compliance with local and state regulations and a referendum in the affected communities.

The use of genetically modified organisms (GMOs) to control a disease vector has its advocates as well as its opponents. In theory, the system could be used to drive the *A. aegypti* mosquito extinct—a noble goal according to some, given the damage they do to human populations.[6] But opponents of the idea are concerned that the gene could escape the species boundary of *A. aegypti* and cause problems in other species, leading to unforeseen ecological consequences. Opponents are also wary of the program because it is being administered by a for-profit corporation, creating the potential for conflicts of interest that would have to be tightly regulated; and it is not clear how any unintended consequences of the program could be reversed.

There are other epidemiological considerations as well. *Aedes aegypti* is apparently not the only vector for the Zika virus. *Aedes albopictus*, the Asian tiger mosquito, is also a vector for the Zika virus.[7] *A. albopictus* is now widespread around the planet including much of the United States (Figure **18.3.6**). Many other mosquitoes have been found to harbor Zika virus, though their capacity to act as vectors is unknown.[8] Genetically modified strains of *A. aegypti* will not control the other species of vectors. Finally, the Zika virus can apparently be transmitted sexually between human hosts, from mother to child, and possibly through blood transfusion. All of these factors must be considered in any approach to controlling the spread of the virus.

Clearly there are risks and unknowns involved in conducting an open-environment experiment of an as-yet poorly understood technology. But allowing the Zika virus to spread unchecked is also risky. Does the threat of a Zika epidemic justify the ecological risk of genetically engineering mosquitos? Are current methods of mosquito control sufficiently ineffective or harmful that we need to try untested alternatives? These are the questions being put to public health officials now.

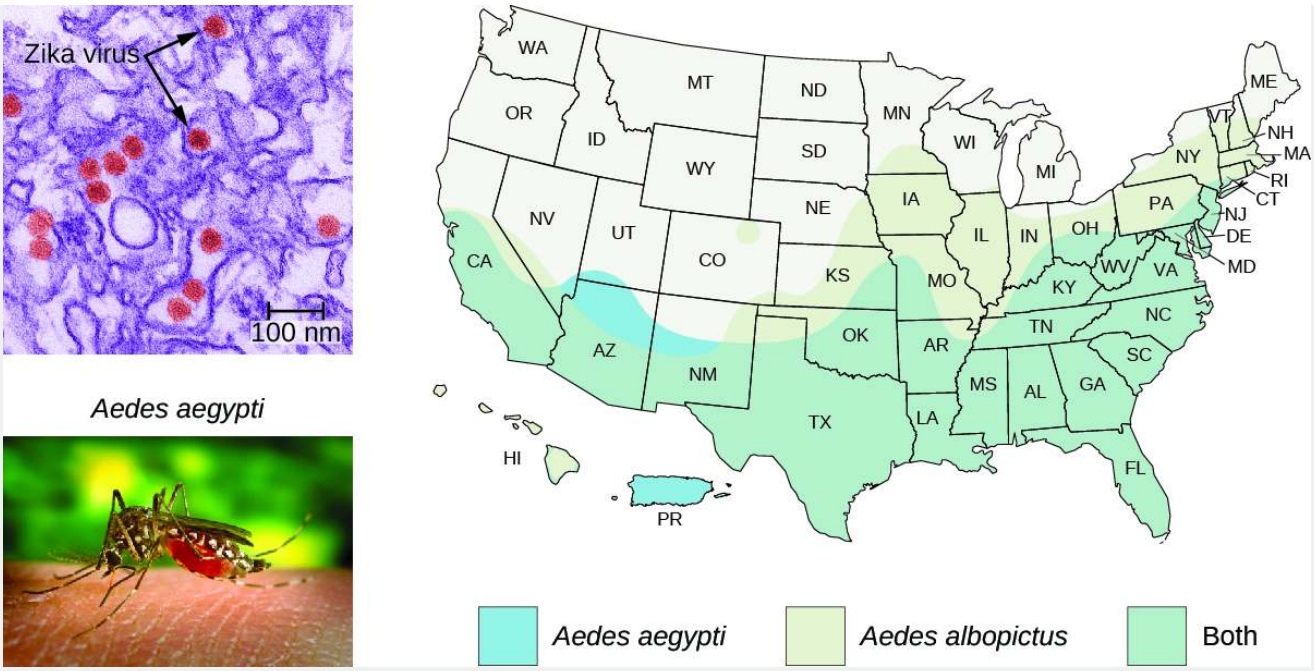


Figure 18.3.6: The Zika virus is an enveloped virus transmitted by mosquitoes, especially *Aedes aegypti*. The range of this mosquito includes much of the United States, from the Southwest and Southeast to as far north as the Mid-Atlantic. The range of *A. albopictus*, another vector, extends even farther north to New England and parts of the Midwest. (credit micrograph: modification of work by Cynthia Goldsmith, Centers for Disease Control and Prevention; credit photo: modification of work by James Gathany, Centers for Disease Control and Prevention; credit map: modification of work by Centers for Disease Control and Prevention)

Quarantining

Individuals suspected or known to have been exposed to certain contagious pathogens may be **quarantined**, or isolated to prevent transmission of the disease to others. Hospitals and other health-care facilities generally set up special wards to isolate patients with particularly hazardous diseases such as tuberculosis or Ebola (Figure 18.3.7). Depending on the setting, these wards may be equipped with special air-handling methods, and personnel may implement special protocols to limit the risk of transmission, such as personal protective equipment or the use of chemical disinfectant sprays upon entry and exit of medical personnel.

The duration of the quarantine depends on factors such as the incubation period of the disease and the evidence suggestive of an infection. The patient may be released if signs and symptoms fail to materialize when expected or if preventive treatment can be administered in order to limit the risk of transmission. If the infection is confirmed, the patient may be compelled to remain in isolation until the disease is no longer considered contagious.

In the United States, public health authorities may only quarantine patients for certain diseases, such as cholera, diphtheria, infectious tuberculosis, and strains of influenza capable of causing a pandemic. Individuals entering the United States or moving between states may be quarantined by the CDC if they are suspected of having been exposed to one of these diseases. Although the CDC routinely monitors entry points to the United States for crew or passengers displaying illness, quarantine is rarely implemented.



(a) (b)

Figure 18.3.7: (a) The Aeromedical Biological Containment System (ABCS) is a module designed by the CDC and Department of Defense specifically for transporting highly contagious patients by air. (b) An isolation ward for Ebola patients in Lagos, Nigeria. (credit a: modification of work by Centers for Disease Control and Prevention; credit b: modification of work by CDC Global)

Healthcare-Associated (Nosocomial) Infections

Hospitals, retirement homes, and prisons attract the attention of epidemiologists because these settings are associated with increased incidence of certain diseases. Higher rates of transmission may be caused by characteristics of the environment itself, characteristics of the population, or both. Consequently, special efforts must be taken to limit the risks of infection in these settings.

Infections acquired in health-care facilities, including hospitals, are called **nosocomial infections** or **healthcare-associated infections (HAI)**. HAIs are often connected with surgery or other invasive procedures that provide the pathogen with access to the portal of infection. For an infection to be classified as an HAI, the patient must have been admitted to the health-care facility for a reason other than the infection. In these settings, patients suffering from primary disease are often afflicted with compromised immunity and are more susceptible to secondary infection and opportunistic pathogens.

In 2011, more than 720,000 HAIs occurred in hospitals in the United States, according to the CDC. About 22% of these HAIs occurred at a surgical site, and cases of pneumonia accounted for another 22%; urinary tract infections accounted for an additional 13%, and primary bloodstream infections 10%.[9] Such HAIs often occur when pathogens are introduced to patients' bodies through contaminated surgical or medical equipment, such as catheters and respiratory ventilators. Health-care facilities seek to limit nosocomial infections through training and hygiene protocols such as those described in Control of Microbial Growth.

Key Concepts and Summary

- **Reservoirs** of human disease can include the human and animal populations, soil, water, and inanimate objects or materials.
- **Contact transmission** can be **direct** or **indirect** through physical contact with either an infected host (direct) or contact with a fomite that an infected host has made contact with previously (indirect).
- Vector transmission occurs when a living organism carries an infectious agent on its body (**mechanical**) or as an infection host itself (**biological**), to a new host.
- **Vehicle transmission** occurs when a substance, such as soil, water, or air, carries an infectious agent to a new host.
- **Healthcare-associated infections (HAI)**, or **nosocomial infections**, are acquired in a clinical setting. Transmission is facilitated by medical interventions and the high concentration of susceptible, immunocompromised individuals in clinical settings.

Footnotes

1. 1 Yves Thomas, Guido Vogel, Werner Wunderli, Patricia Suter, Mark Witschi, Daniel Koch, Caroline Tapparel, and Laurent Kaiser. "Survival of Influenza Virus on Banknotes." *Applied and Environmental Microbiology* 74, no. 10 (2008): 3002–3007.

2. 2 Filio Marineli, Gregory Tsoucalas, Marianna Karamanou, and George Androutsos. “Mary Mallon (1869–1938) and the History of Typhoid Fever.” *Annals of Gastroenterology* 26 (2013): 132–134. www.ncbi.nlm.nih.gov/pmc/arti...rol-26-132.pdf.
3. 3 World Health Organization. Fact sheet No. 391—*Drinking Water.* June 2005. www.who.int/mediacentre/factsheets/fs391/en.
4. 4 Blandine Massonnet-Bruneel, Nicole Corre-Catelin, Renaud Lacroix, Rosemary S. Lees, Kim Phuc Hoang, Derric Nimmo, Luke Alphey, and Paul Reiter. “Fitness of Transgenic Mosquito *Aedes aegypti* Males Carrying a Dominant Lethal Genetic System.” *PLOS ONE* 8, no. 5 (2013): e62711.
5. 5 Richard Levine. “Cases of Dengue Drop 91 Percent Due to Genetically Modified Mosquitoes.” *Entomology Today.* entomologytoday.org/2016/07/...ied-mosquitoes.
6. 6 Olivia Judson. “A Bug’s Death.” *The New York Times*, September 25, 2003. www.nytimes.com/2003/09/25/op...g-s-death.html.
7. 7 Gilda Grard, Mélanie Caron, Illich Manfred Mombo, Dieudonné Nkoghe, Statiana Mboui Ondo, Davy Jiolle, Didier Fontenille, Christophe Paupy, and Eric Maurice Leroy. “Zika Virus in Gabon (Central Africa)–2007: A New Threat from *Aedes albopictus*?” *PLOS Neglected Tropical Diseases* 8, no. 2 (2014): e2681.
8. 8 Constância F.J. Ayres. “Identification of Zika Virus Vectors and Implications for Control.” *The Lancet Infectious Diseases* 16, no. 3 (2016): 278–279.
9. 9 Centers for Disease Control and Prevention. “HAI Data and Statistics.” 2016. http://www.cdc.gov/hai/surveillance. Accessed Jan 2, 2016.

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18.4: Global Public Health

Learning Objectives

- Describe the entities involved in international public health and their activities
- Identify and differentiate between emerging and reemerging infectious diseases

A large number of international programs and agencies are involved in efforts to promote global public health. Among their goals are developing infrastructure in health care, public sanitation, and public health capacity; monitoring infectious disease occurrences around the world; coordinating communications between national public health agencies in various countries; and coordinating international responses to major health crises. In large part, these international efforts are necessary because disease-causing microorganisms know no national boundaries.

The World Health Organization (WHO)

International public health issues are coordinated by the **World Health Organization (WHO)**, an agency of the United Nations. Of its roughly $4 billion budget for 2015–16[1], about $1 billion was funded by member states and the remaining $3 billion by voluntary contributions. In addition to monitoring and reporting on infectious disease, WHO also develops and implements strategies for their control and prevention. WHO has had a number of successful international public health campaigns. For example, its vaccination program against smallpox, begun in the mid-1960s, resulted in the global eradication of the disease by 1980. WHO continues to be involved in infectious disease control, primarily in the developing world, with programs targeting malaria, HIV/AIDS, and tuberculosis, among others. It also runs programs to reduce illness and mortality that occur as a result of violence, accidents, lifestyle-associated illnesses such as diabetes, and poor health-care infrastructure.

WHO maintains a global alert and response system that coordinates information from member nations. In the event of a public health emergency or epidemic, it provides logistical support and coordinates international response to the emergency. The United States contributes to this effort through the CDC. The CDC carries out international monitoring and public health efforts, mainly in the service of protecting US public health in an increasingly connected world. Similarly, the European Union maintains a Health Security Committee that monitors disease outbreaks within its member countries and internationally, coordinating with WHO.

Emerging and Reemerging Infectious Diseases

Both WHO and some national public health agencies such as the CDC monitor and prepare for **emerging infectious diseases**. An emerging infectious disease is either new to the human population or has shown an increase in prevalence in the previous twenty years. Whether the disease is new or conditions have changed to cause an increase in frequency, its status as emerging implies the need to apply resources to understand and control its growing impact.

Emerging diseases may change their frequency gradually over time, or they may experience sudden epidemic growth. The importance of vigilance was made clear during the Ebola hemorrhagic fever epidemic in western Africa through 2014–2015. Although health experts had been aware of the Ebola virus since the 1970s, an outbreak on such a large scale had never happened before (Figure 18.4.1). Previous human epidemics had been small, isolated, and contained. Indeed, the gorilla and chimpanzee populations of western Africa had suffered far worse from Ebola than the human population. The pattern of small isolated human epidemics changed in 2014. Its high transmission rate, coupled with cultural practices for treatment of the dead and perhaps its emergence in an urban setting, caused the disease to spread rapidly, and thousands of people died. The international public health community responded with a large emergency effort to treat patients and contain the epidemic.

Emerging diseases are found in all countries, both developed and developing (Table 18.4.1). Some nations are better equipped to deal with them. National and international public health agencies watch for epidemics like the Ebola outbreak in developing countries because those countries rarely have the health-care infrastructure and expertise to deal with large outbreaks effectively. Even with the support of international agencies, the systems in western Africa struggled to identify and care for the sick and control spread. In addition to the altruistic goal of saving lives and assisting nations lacking in resources, the global nature of transportation

means that an outbreak anywhere can spread quickly to every corner of the planet. Managing an epidemic in one location—its source—is far easier than fighting it on many fronts.

Ebola is not the only disease that needs to be monitored in the global environment. In 2015, WHO set priorities on several emerging diseases that had a high probability of causing epidemics and that were poorly understood (and thus urgently required research and development efforts).

A **reemerging infectious disease** is a disease that is increasing in frequency after a previous period of decline. Its reemergence may be a result of changing conditions or old prevention regimes that are no longer working. Examples of such diseases are drug-resistant forms of tuberculosis, bacterial pneumonia, and malaria. Drug-resistant strains of the bacteria causing gonorrhea and syphilis are also becoming more widespread, raising concerns of untreatable infections.

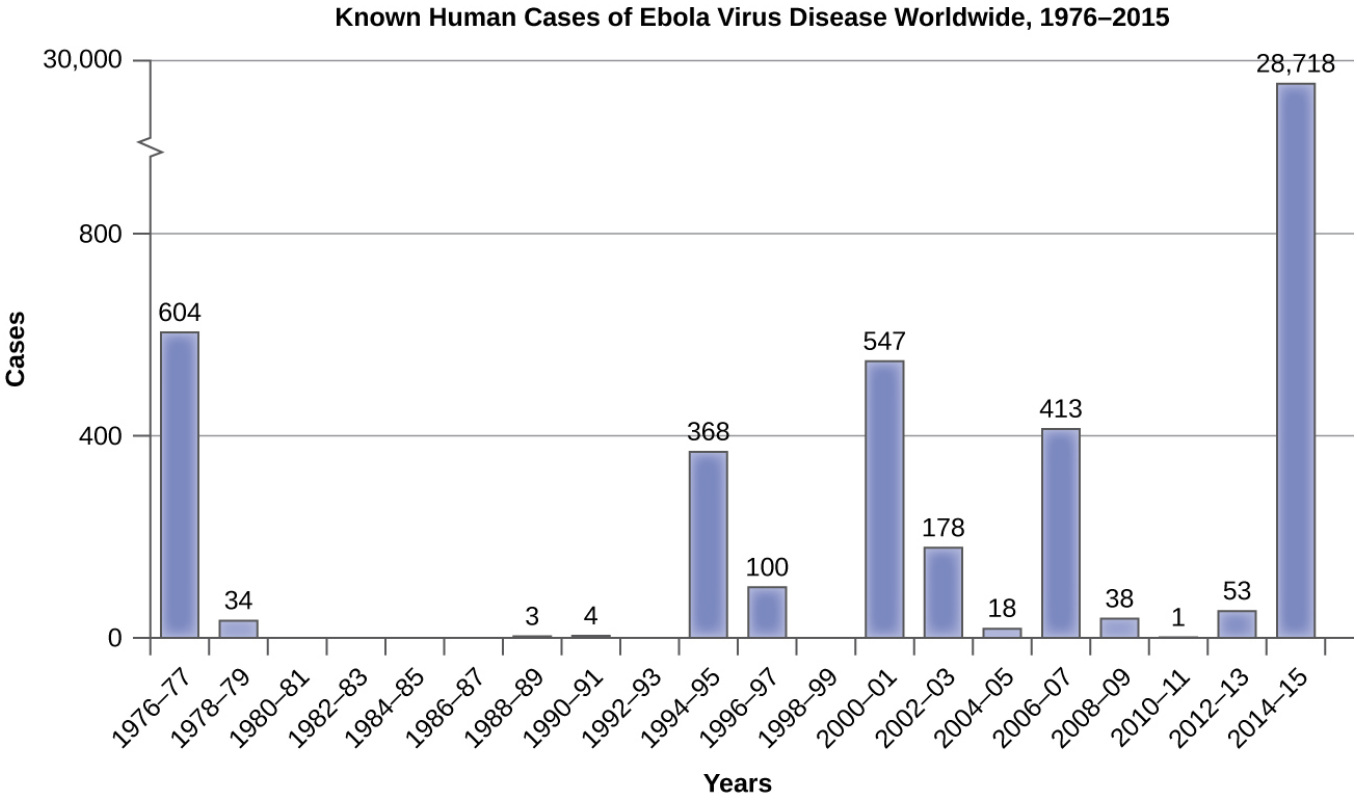


Figure 18.4.1: Even before the Ebola epidemic of 2014–15, Ebola was considered an emerging disease because of several smaller outbreaks between the mid-1990s and 2000s.

Table 18.4.1: Some Emerging and Reemerging Infectious Diseases

Disease	Pathogen	Year Discovered	Affected Regions	Transmission
AIDS	HIV	1981	Worldwide	Contact with infected body fluids
Chikungunya fever	Chikungunya virus	1952	Africa, Asia, India; spreading to Europe and the Americas	Mosquito-borne
Ebola virus disease	Ebola virus	1976	Central and Western Africa	Contact with infected body fluids
H1N1 Influenza (swine flu)	H1N1 virus	2009	Worldwide	Droplet transmission
Lyme disease	*Borrelia burgdorferi* bacterium	1981	Northern hemisphere	From mammal reservoirs to humans by tick vectors
West Nile virus disease	West Nile virus	1937	Africa, Australia, Canada to Venezuela, Europe, Middle East, Western Asia	Mosquito-borne

SARS Outbreak and Identification

On November 16, 2002, the first case of a SARS outbreak was reported in Guangdong Province, China. The patient exhibited influenza-like symptoms such as fever, cough, myalgia, sore throat, and shortness of breath. As the number of cases grew, the Chinese government was reluctant to openly communicate information about the epidemic with the World Health Organization (WHO) and the international community. The slow reaction of Chinese public health officials to this new disease contributed to the spread of the epidemic within and later outside China. In April 2003, the Chinese government finally responded with a huge public health effort involving quarantines, medical checkpoints, and massive cleaning projects. Over 18,000 people were quarantined in Beijing alone. Large funding initiatives were created to improve health-care facilities, and dedicated outbreak teams were created to coordinate the response. By August 16, 2003, the last SARS patients were released from a hospital in Beijing nine months after the first case was reported in China.

In the meantime, SARS spread to other countries on its way to becoming a global pandemic. Though the infectious agent had yet to be identified, it was thought to be an influenza virus. The disease was named SARS, an acronym for severe acute respiratory syndrome, until the etiologic agent could be identified. Travel restrictions to Southeast Asia were enforced by many countries. By the end of the outbreak, there were 8,098 cases and 774 deaths worldwide. China and Hong Kong were hit hardest by the epidemic, but Taiwan, Singapore, and Toronto, Canada, also saw significant numbers of cases (Figure 18.4.2).

Fortunately, timely public health responses in many countries effectively suppressed the outbreak and led to its eventual containment. For example, the disease was introduced to Canada in February 2003 by an infected traveler from Hong Kong, who died shortly after being hospitalized. By the end of March, hospital isolation and home quarantine procedures were in place in the Toronto area, stringent anti-infection protocols were introduced in hospitals, and the media were actively reporting on the disease. Public health officials tracked down contacts of infected individuals and quarantined them. A total of 25,000 individuals were quarantined in the city. Thanks to the vigorous response of the Canadian public health community, SARS was brought under control in Toronto by June, a mere four months after it was introduced.

In 2003, WHO established a collaborative effort to identify the causative agent of SARS, which has now been identified as a coronavirus that was associated with horseshoe bats. The genome of the SARS virus was sequenced and published by researchers at the CDC and in Canada in May 2003, and in the same month researchers in the Netherlands confirmed the etiology of the disease by fulfilling Koch's postulates for the SARS coronavirus. The last known case of SARS worldwide was reported in 2004.



Figure **18.4.2**: This map shows the spread of SARS as of March 28, 2003. (credit: modification of work by Central Intelligence Agency)

Link to Learning

This database of reports chronicles outbreaks of infectious disease around the world. It was on this system that the first information about the SARS outbreak in China emerged.

The CDC publishes *Emerging Infectious Diseases*, a monthly journal available online.

Key Concepts and Summary

- The **World Health Organization (WHO)** is an agency of the United Nations that collects and analyzes data on disease occurrence from member nations. WHO also coordinates public health programs and responses to international health emergencies.
- **Emerging diseases** are those that are new to human populations or that have been increasing in the past two decades. **Reemerging diseases** are those that are making a resurgence in susceptible populations after previously having been controlled in some geographic areas.

Footnotes

1. 1 World Health Organization. “Programme Budget 2014–2015.” www.who.int/about/finances-ac...lity/budget/en.

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18.E: Disease and Epidemiology (Exercises)

18.1: The Language of Epidemiologists

The field of epidemiology concerns the geographical distribution and timing of infectious disease occurrences and how they are transmitted and maintained in nature, with the goal of recognizing and controlling outbreaks. The science of epidemiology includes etiology (the study of the causes of disease) and investigation of disease transmission (mechanisms by which a disease is spread).

Matching

Match each term with its description.

___sporadic disease	A. the number of disease cases per 100,000 individuals
___endemic disease	B. a disease in higher than expected numbers around the world
___pandemic disease	C. the number of deaths from a disease for every 10,000 individuals
___morbidity rate	D. a disease found occasionally in a region with cases occurring mainly in isolation from each other
___mortality rate	E. a disease found regularly in a region

Answer

D, E, B, A, C

Fill in the Blank

The _____ collects data and conducts epidemiologic studies in the United States.

Answer

Centers for Disease Control and Prevention, or CDC

Short Answer

During an epidemic, why might the prevalence of a disease at a particular time not be equal to the sum of the incidences of the disease?

In what publication would you find data on emerging/reemerging diseases in the United States?

Critical Thinking

Why might an epidemiological population in a state not be the same size as the number of people in a state? Use an example.

18.2: Tracking Infectious Diseases

Some important researchers, such as Florence Nightingale, subscribed to the miasma hypothesis. The transition to acceptance of the germ theory during the 19th century provided a solid mechanistic grounding to the study of disease patterns. The studies of 19th century physicians and researchers such as John Snow, Florence Nightingale, Ignaz Semmelweis, Joseph Lister, Robert Koch, Louis Pasteur, and others sowed the seeds of modern epidemiology.

Matching

Match each type of epidemiology study with its description.

___experimental	A. examination of past case histories and medical test results conducted on patients in an outbreak
___analytical	B. examination of current case histories, interviews with patients and their contacts, interpretation of medical test results; frequently conducted while outbreak is still in progress
___prospective	C. use of a set of test subjects (human or animal) and control subjects that are treated the same as the test subjects except for the specific treatment being studied
___descriptive	D. observing groups of individuals to look for associations with disease
___retrospective	E. a comparison of a cohort of individuals through the course of the study

Answer

C, D, E, B, A

Match each pioneer of epidemiology with his or her contribution.

___Florence Nightingale	A. determined the source of a cholera outbreak in London
___Robert Koch	B. showed that surgical wound infection rates could be dramatically reduced by using carbolic acid to disinfect surgical tools, bandages, and surgical sites
___Joseph Lister	C. compiled data on causes of mortality in soldiers, leading to innovations in military medical care
___John Snow	D. developed a methodology for conclusively determining the etiology of disease

Answer

C, D, B, A

Fill in the Blank

_____occurs when an infected individual passes the infection on to other individuals, who pass it on to still others, increasing the penetration of the infection into the susceptible population.

Answer

Propagated spread

A batch of food contaminated with botulism exotoxin, consumed at a family reunion by most of the members of a family, would be an example of a _____ outbreak.

Answer

point source

Short Answer

What activity did John Snow conduct, other than mapping, that contemporary epidemiologists also use when trying to understand how to control a disease?

18.3: How Diseases Spread

Pathogens often have elaborate adaptations to exploit host biology, behavior, and ecology to live in and move between hosts. Hosts have evolved defenses against pathogens, but because their rates of evolution are typically slower than their pathogens (because their generation times are longer), hosts are usually at an evolutionary disadvantage. This section will explore where pathogens survive—both inside and outside hosts—and some of the many ways they move from one host to another.

Multiple Choice

Which is the most common type of biological vector of human disease?

A. viruses
B. bacteria
C. mammals
D. arthropods

Answer

D

A mosquito bites a person who subsequently develops a fever and abdominal rash. What type of transmission would this be?

A. mechanical vector transmission
B. biological vector transmission
C. direct contact transmission
D. vehicle transmission

Answer

B

Cattle are allowed to pasture in a field that contains the farmhouse well, and the farmer's family becomes ill with a gastrointestinal pathogen after drinking the water. What type of transmission of infectious agents would this be?

A. biological vector transmission
B. direct contact transmission
C. indirect contact transmission
D. vehicle transmission

Answer

D

A blanket from a child with chickenpox is likely to be contaminated with the virus that causes chickenpox (Varicella-zoster virus). What is the blanket called?

A. fomite
B. host
C. pathogen
D. vector

Answer

A

Fill in the Blank

A patient in the hospital with a urinary catheter develops a bladder infection. This is an example of a(n) _____ infection.

Answer

nosocomial or healthcare-associated

A _____ is an animal that can transfer infectious pathogens from one host to another.

Answer

vector

Short Answer

Differentiate between droplet vehicle transmission and airborne transmission.

Critical Thinking

Many people find that they become ill with a cold after traveling by airplane. The air circulation systems of commercial aircraft use HEPA filters that should remove any infectious agents that pass through them. What are the possible reasons for increased incidence of colds after flights?

18.4: Global Public Health

A large number of international programs and agencies are involved in efforts to promote global public health. Among their goals are developing infrastructure in health care, public sanitation, and public health capacity; monitoring infectious disease occurrences around the world; coordinating communications between national public health agencies in various countries; and coordinating international responses to major health crises.

Multiple Choice

Which of the following would NOT be considered an emerging disease?

A. Ebola hemorrhagic fever
B. West Nile virus fever/encephalitis
C. Zika virus disease
D. Tuberculosis

Answer

D

Which of the following would NOT be considered a reemerging disease?

A. Drug-resistant tuberculosis
B. Drug-resistant gonorrhea
C. Malaria
D. West Nile virus fever/encephalitis

Answer

D

Which of the following factors can lead to reemergence of a disease?

A. A mutation that allows it to infect humans
B. A period of decline in vaccination rates
C. A change in disease reporting procedures
D. Better education on the signs and symptoms of the disease

Answer

B

Why are emerging diseases with very few cases the focus of intense scrutiny?

A. They tend to be more deadly
B. They are increasing and therefore not controlled
C. They naturally have higher transmission rates
D. They occur more in developed countries

Answer

B

Fill in the Blank

The _____ collects data and conducts epidemiologic studies at the global level.

Answer

WHO (World Health Organization)

Critical Thinking

An Atlantic crossing by boat from England to New England took 60–80 days in the 18th century. In the late 19th century the voyage took less than a week. How do you think these time differences for travel might have impacted the spread of infectious diseases from Europe to the Americas, or vice versa?

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CHAPTER OVERVIEW

19: Epidemiological Studies and Public Health

Epidemiology is the study and analysis of the distribution (who, when, and where) and determinants of health and disease conditions in defined populations.

19.1: Descriptive Epidemiology
19.2: Analytical Epidemiology
19.3: Experimental Epidemiology
19.4: Public Health Measures for Disease Control
19.5: Global Health
19.6: Emerging and Reemerging Infectious Diseases
19.7: Biological Weapons
19.8: Technology and New Infectious Agents
19.9: Current Epidemics

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19.1: Descriptive Epidemiology

20.01: Descriptive Epidemiology

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19.2: Analytical Epidemiology

20.02: Analytical Epidemiology

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19.3: Experimental Epidemiology

20.03: Experimental Epidemiology

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19.4: Public Health Measures for Disease Control

20.04: Public Health Measures for Disease Control

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19.5: Global Health

20.05: Global Health

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19.6: Emerging and Reemerging Infectious Diseases

20.06: Emerging and Reemerging Infectious Diseases

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19.7: Biological Weapons

20.07: Biological Weapons

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19.8: Technology and New Infectious Agents

20.08: Technology and New Infectious Agents

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19.9: Current Epidemics

20.09: Current Epidemics

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CHAPTER OVERVIEW

20: Innate Immunity 1

Despite relatively constant exposure to pathogenic microbes in the environment, humans do not generally suffer from constant infection or disease. Under most circumstances, the body is able to defend itself from the threat of infection thanks to a complex immune system designed to repel, kill, and expel disease-causing invaders. Immunity as a whole can be described as two interrelated parts: nonspecific innate immunity, which is the subject of this chapter, and specific adaptive host defenses, which are discussed in the next chapter.

The nonspecific innate immune response provides a first line of defense that can often prevent infections from gaining a solid foothold in the body. These defenses are described as *nonspecific* because they do not target any specific pathogen; rather, they defend against a wide range of potential pathogens. They are called *innate* because they are built-in mechanisms of the human organism. Unlike the specific adaptive defenses, they are not acquired over time and they have no “memory” (they do not improve after repeated exposures to specific pathogens).

Broadly speaking, nonspecific innate defenses provide an immediate (or very rapid) response against potential pathogens. However, these responses are neither perfect nor impenetrable. They can be circumvented by pathogens on occasion, and sometimes they can even cause damage to the body, contributing to the signs and symptoms of infection.

20.1: Physical Defenses Overview

20.2: Physical Barreirs - Skin and Mucous Membranes

20.3: Chemical Defenses - Overview

20.4: Chemical Defenses - Antimicrobial Peptides (AMP) and Plasma Protein Mediators

20.5: Chemical Defenses - Complement System

20.6: Chemical Defenses - Cytokines

20.E: Innate Immune Defenses I (Exercises)

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20.1: Physical Defenses Overview

Learning Objectives

- Describe the various physical barriers and mechanical defenses that protect the human body against infection and disease
- Describe the role of microbiota as a first-line defense against infection and disease

Nonspecific innate immunity can be characterized as a multifaceted system of defenses that targets invading pathogens in a nonspecific manner. In this chapter, we have divided the numerous defenses that make up this system into three categories: **physical defenses**, **chemical defenses**, and **cellular defenses**. However, it is important to keep in mind that these defenses do not function independently, and the categories often overlap. Table **20.1.1** provides an overview of the nonspecific defenses discussed in this chapter.

Table **20.1.1**: Overview of Nonspecific Innate Immune Defenses

Overview of Nonspecific Innate Immune Defenses	
Physical defenses	Physical barriers
	Mechanical defenses
	Microbiome
Chemical defenses	Chemicals and enzymes in body fluids
	Antimicrobial peptides
	Plasma protein mediators
	Cytokines
	Inflammation-eliciting mediators
Cellular defenses	Granulocytes
	Agranulocytes

Physical defenses provide the body's most basic form of nonspecific defense. They include physical barriers to microbes, such as the skin and mucous membranes, as well as mechanical defenses that physically remove microbes and debris from areas of the body where they might cause harm or infection. In addition, the microbiome provides a measure of physical protection against disease, as microbes of the normal microbiota compete with pathogens for nutrients and cellular binding sites necessary to cause infection.

Key Concepts and Summary

- **Nonspecific innate immunity** provides a first line of defense against infection by nonspecifically blocking entry of microbes and targeting them for destruction or removal from the body.
- The physical defenses of innate immunity include physical barriers, mechanical actions that remove microbes and debris, and the microbiome, which competes with and inhibits the growth of pathogens.
- The skin, mucous membranes, and endothelia throughout the body serve as physical barriers that prevent microbes from reaching potential sites of infection. Tight cell junctions in these tissues prevent microbes from passing through.
- Microbes trapped in dead skin cells or **mucus** are removed from the body by mechanical actions such as shedding of skin cells, mucociliary sweeping, coughing, **peristalsis**, and flushing of bodily fluids (e.g., urination, tears)
- The resident microbiota provide a physical defense by occupying available cellular binding sites and competing with pathogens for available nutrients.

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20.2: Physical Barreirs - Skin and Mucous Membranes

Learning Objectives

- Summarize cellular junctions in physical barriers
- Describe how skin acts as a barrier to protect us from infections from the outside world
- Evaluate the roles of mucous membranes in protecting against infections from inside of the body
- Identify the importance of endothelia in CNS immune defense

Physical Barriers

Physical barriers play an important role in preventing microbes from reaching tissues that are susceptible to infection. At the cellular level, barriers consist of cells that are tightly joined to prevent invaders from crossing through to deeper tissue. For example, the endothelial cells that line blood vessels have very tight cell-to-cell junctions, blocking microbes from gaining access to the bloodstream.

Cell junctions are generally composed of cell membrane proteins that may connect with the extracellular matrix or with complementary proteins from neighboring cells. Tissues in various parts of the body have different types of cell junctions. These include **tight junctions, desmosomes, and gap junctions**, as illustrated in Figure 20.2.1. Invading microorganisms may attempt to break down these substances chemically, using enzymes such as proteases that can cause structural damage to create a point of entry for pathogens.

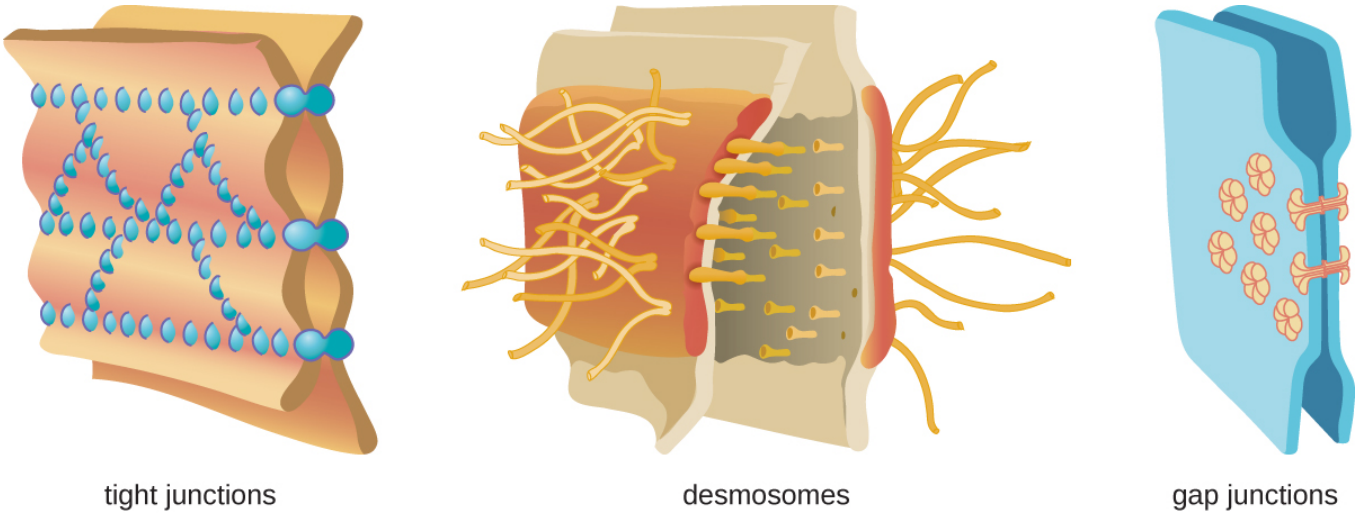


Figure 20.2.1: There are multiple types of cell junctions in human tissue, three of which are shown here. Tight junctions rivet two adjacent cells together, preventing or limiting material exchange through the spaces between them. Desmosomes have intermediate fibers that act like shoelaces, tying two cells together, allowing small materials to pass through the resulting spaces. Gap junctions are channels between two cells that permit their communication via signals. (credit: modification of work by Mariana Ruiz Villareal)

The Skin Barrier

One of the body's most important physical barriers is the **skin barrier**, which is composed of three layers of closely packed cells. The thin upper layer is called the **epidermis**. A second, thicker layer, called the **dermis**, contains hair follicles, sweat glands, nerves, and blood vessels. A layer of fatty tissue called the **hypodermis** lies beneath the dermis and contains blood and lymph vessels (Figure 20.2.2).

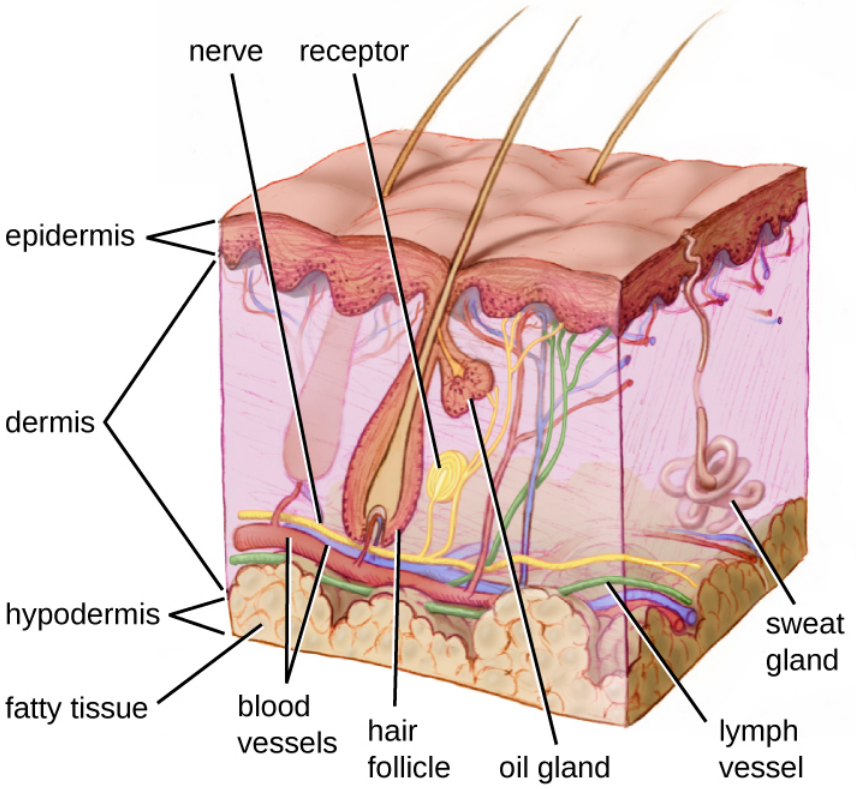


Figure 20.2.2: Human skin has three layers, the epidermis, the dermis, and the hypodermis, which provide a thick barrier between microbes outside the body and deeper tissues. Dead skin cells on the surface of the epidermis are continually shed, taking with them microbes on the skin's surface. (credit: modification of work by National Institutes of Health)

The topmost layer of skin, the epidermis, consists of cells that are packed with **keratin**. These dead cells remain as a tightly connected, dense layer of protein-filled cell husks on the surface of the skin. The keratin makes the skin's surface mechanically tough and resistant to degradation by bacterial enzymes. **Fatty acids** on the skin's surface create a dry, salty, and acidic environment that inhibits the growth of some microbes and is highly resistant to breakdown by bacterial enzymes. In addition, the **dead cells** of the epidermis are frequently shed, along with any microbes that may be clinging to them. Shed skin cells are continually replaced with new cells from below, providing a new barrier that will soon be shed in the same way.

Infections can occur when the skin barrier is compromised or broken. A wound can serve as a point of entry for opportunistic pathogens, which can infect the skin tissue surrounding the wound and possibly spread to deeper tissues.

Mucous Membranes

The **mucous membranes** lining the nose, mouth, lungs, and urinary and digestive tracts provide another nonspecific barrier against potential pathogens. Mucous membranes consist of a layer of epithelial cells bound by tight junctions. The epithelial cells secrete a moist, sticky substance called **mucus**, which covers and protects the more fragile cell layers beneath it and traps debris and particulate matter, including microbes. Mucus secretions also contain **antimicrobial peptides**.

In many regions of the body, mechanical actions serve to flush mucus (along with trapped or dead microbes) out of the body or away from potential sites of infection. For example, in the respiratory system, inhalation can bring microbes, dust, mold spores, and other small airborne debris into the body. This debris becomes trapped in the mucus lining the respiratory tract, a layer known as the mucociliary blanket. The epithelial cells lining the upper parts of the respiratory tract are called ciliated epithelial cells because they have hair-like appendages known as cilia. Movement of the cilia propels debris-laden mucus out and away from the lungs. The expelled mucus is then swallowed and destroyed in the stomach, or coughed up, or sneezed out (Figure 20.2.4). This system of removal is often called the **mucociliary escalator**.

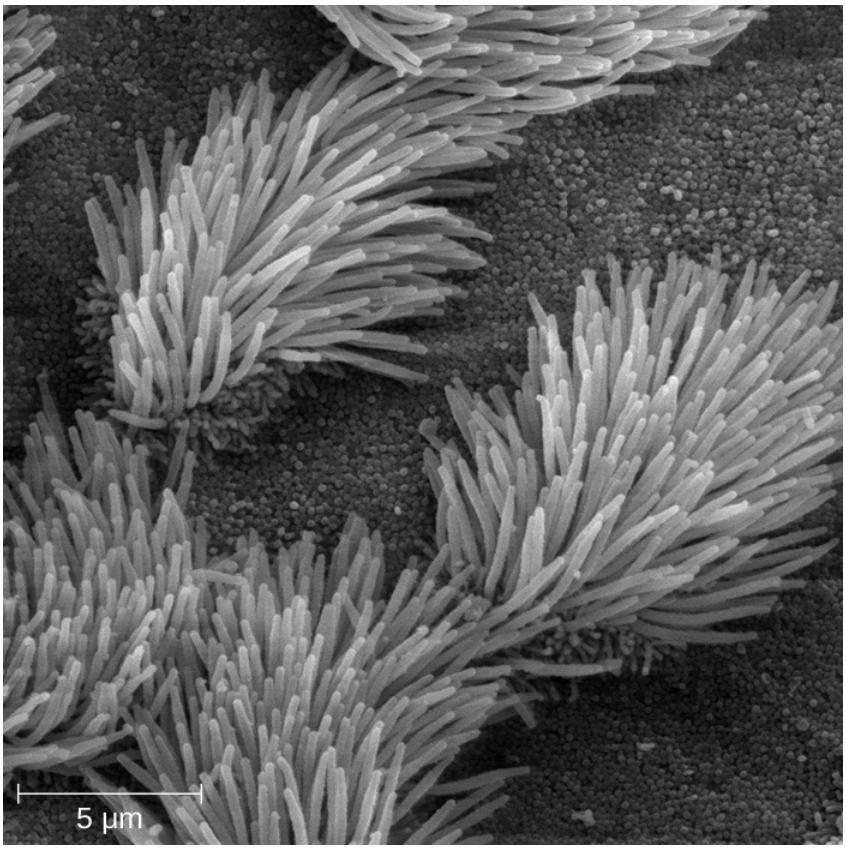


Figure 20.2.4: This scanning electron micrograph shows ciliated and nonciliated epithelial cells from the human trachea. The mucociliary escalator pushes mucus away from the lungs, along with any debris or microorganisms that may be trapped in the sticky mucus, and the mucus moves up to the esophagus where it can be removed by swallowing.

The mucociliary escalator is such an effective barrier to microbes that the lungs, the lowermost (and most sensitive) portion of the respiratory tract, were long considered to be a sterile environment in healthy individuals. Only recently has research suggested that healthy lungs may have a small normal microbiota. Disruption of the mucociliary escalator by the damaging effects of smoking or diseases such as cystic fibrosis can lead to increased colonization of bacteria in the lower respiratory tract and frequent infections, which highlights the importance of this physical barrier to host defenses.

Like the respiratory tract, the digestive tract is a portal of entry through which microbes enter the body, and the mucous membranes lining the digestive tract provide a nonspecific physical barrier against ingested microbes. The intestinal tract is lined with epithelial cells, interspersed with mucus-secreting **goblet cells** (Figure 20.2.5). This mucus mixes with material received from the stomach, trapping foodborne microbes and debris. The mechanical action of **peristalsis**, a series of muscular contractions in the digestive tract, moves the sloughed mucus and other material through the intestines, rectum, and anus, excreting the material in feces.

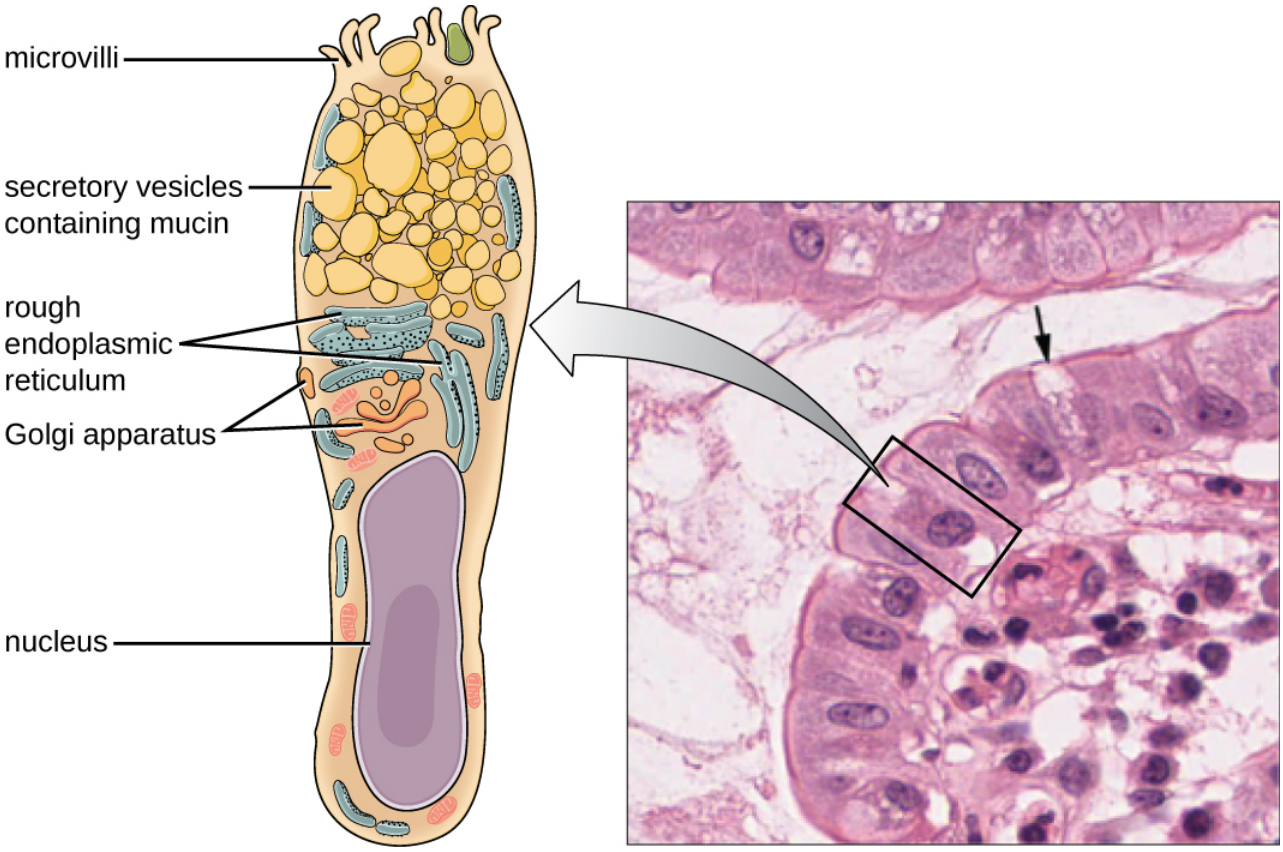


Figure **20.2.5**: Goblet cells produce and secrete mucus. The arrows in this micrograph point to the mucus-secreting goblet cells (magnification 1600×) in the intestinal epithelium. (credit micrograph: Micrograph provided by the Regents of University of Michigan Medical School © 2012)

Endothelia

The **epithelial cells** lining the urogenital tract, blood vessels, lymphatic vessels, and certain other tissues are known as endothelia. These tightly packed cells provide a particularly effective frontline barrier against invaders. The endothelia of the **blood-brain barrier**, for example, protect the central nervous system (CNS), which consists of the brain and the spinal cord. The CNS is one of the most sensitive and important areas of the body, as microbial infection of the CNS can quickly lead to serious and often fatal inflammation. The cell junctions in the blood vessels traveling through the CNS are some of the tightest and toughest in the body, preventing any transient microbes in the bloodstream from entering the CNS. This keeps the cerebrospinal fluid that surrounds and bathes the brain and spinal cord sterile under normal conditions.

Key Concepts and Summary

- The physical defenses of innate immunity include physical barriers, mechanical actions that remove microbes and debris, and the microbiome, which competes with and inhibits the growth of pathogens.
- The skin, mucous membranes, and endothelia throughout the body serve as physical barriers that prevent microbes from reaching potential sites of infection. Tight cell junctions in these tissues prevent microbes from passing through.
- Microbes trapped in dead skin cells or **mucus** are removed from the body by mechanical actions such as shedding of skin cells, mucociliary sweeping, coughing, **peristalsis**, and flushing of bodily fluids (e.g., urination, tears)

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20.3: Chemical Defenses - Overview

Learning Objectives

- Describe how enzymes in body fluids provide protection against infection or disease
- Recognize the chemical defense mechanisms in several organ systems

In addition to physical defenses, the innate nonspecific immune system uses a number of **chemical mediators** that inhibit microbial invaders. The term “chemical mediators” encompasses a wide array of substances found in various body fluids and tissues throughout the body. Chemical mediators may work alone or in conjunction with each other to inhibit microbial colonization and infection.

Some chemical mediators are **endogenously** produced, meaning they are produced by human body cells; others are produced **exogenously**, meaning that they are produced by certain microbes that are part of the microbiome. Some mediators are produced continually, bathing the area in the antimicrobial substance; others are produced or activated primarily in response to some stimulus, such as the presence of microbes.

Chemical and Enzymatic Mediators Found in Body Fluids

Fluids produced by the skin include examples of both endogenous and exogenous mediators. **Sebaceous glands** in the dermis secrete an oil called sebum that is released onto the skin surface through hair follicles. This **sebum** is an **endogenous mediator**, providing an additional layer of defense by helping seal off the pore of the hair follicle, preventing bacteria on the skin’s surface from invading sweat glands and surrounding tissue (Figure 20.3.1). Certain members of the **microbiome**, such as the bacterium *Propionibacterium acnes* and the fungus *Malassezia*, among others, can use lipase enzymes to degrade sebum, using it as a food source. This produces oleic acid, which creates a mildly acidic environment on the surface of the skin that is inhospitable to many pathogenic microbes. **Oleic acid** is an example of an **exogenously produced mediator** because it is produced by resident microbes and not directly by body cells.

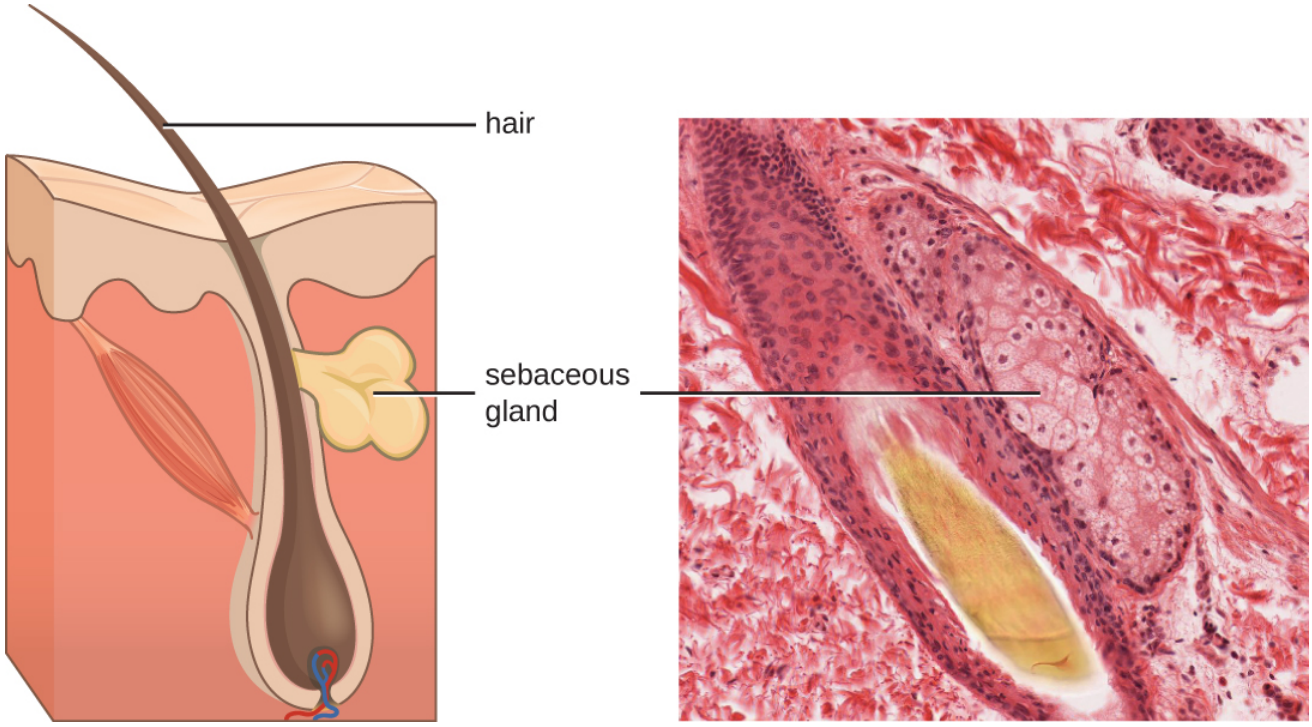


Figure 20.3.1: Sebaceous glands secrete sebum, a chemical mediator that lubricates and protect the skin from invading microbes. Sebum is also a food source for resident microbes that produce oleic acid, an exogenously produced mediator. (credit micrograph: Micrograph provided by the Regents of University of Michigan Medical School © 2012)

Environmental factors that affect the microbiota of the skin can have a direct impact on the production of chemical mediators. Low humidity or decreased sebum production, for example, could make the skin less habitable for microbes that produce oleic acid, thus making the skin more susceptible to pathogens normally inhibited by the skin's low pH. Many skin moisturizers are formulated to counter such effects by restoring moisture and essential oils to the skin.

Chemical Defense Mechanisms in Organ Systems

The **digestive tract** also produces a large number of chemical mediators that inhibit or kill microbes. In the oral cavity, **saliva** contains mediators such as lactoperoxidase enzymes, and **mucus** secreted by the esophagus contains the antibacterial enzyme lysozyme. In the stomach, highly **acidic gastric fluid** kills most microbes. In the lower digestive tract, the intestines have pancreatic and intestinal **enzymes, antibacterial peptides (cryptins), bile** produced from the liver, and specialized Paneth cells that produce lysozyme. Together, these mediators are able to eliminate most pathogens that manage to survive the acidic environment of the stomach.

In the **urinary tract**, urine flushes microbes out of the body during urination. Furthermore, the slight acidity of urine (the average pH is about 6) inhibits the growth of many microbes and potential pathogens in the urinary tract.

The female **reproductive system** employs **lactate**, an exogenously produced chemical mediator, to inhibit microbial growth. The cells and tissue layers composing the vagina produce glycogen, a branched and more complex polymer of glucose. Lactobacilli in the area ferment glycogen to produce lactate, lowering the pH in the vagina and inhibiting transient microbiota, opportunistic pathogens like *Candida* (a yeast associated with vaginal infections), and other pathogens responsible for sexually transmitted diseases.

In the **eyes**, **tears** contain the chemical mediators lysozyme and lactoferrin, both of which are capable of eliminating microbes that have found their way to the surface of the eyes. Lysozyme cleaves the bond between NAG and NAM in peptidoglycan, a component of the cell wall in bacteria. It is more effective against gram-positive bacteria, which lack the protective outer membrane associated with gram-negative bacteria. Lactoferrin inhibits microbial growth by chemically binding and sequestering iron. This effectually starves many microbes that require iron for growth.

In the **ears**, **cerumen (earwax)** exhibits antimicrobial properties due to the presence of fatty acids, which lower the pH to between 3 and 5.

The **respiratory tract** uses various chemical mediators in the nasal passages, trachea, and lungs. The mucus produced in the nasal passages contains a mix of antimicrobial molecules similar to those found in tears and saliva (e.g., lysozyme, lactoferrin, lactoperoxidase). Secretions in the trachea and lungs also contain lysozyme and lactoferrin, as well as a diverse group of additional chemical mediators, such as the lipoprotein complex called surfactant, which has antibacterial properties.

Key Concepts and Summary

- Numerous **chemical mediators** produced endogenously and exogenously exhibit nonspecific antimicrobial functions.
- Many chemical mediators are found in body fluids such as sebum, saliva, mucus, gastric and intestinal fluids, urine, tears, cerumen, and vaginal secretions.

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20.4: Chemical Defenses - Antimicrobial Peptides (AMP) and Plasma Protein Mediators

Learning Objectives

- Describe the functions of antimicrobial peptides (AMPs)
- Distinguish between defensins and bacteriocins

Antimicrobial Peptides (AMPs)

The **antimicrobial peptides (AMPs)** are a special class of nonspecific cell-derived mediators with **broad-spectrum** antimicrobial properties. Some AMPs are produced routinely by the body, whereas others are primarily produced (or produced in greater quantities) in response to the presence of an invading pathogen. Research has begun exploring how AMPs can be used in the diagnosis and treatment of disease.

Functions of AMPs

AMPs may induce cell damage in microorganisms in a variety of ways, including by **inflicting damage to membranes**, **destroying DNA and RNA**, or **interfering with cell-wall synthesis**. Depending on the specific antimicrobial mechanism, a particular AMP may inhibit only certain groups of microbes (e.g., gram-positive or gram-negative bacteria) or it may be more broadly effective against bacteria, fungi, protozoa, and viruses. Many AMPs are found on the skin, but they can also be found in other regions of the body.

Defensins versus Bacteriocins

A family of AMPs called **defensins** can be produced by epithelial cells throughout the body as well as by cellular defenses such as macrophages and neutrophils (see Cellular Defenses). Defensins may be secreted or act inside host cells; they combat microorganisms by damaging their plasma membranes. AMPs called **bacteriocins** are produced exogenously by certain members of the resident microbiota within the gastrointestinal tract. The genes coding for these types of AMPs are often carried on plasmids and can be passed between different species within the resident microbiota through lateral or horizontal gene transfer.

There are numerous other AMPs throughout the body. The characteristics of a few of the more significant AMPs are summarized in Table 20.4.1.

Table 20.4.1: Characteristics of Selected Antimicrobial Peptides (AMPs)

AMP	Secreted by	Body site	Pathogens inhibited	Mode of action
Bacteriocins	Resident microbiota	Gastrointestinal tract	Bacteria	Disrupt membrane
Cathelicidin	Epithelial cells, macrophages, and other cell types	Skin	Bacteria and fungi	Disrupts membrane
Defensins	Epithelial cells, macrophages, neutrophils	Throughout the body	Fungi, bacteria, and many viruses	Disrupt membrane
Dermicidin	Sweat glands	Skin	Bacteria and fungi	Disrupts membrane integrity and ion channels
Histatins	Salivary glands	Oral cavity	Fungi	Disrupt intracellular function

Plasma Protein Mediators

Many nonspecific innate immune factors are found in plasma, the fluid portion of blood. **Plasma** contains electrolytes, sugars, lipids, and proteins, each of which helps to maintain homeostasis (i.e., stable internal body functioning), and contains the proteins involved in the clotting of blood. Additional proteins found in blood plasma, such as acute-phase proteins, complement proteins, and cytokines, are involved in the nonspecific innate immune response.

Acute-Phase Proteins

The acute-phase proteins are another class of antimicrobial mediators. **Acute-phase proteins** are primarily produced in the **liver** and secreted into the blood in response to inflammatory molecules from the immune system. Examples of acute-phase proteins include C-reactive protein, serum amyloid A, ferritin, transferrin, fibrinogen, and mannose-binding lectin. Each of these proteins has a different chemical structure and inhibits or destroys microbes in some way (Table **20.4.1**).

Table **20.4.2**: Some Acute-Phase Proteins and Their Functions

Some Acute-Phase Proteins and Their Functions	
C-reactive protein	Coats bacteria (opsonization), preparing them for ingestion by phagocytes
Serum amyloid A	
Ferritin	Bind and sequester iron, thereby inhibiting the growth of pathogens
Transferrin	
Fibrinogen	Involved in formation of blood clots that trap bacterial pathogens
Mannose-binding lectin	Activates complement cascade

Key Concepts and Summary

- **Antimicrobial peptides (AMPs)** found on the skin and in other areas of the body are largely produced in response to the presence of pathogens. These include dermcidin, cathelicidin, defensins, histatins, and bacteriocins.

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20.5: Chemical Defenses - Complement System

Learning Objectives

- Identify the three mechanisms that trigger complement activation
- Describe the outcome of complement activation

The Complement System

The **complement system** is a group of plasma protein mediators that can act as an innate nonspecific defense while also serving to connect innate and adaptive immunity (discussed in the next chapter). The complement system is composed of more than 30 proteins (including C1 through C9) that normally circulate as precursor proteins in blood. These precursor proteins become activated when stimulated or triggered by a variety of factors, including the presence of microorganisms. Complement proteins are considered part of innate nonspecific immunity because they are always present in the blood and tissue fluids, allowing them to be activated quickly. Also, when activated through the alternative pathway (described later in this section), complement proteins target pathogens in a nonspecific manner.

Complement Activation

The process by which circulating complement precursors become functional is called **complement activation**. This process is a cascade that can be triggered by one of **three different mechanisms**, known as the alternative, classical, and lectin pathways.

The **alternative pathway** is initiated by the spontaneous activation of the complement protein C3. The hydrolysis of C3 produces two products, C3a and C3b. When no invader microbes are present, C3b is very quickly degraded in a hydrolysis reaction using the water in the blood. However, if invading microbes are present, C3b attaches to the surface of these microbes. Once attached, C3b will recruit other complement proteins in a cascade (Figure **20.5.2**).

The **classical pathway** provides a more efficient mechanism of activating the complement cascade, but it depends upon the production of antibodies by the specific adaptive immune defenses. To initiate the classical pathway, a specific antibody must first bind to the pathogen to form an **antibody-antigen complex**. This activates the first protein in the complement cascade, the C1 complex. The C1 complex is a multipart protein complex, and each component participates in the full activation of the overall complex. Following recruitment and activation of the C1 complex, the remaining classical pathway complement proteins are recruited and activated in a cascading sequence (Figure **20.5.2**).

The **lectin activation pathway** is similar to the classical pathway, but it is triggered by the binding of **mannose-binding lectin**, an acute-phase protein, to carbohydrates on the microbial surface. Like other acute-phase proteins, lectins are produced by liver cells and are commonly upregulated in response to inflammatory signals received by the body during an infection (Figure **20.5.2**).

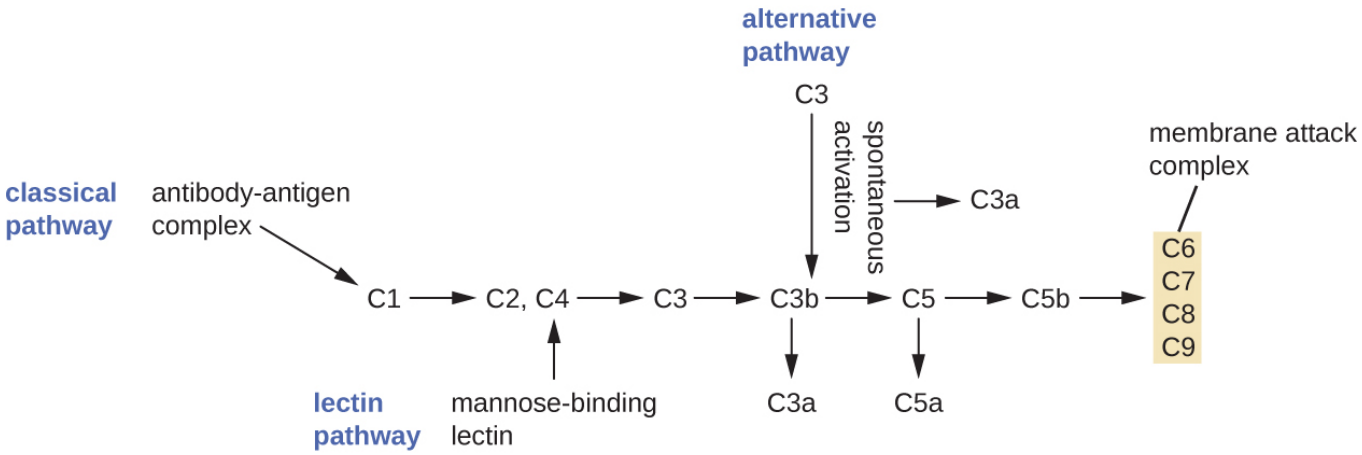


Figure 20.5.2: The three complement activation pathways have different triggers, as shown here, but all three result in the activation of the complement protein C3, which produces C3a and C3b. The latter binds to the surface of the target cell and then works with other complement proteins to cleave C5 into C5a and C5b. C5b also binds to the cell surface and then recruits C6 through C9; these molecules form a ring structure called the membrane attack complex (MAC), which punches through the cell membrane of the invading pathogen, causing it to swell and burst.

Outcome of Complement Activation

Although each complement activation pathway is initiated in a different way, they all provide the same protective outcomes: **opsonization, inflammation, chemotaxis, and cytolysis**. The term opsonization refers to the coating of a pathogen by a chemical substance (called an opsonin) that allows phagocytic cells to recognize, engulf, and destroy it more easily. Opsonins from the complement cascade include C1q, C3b, and C4b. Additional important opsonins include mannose-binding proteins and antibodies. The complement fragments C3a and C5a are well-characterized anaphylatoxins with potent proinflammatory functions. Anaphylatoxins activate mast cells, causing degranulation and the release of inflammatory chemical signals, including mediators that cause vasodilation and increased vascular permeability. C5a is also one of the most potent chemoattractants for neutrophils and other white blood cells, cellular defenses that will be discussed in the next section.

The complement proteins C6, C7, C8, and C9 assemble into a **membrane attack complex (MAC)**, which allows C9 to polymerize into pores in the membranes of gram-negative bacteria. These pores allow water, ions, and other molecules to move freely in and out of the targeted cells, eventually leading to cell lysis and death of the pathogen (Figure 20.5.2). However, the MAC is only effective against gram-negative bacteria; it cannot penetrate the thick layer of peptidoglycan associated with cell walls of gram-positive bacteria. Since the MAC does not pose a lethal threat to gram-positive bacterial pathogens, complement-mediated opsonization is more important for their clearance.

Key Concepts and Summary

- The **complement system** involves numerous precursor proteins that circulate in plasma. These proteins become activated in a cascading sequence in the presence of microbes, resulting in the **opsonization** of pathogens, chemoattraction of leukocytes, induction of inflammation, and cytolysis through the formation of a **membrane attack complex (MAC)**.

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20.6: Chemical Defenses - Cytokines

Learning Objectives

- Distinguish the three classes of cytokines
- Describe the actions of inflammation-eliciting mediators

Cytokines

Cytokines are soluble proteins that act as communication signals between cells. In a nonspecific innate immune response, various cytokines may be released to stimulate production of chemical mediators or other cell functions, such as cell proliferation, cell differentiation, inhibition of cell division, apoptosis, and chemotaxis.

When a cytokine binds to its target receptor, the effect can vary widely depending on the type of cytokine and the type of cell or receptor to which it has bound. The function of a particular cytokine can be described as autocrine, paracrine, or endocrine (Figure 20.6.3). In autocrine function, the same cell that releases the cytokine is the recipient of the signal; in other words, autocrine function is a form of self-stimulation by a cell. In contrast, paracrine function involves the release of cytokines from one cell to other nearby cells, stimulating some response from the recipient cells. Last, endocrine function occurs when cells release cytokines into the bloodstream to be carried to target cells much farther away.

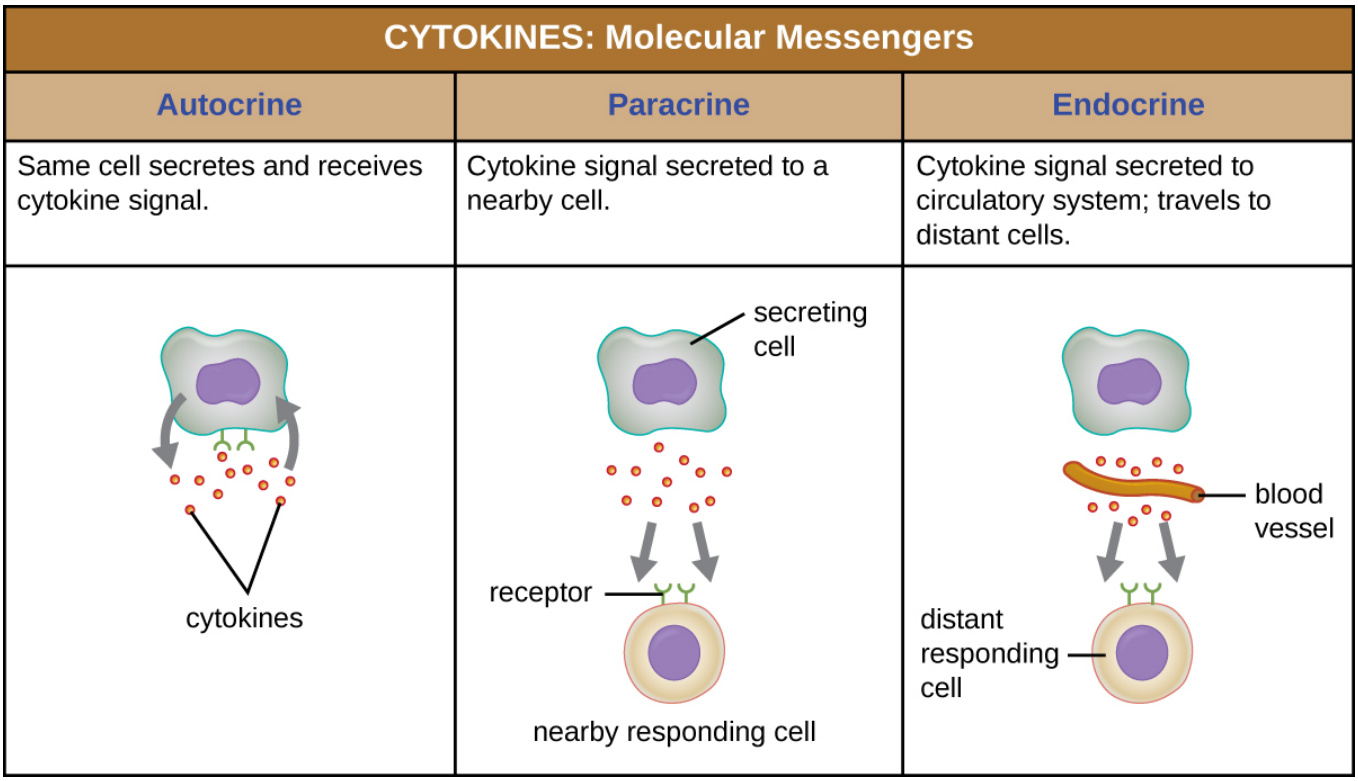


Figure 20.6.3: Autocrine, paracrine, and endocrine actions describe which cells are targeted by cytokines and how far the cytokines must travel to bind to their intended target cells' receptors.

Three important classes of cytokines are the interleukins, chemokines, and interferons. The **interleukins** were originally thought to be produced only by leukocytes (white blood cells) and to only stimulate leukocytes, thus the reasons for their name. Although interleukins are involved in modulating almost every function of the immune system, their role in the body is not restricted to immunity. Interleukins are also produced by and stimulate a variety of cells unrelated to immune defenses.

The **chemokines** are chemotactic factors that recruit leukocytes to sites of infection, tissue damage, and inflammation. In contrast to more general chemotactic factors, like complement factor C5a, chemokines are very specific in the subsets of leukocytes they recruit.

Interferons are a diverse group of immune signaling molecules and are especially important in our defense against viruses. Type I interferons (interferon-α and interferon-β) are produced and released by cells infected with virus. These interferons stimulate nearby cells to stop production of mRNA, destroy RNA already produced, and reduce protein synthesis. These cellular changes inhibit viral replication and production of mature virus, slowing the spread of the virus. Type I interferons also stimulate various immune cells involved in viral clearance to more aggressively attack virus-infected cells. Type II interferon (interferon-γ) is an important activator of immune cells (Figure **20.6.4**).

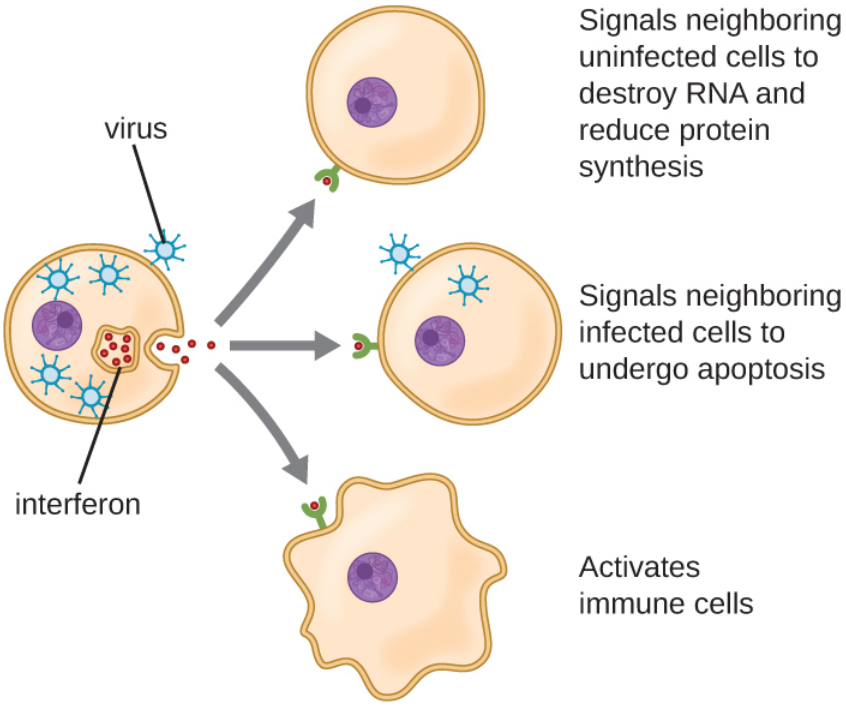


Figure **20.6.4**: Interferons are cytokines released by a cell infected with a virus. Interferon-α and interferon-β signal uninfected neighboring cells to inhibit mRNA synthesis, destroy RNA, and reduce protein synthesis (top arrow). Interferon-α and interferon-β also promote apoptosis in cells infected with the virus (middle arrow). Interferon-γ alerts neighboring immune cells to an attack (bottom arrow). Although interferons do not cure the cell releasing them or other infected cells, which will soon die, their release may prevent additional cells from becoming infected, thus stemming the infection.

Inflammation-Eliciting Mediators

Many of the chemical mediators discussed in this section contribute in some way to inflammation and fever, which are nonspecific immune responses discussed in more detail in Inflammation and Fever. Cytokines stimulate the production of acute-phase proteins such as C-reactive protein and mannose-binding lectin in the liver. These acute-phase proteins act as opsonins, activating complement cascades through the lectin pathway.

Some cytokines also bind mast cells and basophils, inducing them to release histamine, a proinflammatory compound. Histamine receptors are found on a variety of cells and mediate proinflammatory events, such as bronchoconstriction (tightening of the airways) and smooth muscle contraction.

In addition to **histamine**, mast cells may release other chemical mediators, such as leukotrienes. **Leukotrienes** are lipid-based proinflammatory mediators that are produced from the metabolism of arachidonic acid in the cell membrane of leukocytes and tissue cells. Compared with the proinflammatory effects of histamine, those of leukotrienes are more potent and longer lasting. Together, these chemical mediators can induce coughing, vomiting, and diarrhea, which serve to expel pathogens from the body.

Certain cytokines also stimulate the production of **prostaglandins**, chemical mediators that promote the inflammatory effects of kinins and histamines. Prostaglandins can also help to set the body temperature higher, leading to fever, which promotes the activities of white blood cells and slightly inhibits the growth of pathogenic microbes (see Inflammation and Fever).

Another inflammatory mediator, **bradykinin**, contributes to edema, which occurs when fluids and leukocytes leak out of the bloodstream and into tissues. It binds to receptors on cells in the capillary walls, causing the capillaries to dilate and become more permeable to fluids.

Table **20.6.3** provides a summary of the chemical defenses discussed in this section.

Table **20.6.3**: Chemical Defenses of Nonspecific Innate Immunity

Defense	Examples	Function
Chemicals and enzymes in body fluids	Sebum from sebaceous glands	Provides oil barrier protecting hair follicle pores from pathogens
	Oleic acid from sebum and skin microbiota	Lowers pH to inhibit pathogens
	Lysozyme in secretions	Kills bacteria by attacking cell wall
	Acid in stomach, urine, and vagina	Inhibits or kills bacteria
	Digestive enzymes and bile	Kill bacteria
	Lactoferrin and transferrin	Bind and sequester iron, inhibiting bacterial growth
	Surfactant in lungs	Kills bacteria
Antimicrobial peptides	Defensins, bacteriocins, dermicidin, cathelicidin, histatins,	Kill bacteria by attacking membranes or interfering with cell functions
Plasma protein mediators	Acute-phase proteins (C-reactive protein, serum amyloid A, ferritin, fibrinogen, transferrin, and mannose-binding lectin)	Inhibit the growth of bacteria and assist in the trapping and killing of bacteria
	Complements C3b and C4b	Opsonization of pathogens to aid phagocytosis
	Complement C5a	Chemoattractant for phagocytes
	Complements C3a and C5a	Proinflammatory anaphylatoxins
Cytokines	Interleukins	Stimulate and modulate most functions of immune system
	Chemokines	Recruit white blood cells to infected area
	Interferons	Alert cells to viral infection, induce apoptosis of virus-infected cells, induce antiviral defenses in infected and nearby uninfected cells, stimulate immune cells to attack virus-infected cells
Inflammation-eliciting mediators	Histamine	Promotes vasodilation, bronchoconstriction, smooth muscle contraction, increased secretion and mucus production
	Leukotrienes	Promote inflammation; stronger and longer lasting than histamine
	Prostaglandins	Promote inflammation and fever
	Bradykinin	Increases vasodilation and vascular permeability, leading to edema

Key Concepts and Summary

- **Cytokines** are proteins that facilitate various nonspecific responses by innate immune cells, including production of other chemical mediators, cell proliferation, cell death, and differentiation.
- Cytokines play a key role in the inflammatory response, triggering production of inflammation-eliciting mediators such as acute-phase proteins, **histamine**, leukotrienes, **prostaglandins**, and **bradykinin**.

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20.E: Innate Immune Defenses I (Exercises)

20.1: Physical Defenses

Nonspecific innate immunity provides a first line of defense against infection by nonspecifically blocking entry of microbes and targeting them for destruction or removal from the body. The physical defenses of innate immunity include physical barriers, mechanical actions that remove microbes and debris, and the microbiome, which competes with and inhibits the growth of pathogens. The skin, mucous membranes, and endothelia throughout the body serve as physical barriers.

Multiple Choice

Which of the following best describes the innate nonspecific immune system?

A. a targeted and highly specific response to a single pathogen or molecule
B. a generalized and nonspecific set of defenses against a class or group of pathogens
C. a set of barrier mechanisms that adapts to specific pathogens after repeated exposure
D. the production of antibody molecules against pathogens

Answer

B

Which of the following constantly sheds dead cells along with any microbes that may be attached to those cells?

A. epidermis
B. dermis
C. hypodermis
D. mucous membrane

Answer

A

Which of the following uses a particularly dense suite of tight junctions to prevent microbes from entering the underlying tissue?

A. the mucociliary escalator
B. the epidermis
C. the blood-brain barrier
D. the urethra

Answer

C

Fill in the Blank

The muscular contraction of the intestines that results in movement of material through the digestive tract is called _____.

Answer

peristalsis

_____ are the hair-like appendages of cells lining parts of the respiratory tract that sweep debris away from the lungs.

Answer

cilia

Secretions that bathe and moisten the interior of the intestines are produced by _____ cells.

Answer

goblet

Short Answer

Differentiate a physical barrier from a mechanical removal mechanism and give an example of each.

Identify some ways that pathogens can breach the physical barriers of the innate immune system.

20.2: Chemical Defenses

Numerous chemical mediators produced endogenously and exogenously exhibit nonspecific antimicrobial functions. Many chemical mediators are found in body fluids such as sebum, saliva, mucus, gastric and intestinal fluids, urine, tears, cerumen, and vaginal secretions. Antimicrobial peptides (AMPs) found on the skin and in other areas of the body are largely produced in response to the presence of pathogens. These include dermcidin, cathelicidin, defensins, histatins, and bacteriocins.

Multiple Choice

Which of the following serve as chemical signals between cells and stimulate a wide range of nonspecific defenses?

A. cytokines
B. antimicrobial peptides
C. complement proteins
D. antibodies

Answer

A

Bacteriocins and defensins are types of which of the following?

A. leukotrienes
B. cytokines
C. inflammation-eliciting mediators
D. antimicrobial peptides

Answer

D

Which of the following chemical mediators is secreted onto the surface of the skin?

A. cerumen
B. sebum
C. gastric acid
D. prostaglandin

Answer

B

Identify the complement activation pathway that is triggered by the binding of an acute-phase protein to a pathogen.

A. classical
B. alternate
C. lectin
D. cathelicidin

Answer

C

Histamine, leukotrienes, prostaglandins, and bradykinin are examples of which of the following?

A. chemical mediators primarily found in the digestive system
B. chemical mediators that promote inflammation
C. antimicrobial peptides found on the skin
D. complement proteins that form MACs

Answer

B

Fill in the Blank

_____ are antimicrobial peptides produced by members of the normal microbiota.

Answer

bacteriocins

_____ is the fluid portion of a blood sample that has been drawn in the presence of an anticoagulant compound.

Answer

plasma

The process by which cells are drawn or attracted to an area by a microbe invader is known as _____.

Answer

chemotaxis

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CHAPTER OVERVIEW

21: Innate Immunity II

Despite relatively constant exposure to pathogenic microbes in the environment, humans do not generally suffer from constant infection or disease. Under most circumstances, the body is able to defend itself from the threat of infection thanks to a complex immune system designed to repel, kill, and expel disease-causing invaders. Immunity as a whole can be described as two interrelated parts: nonspecific innate immunity, which is the subject of this chapter, and specific adaptive host defenses, which are discussed in the next chapter.

The nonspecific innate immune response provides a first line of defense that can often prevent infections from gaining a solid foothold in the body. These defenses are described as *nonspecific* because they do not target any specific pathogen; rather, they defend against a wide range of potential pathogens. They are called *innate* because they are built-in mechanisms of the human organism. Unlike the specific adaptive defenses, they are not acquired over time and they have no “memory” (they do not improve after repeated exposures to specific pathogens).

Broadly speaking, nonspecific innate defenses provide an immediate (or very rapid) response against potential pathogens. However, these responses are neither perfect nor impenetrable. They can be circumvented by pathogens on occasion, and sometimes they can even cause damage to the body, contributing to the signs and symptoms of infection.

21.1: Cellular Defenses - Overview
21.2: Cellular Defenses - Granulocytes
21.3: Cellular Defenses - Agranulocytes
21.4: Pathogen Recognition
21.5: Phagocytosis
21.6: Inflammation
21.7: Fever
21.E: Innate Immune Defenses II (Exercises)

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21.1: Cellular Defenses - Overview

Learning Objectives

- Identify and describe the components of blood
- Explain the process by which the formed elements of blood are formed (hematopoiesis)
- Describe the characteristics of formed elements found in peripheral blood, as well as their respective functions within the innate immune system

In the previous section, we discussed some of the chemical mediators found in plasma, the fluid portion of blood. The nonfluid portion of blood consists of various types of **formed elements**, so called because they are all formed from the same stem cells found in bone marrow. **The three major categories of formed elements are: red blood cells (RBCs), also called erythrocytes; platelets, also called thrombocytes; and white blood cells (WBCs), also called leukocytes.**

Red blood cells are primarily responsible for carrying oxygen to tissues. Platelets are cellular fragments that participate in blood clot formation and tissue repair. Several different types of WBCs participate in various nonspecific mechanisms of innate and adaptive immunity. In this section, we will focus primarily on the innate mechanisms of various types of WBCs.

Hematopoiesis

All of the formed elements of blood are derived from **hematopoietic stem cells (HSCs)** in the bone marrow. As the HSCs make copies of themselves in the bone marrow, individual cells receive different cues from the body that control how they develop and mature. As a result, the HSCs differentiate into different types of blood cells that, once mature, circulate in peripheral blood. This process of differentiation, called hematopoiesis, is shown in more detail in Figure **21.1.1**.

In terms of sheer numbers, the vast majority of HSCs become erythrocytes. Much smaller numbers become leukocytes and platelets. Leukocytes can be further subdivided into granulocytes, which are characterized by numerous granules visible in the cytoplasm, and agranulocytes, which lack granules. Figure **21.1.2** provides an overview of the various types of formed elements, including their relative numbers, primary function, and lifespans.

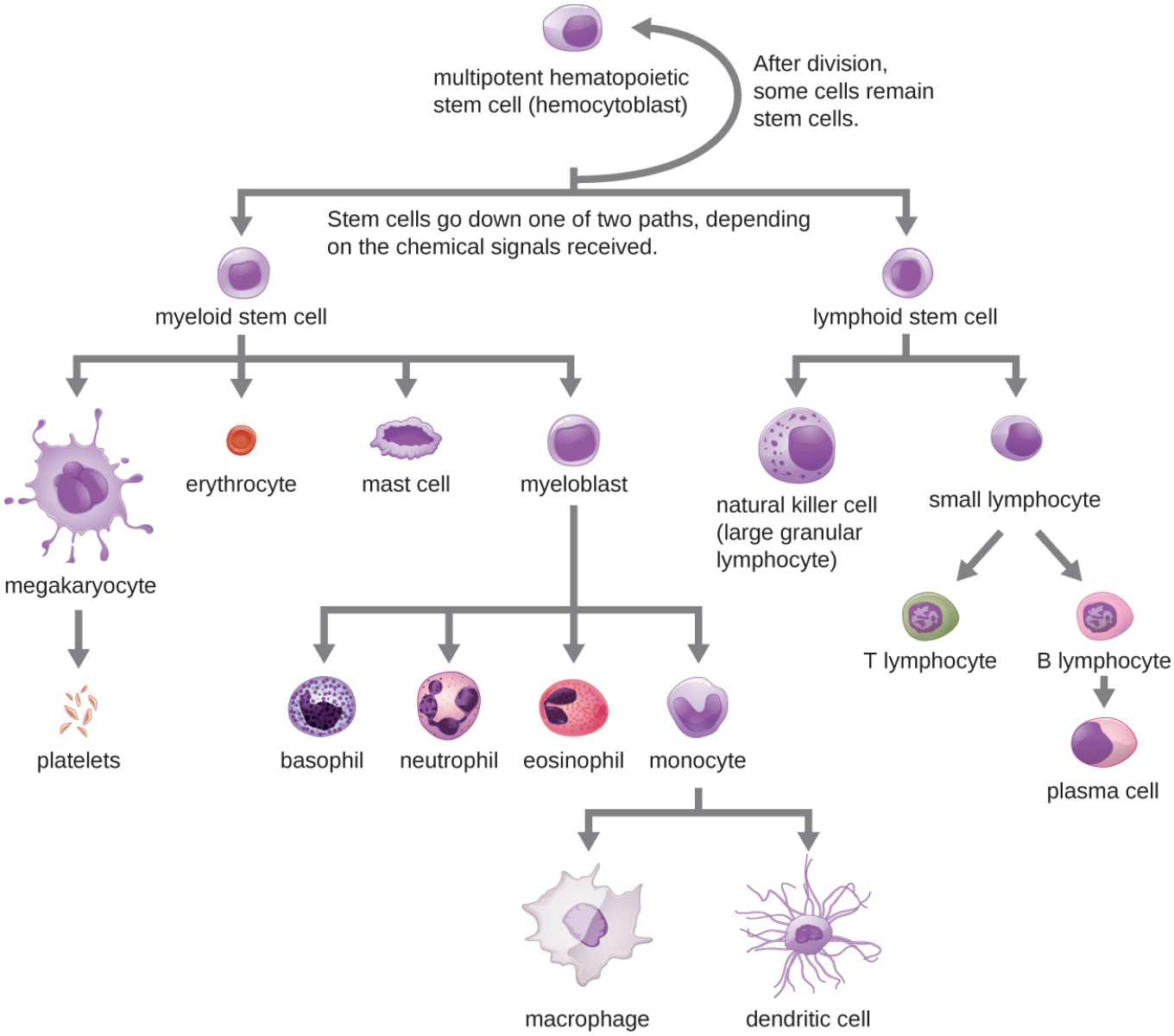


Figure **21.1.1**: All the formed elements of the blood arise by differentiation of hematopoietic stem cells in the bone marrow.

Formed Element	Major Subtypes		Numbers Present per Microliter (μL) and Mean (Range)	Appearance in a Standard Blood Smear	Summary of Functions	Comments
Erythrocytes (red blood cells) 			5.2 million (4.4–6.0 million)	Flattened biconcave disk; no nucleus; pale red	Transport oxygen and some carbon dioxide between tissue and lungs	Lifespan of approximately 120 days
Leukocytes (white blood cells)			7000 (5000–10,000)	Obvious dark-staining nucleus	All function in body defenses	Exit capillaries and move into tissues; lifespan of usually a few hours or days
	Granulocytes, including neutrophils, eosinophils, and basophils	***Total leukocytes (%)***	4360 (1800–9950)	Abundant granules in cytoplasm; nucleus normally lobed	Nonspecific (innate) resistance to disease	Classified according to membrane-bound granules in cytoplasm
	Neutrophils 	50–70	4150 (1800–7300)	Nucleus lobes increase with age; pale lilac granules	Phagocytic; particularly effective against bacteria; release cytotoxic chemicals from granules	Most common leukocyte; lifespan of minutes to days
	Eosinophils 	1–3	165 (0–700)	Nucleus generally two-lobed; bright red-orange granules	Phagocytic cells; particularly effective with antigen-antibody complexes; release antihistamines; combat parasitic infections	Lifespan of minutes to days
	Basophils 	<1	44 (0–150)	Nucleus generally two-lobed but difficult to see due to presence of heavy, dense, dark purple granules	Pro-inflammatory	Least common leukocyte; lifespan unknown
	Agranulocytes, including lymphocytes and monocytes		2640 (1700–4950)	Lack abundant granules in cytoplasm; have a simple-shaped nucleus that may be indented	Body defenses	Group consists of two major cell types from different lineages
	Lymphocytes 	20–40	2185 (1500–4000)	Spherical cells with a single, often large, nucleus occupying much of the cell's volume; stains purple; seen in large (natural killer cells) and small (B and T cells) variants	Primarily specific (adaptive) immunity: T cells directly attack other cells (cellular immunity); B cells release antibodies (humoral immunity); natural killer cells are similar to T cells but nonspecific	Initial cells originate in bone marrow, but secondary production occurs in lymphatic tissue; several distinct subtypes; memory cells form after exposure to a pathogen and rapidly increase responses to subsequent exposure; lifespan of many years
	Monocytes 	1–6	455 (200–950)	Largest leukocyte; has an indented or horseshoe-shaped nucleus	Very effective phagocytic cells engulfing pathogens or worn-out cells; also serve as antigen-presenting cells (APCs) or other components of the immune system	Produced in red bone marrow; referred to as macrophages and dendritic cells after leaving the circulation
Platelets 			350,000 (150,000–500,000)	Cellular fragments surrounded by a plasma membrane and containing granules; stains purple	Hemostasis; release growth factors for repair and healing of tissue	Formed from megakaryocytes that remain in the red bone marrow and shed platelets into circulation

Figure **21.1.2**: Formed elements of blood include erythrocytes (red blood cells), leukocytes (white blood cells), and platelets.

Key Concepts and Summary

- The **formed elements** of the blood include red blood cells (**erythrocytes**), white blood cells (**leukocytes**), and **platelets (thrombocytes**). Of these, leukocytes are primarily involved in the immune response.
- All formed elements originate in the bone marrow as stem cells (HSCs) that differentiate through **hematopoiesis**.

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21.2: Cellular Defenses - Granulocytes

Learning Objectives

- Distinguish the morphology and function of different granulocytes
- Name three antimicrobial mechanisms of neutrophils (PMNs)

Granulocytes

The various types of granulocytes can be distinguished from one another in a blood smear by the appearance of their nuclei and the contents of their granules, which confer different traits, functions, and staining properties. The **neutrophils**, also called **polymorphonuclear neutrophils (PMNs)**, have a nucleus with three to five lobes and small, numerous, lilac-colored granules. Each lobe of the nucleus is connected by a thin strand of material to the other lobes. The **eosinophils** have fewer lobes in the nucleus (typically 2–3) and larger granules that stain reddish-orange. The **basophils** have a two-lobed nucleus and large granules that stain dark blue or purple (Figure **21.2.3**).

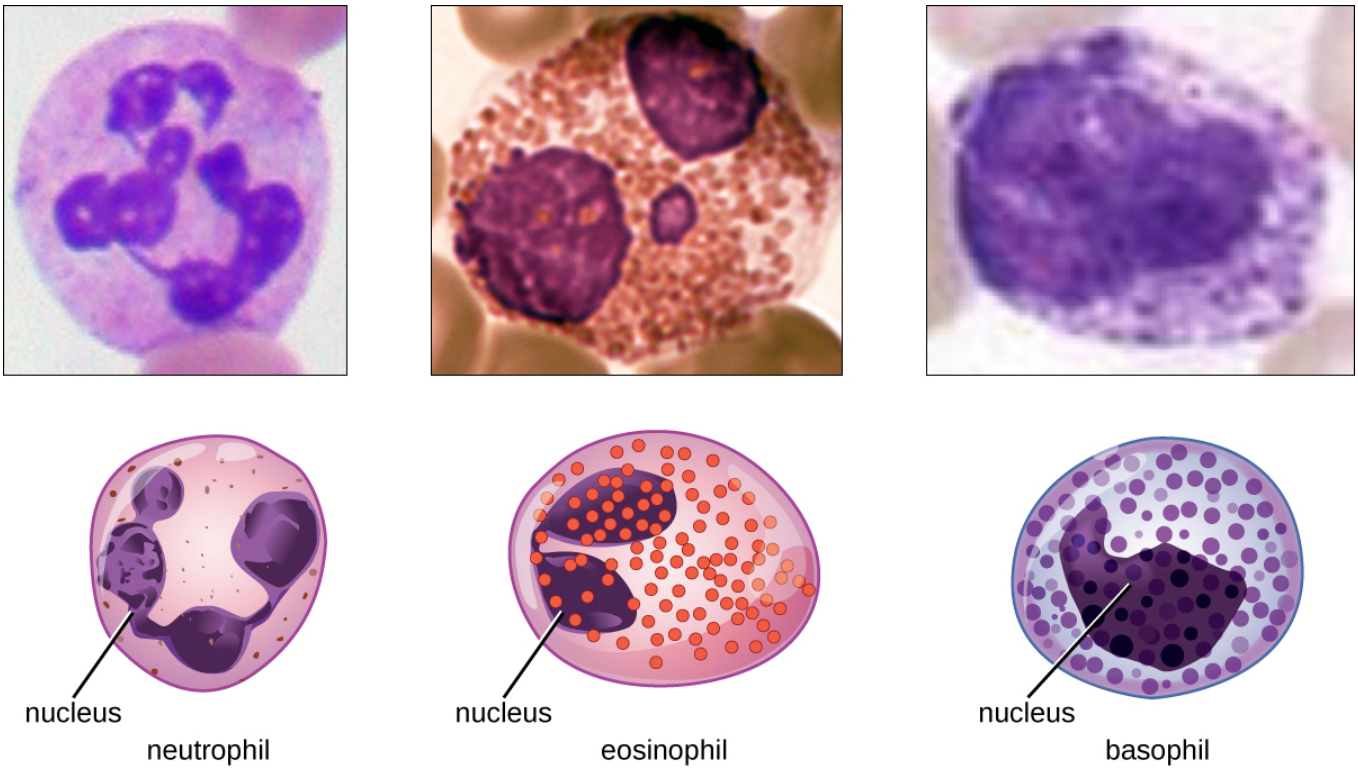


Figure **21.2.3**: Granulocytes can be distinguished by the number of lobes in their nuclei and the staining properties of their granules. (credit "neutrophil" micrograph: modification of work by Ed Uthman)

Neutrophils (PMNs)

Neutrophils (PMNs) are frequently involved in the elimination and destruction of extracellular bacteria. They are capable of migrating through the walls of blood vessels to areas of bacterial infection and tissue damage, where they seek out and kill infectious bacteria. **PMN granules contain a variety of defensins and hydrolytic enzymes** that help them destroy bacteria through phagocytosis (described in more detail in Pathogen Recognition and Phagocytosis) In addition, when many neutrophils are brought into an infected area, they can be stimulated to release toxic molecules into the surrounding tissue to better clear infectious agents. This is called degranulation.

Another mechanism used by neutrophils is **neutrophil extracellular traps (NETs)**, which are extruded meshes of chromatin that are closely associated with antimicrobial granule proteins and components. Chromatin is DNA with associated proteins (usually histone proteins, around which DNA wraps for organization and packing within a cell). By creating and releasing a mesh or lattice-like structure of chromatin that is coupled with antimicrobial proteins, the neutrophils can mount a highly concentrated and

efficient attack against nearby pathogens. Proteins frequently associated with NETs include lactoferrin, gelatinase, cathepsin G, and myeloperoxidase. Each has a different means of promoting antimicrobial activity, helping neutrophils eliminate pathogens. The toxic proteins in NETs may kill some of the body's own cells along with invading pathogens. However, this collateral damage can be repaired after the danger of the infection has been eliminated.

As neutrophils fight an infection, a visible accumulation of leukocytes, cellular debris, and bacteria at the site of infection can be observed. This buildup is what we call **pus** (also known as purulent or suppurative discharge or drainage). The presence of pus is a sign that the immune defenses have been activated against an infection; historically, some physicians believed that inducing pus formation could actually promote the healing of wounds. The practice of promoting "laudable pus" (by, for instance, wrapping a wound in greasy wool soaked in wine) dates back to the ancient physician Galen in the 2nd century AD, and was practiced in variant forms until the 17th century (though it was not universally accepted). Today, this method is no longer practiced because we now know that it is not effective. Although a small amount of pus formation can indicate a strong immune response, artificially inducing pus formation does not promote recovery.

Eosinophils

Eosinophils are granulocytes that protect against protozoa and helminths; they also play a role in allergic reactions. The granules of eosinophils, which readily absorb the acidic reddish dye eosin, contain histamine, degradative enzymes, and a compound known as major basic protein (MBP) (Figure 21.2.3). MBP binds to the surface carbohydrates of parasites, and this binding is associated with disruption of the cell membrane and membrane permeability.

Basophils

Basophils have cytoplasmic granules of varied size and are named for their granules' ability to absorb the basic dye methylene blue (Figure 21.2.3). Their stimulation and degranulation can result from multiple triggering events. Activated complement fragments C3a and C5a, produced in the activation cascades of complement proteins, act as anaphylatoxins by inducing degranulation of basophils and inflammatory responses. This cell type is important in allergic reactions and other responses that involve inflammation. One of the most abundant components of basophil granules is histamine, which is released along with other chemical factors when the basophil is stimulated. These chemicals can be chemotactic and can help to open the gaps between cells in the blood vessels. Other mechanisms for basophil triggering require the assistance of antibodies, as discussed in B Lymphocytes and Humoral Immunity.

Mast Cells

Hematopoiesis also gives rise to mast cells, which appear to be derived from the same common myeloid progenitor cell as neutrophils, eosinophils, and basophils. Functionally, mast cells are very similar to basophils, containing many of the same components in their granules (e.g., histamine) and playing a similar role in allergic responses and other inflammatory reactions. However, unlike basophils, mast cells leave the circulating blood and are most frequently found residing in tissues. They are often associated with blood vessels and nerves or found close to surfaces that interface with the external environment, such as the skin and mucous membranes in various regions of the body (Figure 21.2.4).

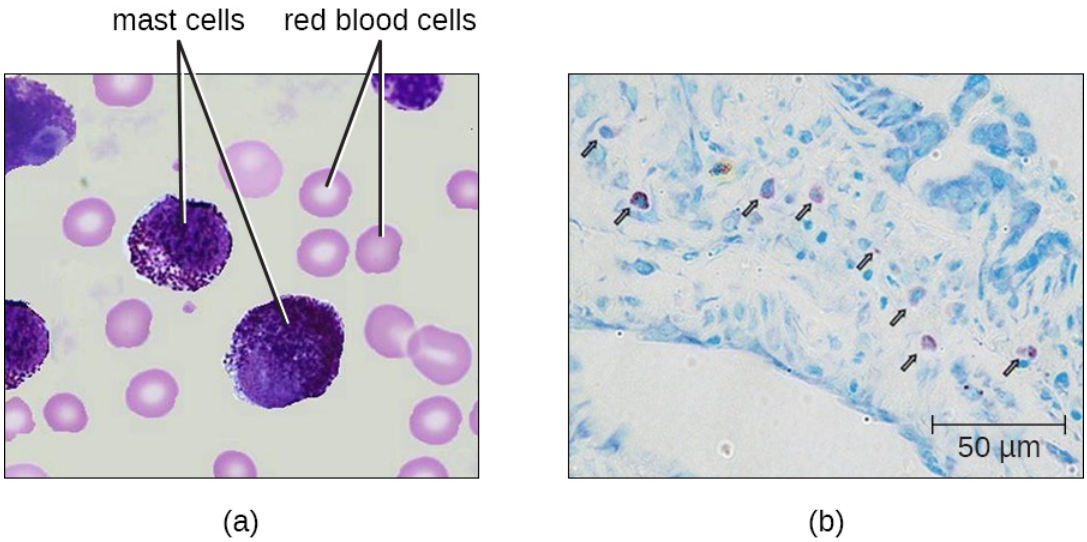


Figure **21.2.4**: Mast cells function similarly to basophils by inducing and promoting inflammatory responses. (a) This figure shows mast cells in blood. In a blood smear, they are difficult to differentiate from basophils (b). Unlike basophils, mast cells migrate from the blood into various tissues. (credit right: modification of work by Greenland JR, Xu X, Sayah DM, Liu FC, Jones KD, Looney MR, Caughey GH)

Key Concepts and Summary

- **Granulocytes** are leukocytes characterized by a lobed nucleus and granules in the cytoplasm. These include **neutrophils (PMNs)**, **eosinophils**, and **basophils**.
- Neutrophils are the leukocytes found in the largest numbers in the bloodstream and they primarily fight bacterial infections.
- Eosinophils target parasitic infections. Eosinophils and basophils are involved in allergic reactions. Both release histamine and other proinflammatory compounds from their granules upon stimulation.
- **Mast cells** function similarly to basophils but can be found in tissues outside the bloodstream.

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21.3: Cellular Defenses - Agranulocytes

Learning Objectives

- Describe the signals that activate natural killer cells
- Identify the mechanisms that NK cells use to kill target cells
- Differentiate monocytes from macrophages

Agranulocytes

As their name suggests, **agranulocytes** lack visible granules in the cytoplasm. **Agranulocytes can be categorized as lymphocytes or monocytes** (Figure **21.3.2**). Among the lymphocytes are natural killer cells, which play an important role in nonspecific innate immune defenses. Lymphocytes also include the B cells and T cells, which are discussed in the next chapter because they are central players in the specific adaptive immune defenses. The monocytes differentiate into macrophages and dendritic cells, which are collectively referred to as the mononuclear phagocyte system.

Natural Killer Cells

Most lymphocytes are primarily involved in the specific adaptive immune response, and thus will be discussed in the following chapter. An exception is the **natural killer cells (NK cells)**; these mononuclear lymphocytes use nonspecific mechanisms to recognize and destroy cells that are abnormal in some way. Cancer cells and cells infected with viruses are two examples of cellular abnormalities that are targeted by NK cells. Recognition of such cells involves a complex process of identifying inhibitory and activating molecular markers on the surface of the target cell. Molecular markers that make up the major histocompatibility complex (MHC) are expressed by healthy cells as an indication of “self.” This will be covered in more detail in next chapter. NK cells are able to recognize normal MHC markers on the surface of healthy cells, and these MHC markers serve as an inhibitory signal preventing NK cell activation. However, cancer cells and virus-infected cells actively diminish or eliminate expression of MHC markers on their surface. When these MHC markers are diminished or absent, the NK cell interprets this as an abnormality and a cell in distress. This is one part of the NK cell activation process (Figure **21.3.5**). NK cells are also activated by binding to activating molecular molecules on the target cell. These activating molecular molecules include “altered self” or “nonself” molecules. When a NK cell recognizes a decrease in inhibitory normal MHC molecules and an increase in activating molecules on the surface of a cell, the NK cell will be activated to eliminate the cell in distress.

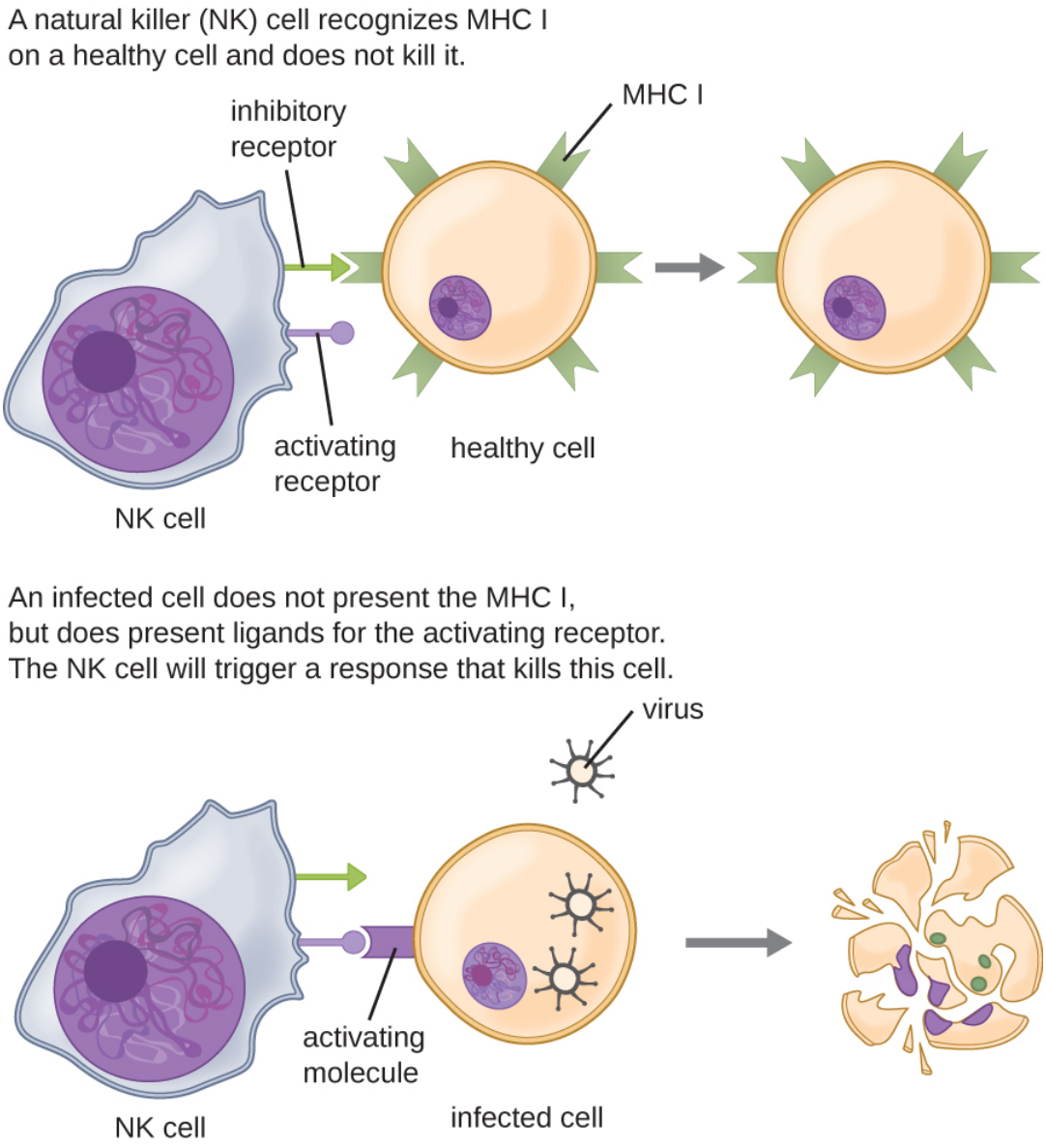


Figure **21.3.5**: Natural killer (NK) cells are inhibited by the presence of the major histocompatibility cell (MHC) receptor on healthy cells. Cancer cells and virus-infected cells have reduced expression of MHC and increased expression of activating molecules. When a NK cell recognizes decreased MHC and increased activating molecules, it will kill the abnormal cell.

Once a cell has been recognized as a target, the NK cell can use several different mechanisms to kill its target. For example, it may express cytotoxic membrane proteins and cytokines that stimulate the target cell to undergo apoptosis, or controlled cell suicide. NK cells may also use **perforin-mediated cytotoxicity** to induce apoptosis in target cells. This mechanism relies on two toxins released from granules in the cytoplasm of the NK cell: **perforin**, a protein that creates pores in the target cell, and **granzymes**, proteases that enter through the pores into the target cell's cytoplasm, where they trigger a cascade of protein activation that leads to apoptosis. The NK cell binds to the abnormal target cell, releases its destructive payload, and detaches from the target cell. While the target cell undergoes apoptosis, the NK cell synthesizes more perforin and proteases to use on its next target.

NK cells contain these toxic compounds in granules in their cytoplasm. When stained, the granules are azurophilic and can be visualized under a light microscope (Figure **21.3.6**). Even though they have granules, NK cells are not considered granulocytes because their granules are far less numerous than those found in true granulocytes. Furthermore, NK cells have a different lineage than granulocytes, arising from lymphoid rather than myeloid stem cells (Figure **21.3.1**).

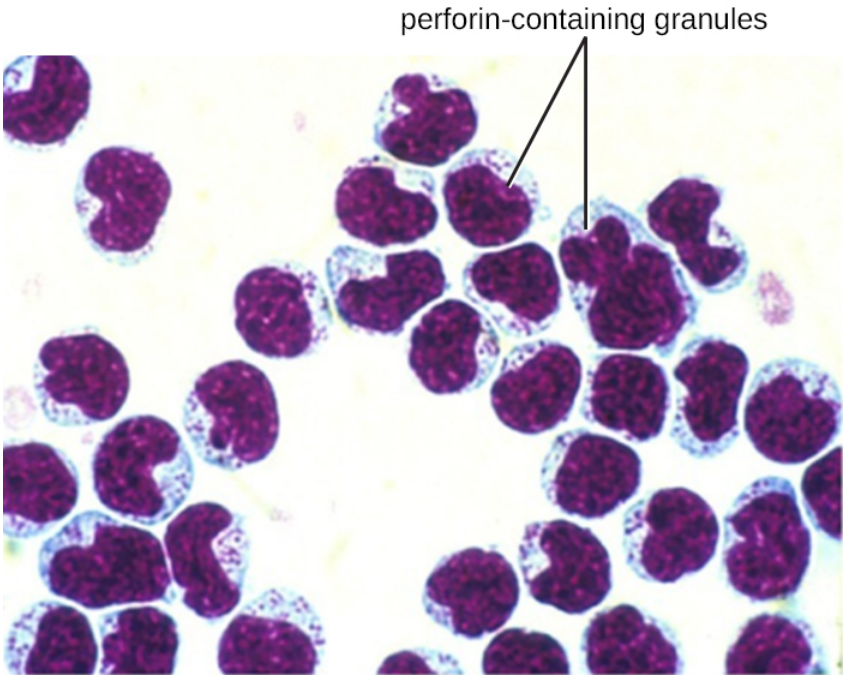


Figure **21.3.6**: Natural killer cell with perforin-containing granules. (credit: modification of work by Rolstad B)

Monocytes

The largest of the white blood cells, **monocytes** have a nucleus that lacks lobes, and they also lack granules in the cytoplasm (Figure **21.3.7**). Nevertheless, they are **effective phagocytes**, engulfing pathogens and apoptotic cells to help fight infection.

When monocytes leave the bloodstream and enter a specific body tissue, they differentiate into tissue-specific phagocytes called **macrophages** and **dendritic cells**. They are particularly important residents of lymphoid tissue, as well as nonlymphoid sites and organs. Macrophages and dendritic cells can reside in body tissues for significant lengths of time. Macrophages in specific body tissues develop characteristics suited to the particular tissue. Not only do they provide immune protection for the tissue in which they reside but they also support normal function of their neighboring tissue cells through the production of cytokines. Macrophages are given tissue-specific names, and a few examples of tissue-specific macrophages are listed in Table **21.3.1**. Dendritic cells are important sentinels residing in the skin and mucous membranes, which are portals of entry for many pathogens. Monocytes, macrophages, and dendritic cells are all highly phagocytic and important promoters of the immune response through their production and release of cytokines. These cells provide an essential bridge between innate and adaptive immune responses, as discussed in the next section as well as the next chapter.

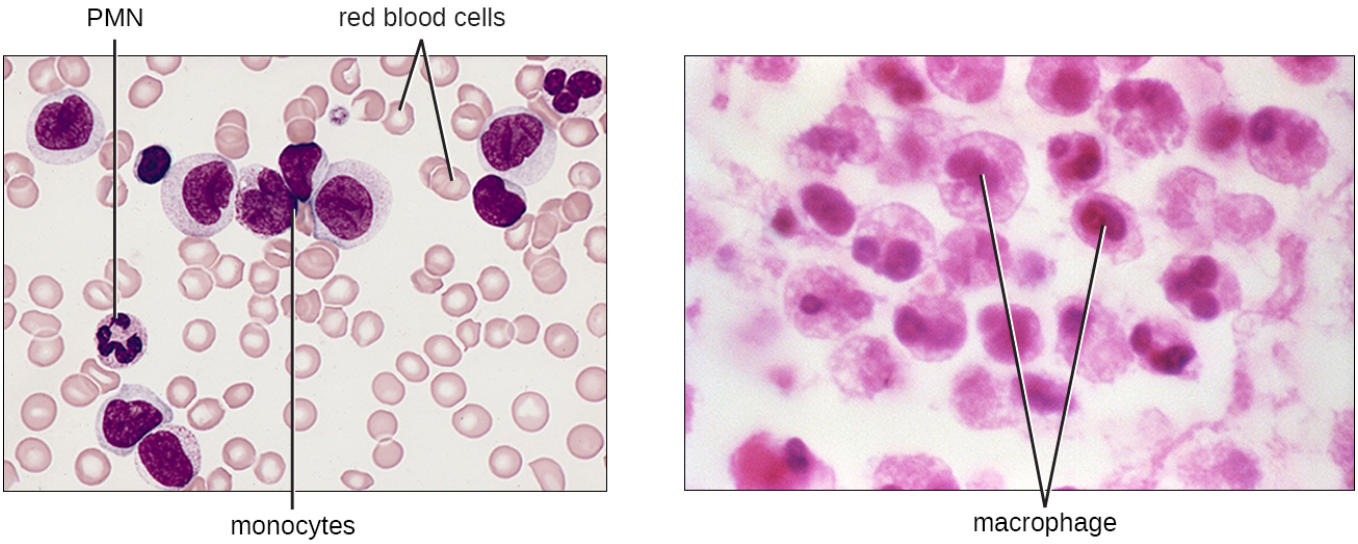


Figure **21.3.7**: Monocytes are large, agranular white blood cells with a nucleus that lacks lobes. When monocytes leave the bloodstream, they differentiate and become macrophages with tissue-specific properties. (credit left: modification of work by Armed Forces Institute of Pathology; credit right: modification of work by Centers for Disease Control and Prevention)

Table **21.3.1**: Macrophages Found in Various Body Tissues

Tissue	Macrophage
Brain and central nervous system	Microglial cells
Liver	Kupffer cells
Lungs	Alveolar macrophages (dust cells)
Peritoneal cavity	Peritoneal macrophages

Key Concepts and Summary

- **Natural killer** (**NK**) cells are lymphocytes that recognize and kill abnormal or infected cells by releasing proteins that trigger apoptosis.
- **Monocytes** are large, mononuclear leukocytes that circulate in the bloodstream. They may leave the bloodstream and take up residence in body tissues, where they differentiate and become tissue-specific **macrophages** and **dendritic cells**.

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21.4: Pathogen Recognition

21.04: Pathogen Recognition

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21.5: Phagocytosis

21.05: Phagocytosis

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21.6: Inflammation

21.06: Inflammation

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21.7: Fever

21.07: Fever

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21.E: Innate Immune Defenses II (Exercises)

21.1: Cellular Defenses

The formed elements of the blood include red blood cells (erythrocytes), white blood cells (leukocytes), and platelets (thrombocytes). Of these, leukocytes are primarily involved in the immune response. All formed elements originate in the bone marrow as stem cells (HSCs) that differentiate through hematopoiesis. Granulocytes are leukocytes characterized by a lobed nucleus and granules in the cytoplasm. These include neutrophils (PMNs), eosinophils, and basophils.

Multiple Choice

White blood cells are also referred to as which of the following?

A. platelets
B. erythrocytes
C. leukocytes
D. megakaryocytes

Answer

C

Hematopoiesis occurs inwhich of the following?

A. liver
B. bone marrow
C. kidneys
D. central nervous system

Answer

B

Granulocytes are which type of cell?

A. lymphocyte
B. erythrocyte
C. megakaryocyte
D. leukocyte

Answer

D

Matching

Match each cell type with its description.

___natural killer cell	A. stains with basic dye methylene blue, has large amounts of histamine in granules, and facilitates allergic responses and inflammation
___basophil	B. stains with acidic dye eosin, has histamine and major basic protein in granules, and facilitates responses to protozoa and helminths
___macrophage	C. recognizes abnormal cells, binds to them, and releases perforin and granzyme molecules, which induce apoptosis
___eosinophil	D. large agranular phagocyte that resides in tissues such as the brain and lungs

Answer

C, A, D, B

Match each cellular defense with the infection it would most likely target.

___natural killer cell	A. virus-infected cell
___neutrophil	B. tapeworm in the intestines
___eosinophil	C. bacteria in a skin lesion

Answer

A, C, B

Fill in the Blank

Platelets are also called _____.

Answer

thrombocytes

The cell in the bone marrow that gives rise to all other blood cell types is the _____.

Answer

pluripotent hematopoietic stem cell (HSC)

PMNs are another name for _____.

Answer

neutrophils

Kupffer cells residing in the liver are a type of _____.

Answer

macrophage

_____ are similar to basophils, but reside in tissues rather than circulating in the blood.

Answer

mast cells

Short Answer

Explain the difference between plasma and the formed elements of the blood.

List three ways that a neutrophil can destroy an infectious bacterium.

Critical Thinking

Neutrophils can sometimes kill human cells along with pathogens when they release the toxic contents of their granules into the surrounding tissue. Likewise, natural killer cells target human cells for destruction. Explain why it is advantageous for the immune system to have cells that can kill human cells as well as pathogens.

Refer to Figure 17.3.2. In a blood smear taken from a healthy patient, which type of leukocyte would you expect to observe in the highest numbers?

21.2: Pathogen Recognition and Phagocytosis

Phagocytes are cells that recognize pathogens and destroy them through phagocytosis. Recognition often takes place by the use of phagocyte receptors that bind molecules commonly found on pathogens, known as pathogen-associated molecular patterns (PAMPs). The receptors that bind PAMPs are called pattern recognition receptors, or PRRs. Toll-like receptors (TLRs) are one type of PRR found on phagocytes.

Multiple Choice

PAMPs would be found on the surface of which of the following?

A. pathogen
B. phagocyte
C. skin cell
D. blood vessel wall

Answer

A

_____ on phagocytes bind to PAMPs on bacteria, which triggers the uptake and destruction of the bacterial pathogens?

A. PRRs
B. AMPs
C. PAMPs
D. PMNs

Answer

A

Which of the following best characterizes the mode of pathogen recognition for opsonin-dependent phagocytosis?

A. Opsonins produced by a pathogen attract phagocytes through chemotaxis.
B. A PAMP on the pathogen’s surface is recognized by a phagocyte’s toll-like receptors.
C. A pathogen is first coated with a molecule such as a complement protein, which allows it to be recognized by phagocytes.
D. A pathogen is coated with a molecule such as a complement protein that immediately lyses the cell.

Answer

C

Fill in the Blank

_____, also known as diapedesis, refers to the exit from the bloodstream of neutrophils and other circulating leukocytes.

Answer

extravasation

Toll-like receptors are examples of _____.

Answer

pattern-recognition receptors (PRRs)

Short Answer

Briefly summarize the events leading up to and including the process of transendothelial migration.

21.3: Inflammation and Fever

Inflammation results from the collective response of chemical mediators and cellular defenses to an injury or infection. Acute inflammation is short lived and localized to the site of injury or infection. Chronic inflammation occurs when the inflammatory response is unsuccessful, and may result in the formation of granulomas (e.g., with tuberculosis) and scarring (e.g., with hepatitis C viral infections and liver cirrhosis).

Multiple Choice

Which refers to swelling as a result of inflammation?

A. erythema
B. edema
C. granuloma
D. vasodilation

Answer

B

Which type of inflammation occurs at the site of an injury or infection?

A. acute
B. chronic
C. endogenous
D. exogenous

Answer

A

Fill in the Blank

A(n) _____ is a walled-off area of infected tissue that exhibits chronic inflammation.

Answer

granuloma

The _____ is the part of the body responsible for regulating body temperature.

Answer

hypothalamus

Heat and redness, or _____, occur when the small blood vessels in an inflamed area dilate (open up), bringing more blood much closer to the surface of the skin.

Answer

erythema

Short Answer

Differentiate exogenous and endogenous pyrogens, and provide an example of each.

Critical Thinking

If a gram-negative bacterial infection reaches the bloodstream, large quantities of LPS can be released into the blood, resulting in a syndrome called septic shock. Death due to septic shock is a real danger. The overwhelming immune and inflammatory responses that occur with septic shock can cause a perilous drop in blood pressure; intravascular blood clotting; development of thrombi and emboli that block blood vessels, leading to tissue death; failure of multiple organs; and death of the patient. Identify and characterize two to three therapies that might be useful in stopping the dangerous events and outcomes of septic shock once it has begun, given what you have learned about inflammation and innate immunity in this chapter.

In Lubeck, Germany, in 1930, a group of 251 infants was accidentally administered a tainted vaccine for tuberculosis that contained live *Mycobacterium tuberculosis*. This vaccine was administered orally, directly exposing the infants to the deadly bacterium. Many of these infants contracted tuberculosis, and some died. However, 44 of the infants never contracted tuberculosis. Based on your knowledge of the innate immune system, what innate defenses might have inhibited *M. tuberculosis* enough to prevent these infants from contracting the disease?

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CHAPTER OVERVIEW

22: Adaptive Immunity I

People living in developed nations and born in the 1960s or later may have difficulty understanding the once heavy burden of devastating infectious diseases. For example, smallpox, a deadly viral disease, once destroyed entire civilizations but has since been eradicated. Thanks to the vaccination efforts by multiple groups, including the World Health Organization, Rotary International, and the United Nations Children's Fund (UNICEF), smallpox has not been diagnosed in a patient since 1977. Polio is another excellent example. This crippling viral disease paralyzed patients, who were often kept alive in "iron lung wards" as recently as the 1950s (Figure **22.1**). Today, vaccination against polio has nearly eradicated the disease. Vaccines have also reduced the prevalence of once-common infectious diseases such as chickenpox, German measles, measles, mumps, and whooping cough. The success of these and other vaccines is due to the very specific and adaptive host defenses that are the focus of this chapter.

Innate Nonspecific Host Defenses described innate immunity against microbial pathogens. Higher animals, such as humans, also possess an adaptive immune defense, which is highly specific for individual microbial pathogens. This specific adaptive immunity is acquired through active infection or vaccination and serves as an important defense against pathogens that evade the defenses of innate immunity.



Figure **22.1**: Polio was once a common disease with potentially serious consequences, including paralysis. Vaccination has all but eliminated the disease from most countries around the world. An iron-lung ward, such as the one shown in this 1953 photograph, housed patients paralyzed from polio and unable to breathe for themselves.

22.1: Adaptive Immune Defenses - Overview

22.2: Antigens

22.3: Antibodies

22.4: Antigen-Antibody Interactions

22.5: Major Histocompatibility Complexes (MHC)

22.6: Antigen Presenting Cells (APC)

22.E: Adaptive Immunity I (Exercises)

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22.1: Adaptive Immune Defenses - Overview

Learning Objectives

- List the defining characteristics of adaptive immunity
- Explain the difference between a primary and a secondary immune response
- Distinguish between humoral and cellular immunity

Adaptive Immunity

Adaptive immunity is defined by two important characteristics: **specificity** and **memory**. Specificity refers to the adaptive immune system's ability to target specific pathogens, and memory refers to its ability to quickly respond to pathogens to which it has previously been exposed. For example, when an individual recovers from chickenpox, the body develops a *memory* of the infection that will *specifically* protect it from the causative agent, the varicella-zoster virus, if it is exposed to the virus again later.

Specificity and memory are achieved by essentially programming certain cells involved in the immune response to respond rapidly to subsequent exposures of the pathogen. This programming occurs as a result of the first exposure to a pathogen or vaccine, which triggers a **primary response**. Subsequent exposures result in a **secondary response** that is faster and stronger as a result of the body's memory of the first exposure (Figure **22.1.1**). This secondary response, however, is specific to the pathogen in question. For example, exposure to one virus (e.g., varicella-zoster virus) will not provide protection against other viral diseases (e.g., measles, mumps, or polio).

Adaptive specific immunity involves the actions of two distinct cell types: **B lymphocytes (B cells)** and **T lymphocytes (T cells)**. Although B cells and T cells arise from a common hematopoietic stem cell differentiation pathway, their sites of maturation and their roles in adaptive immunity are very different.

B cells mature in the **bone marrow** and are responsible for the production of glycoproteins called **antibodies**, or **immunoglobulins**. Antibodies are involved in the body's defense against pathogens and toxins in the extracellular environment. Mechanisms of adaptive specific immunity that involve B cells and antibody production are referred to as **humoral immunity**. The maturation of **T cells** occurs in the **thymus**. T cells function as the central orchestrator of both innate and adaptive immune responses. They are also responsible for destruction of cells infected with intracellular pathogens. The targeting and destruction of intracellular pathogens by T cells is called **cell-mediated immunity**, or **cellular immunity**.

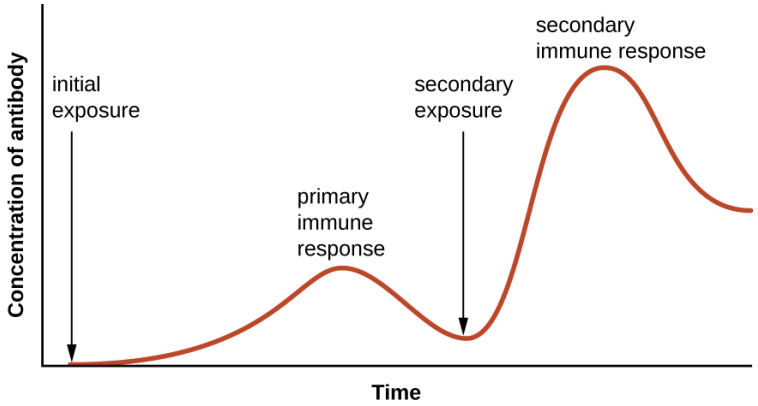


Figure **22.1.1**: This graph illustrates the primary and secondary immune responses related to antibody production after an initial and secondary exposure to an antigen. Notice that the secondary response is faster and provides a much higher concentration of antibody.

Key Concepts and Summary

- **Adaptive immunity** is an acquired defense against foreign pathogens that is characterized by **specificity** and **memory.** The first exposure to an antigen stimulates a **primary response**, and subsequent exposures stimulate a faster and strong **secondary response.**

- Adaptive immunity is a dual system involving **humoral immunity** (antibodies produced by B cells) and **cellular immunity** (T cells directed against intracellular pathogens).

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22.2: Antigens

Learning Objectives

- Compare an antigen with an epitope
- Identify factors that affect an antigen's antigenic potential
- Describe haptens and why they are not antigenic

Antigens

Antigen Definition

Activation of the adaptive immune defenses is triggered by pathogen-specific molecular structures called **antigens**. Antigens are similar to the pathogen-associated molecular patterns (PAMPs) discussed in Pathogen Recognition and Phagocytosis; however, whereas PAMPs are molecular structures found on numerous pathogens, antigens are unique to a specific pathogen. The antigens that stimulate adaptive immunity to chickenpox, for example, are unique to the varicella-zoster virus but significantly different from the antigens associated with other viral pathogens.

The term *antigen* was initially used to describe molecules that stimulate the production of antibodies; in fact, the term comes from a combination of the words antibody and generator, and a molecule that stimulates antibody production is said to be **antigenic**. However, the role of antigens is not limited to humoral immunity and the production of antibodies; antigens also play an essential role in stimulating cellular immunity, and for this reason antigens are sometimes more accurately referred to as **immunogens**. In this text, however, we will typically refer to them as antigens.

Pathogens possess a variety of structures that may contain antigens. For example, antigens from bacterial cells may be associated with their capsules, cell walls, fimbriae, flagella, or pili. Bacterial antigens may also be associated with extracellular toxins and enzymes that they secrete. Viruses possess a variety of antigens associated with their capsids, envelopes, and the spike structures they use for attachment to cells.

Antigen Classes

Antigens may belong to any number of molecular classes, including carbohydrates, lipids, nucleic acids, proteins, and combinations of these molecules. Antigens of different classes vary in their ability to stimulate adaptive immune defenses as well as in the type of response they stimulate (humoral or cellular). The **structural complexity** of an antigenic molecule is an important factor in its antigenic potential. In general, more complex molecules are more effective as antigens. For example, the three-dimensional complex structure of **proteins** make them the most effective and potent antigens, capable of stimulating both humoral and cellular immunity. In comparison, **carbohydrates** are less complex in structure and therefore less effective as antigens; they can only stimulate humoral immune defenses. **Lipids and nucleic acids** are the least antigenic molecules, and in some cases may only become antigenic when combined with proteins or carbohydrates to form glycolipids, lipoproteins, or nucleoproteins.

One reason the three-dimensional complexity of antigens is so important is that antibodies and T cells do not recognize and interact with an entire antigen but with smaller exposed regions on the surface of antigens called **epitopes**. A single antigen may possess several different epitopes (Figure 22.2.2), and different antibodies may bind to different epitopes on the same antigen (Figure 22.2.3). For example, the bacterial flagellum is a large, complex protein structure that can possess hundreds or even thousands of epitopes with unique three-dimensional structures. Moreover, flagella from different bacterial species (or even strains of the same species) contain unique epitopes that can only be bound by specific antibodies.

An antigen's **size** is another important factor in its antigenic potential. Whereas large antigenic structures like flagella possess multiple epitopes, some molecules are too small to be antigenic by themselves. Such molecules, called haptens, are essentially free epitopes that are not part of the complex three-dimensional structure of a larger antigen. For a hapten to become antigenic, it must first attach to a larger carrier molecule (usually a protein) to produce a conjugate antigen. The hapten-specific antibodies produced in response to the conjugate antigen are then able to interact with unconjugated free hapten molecules. Haptens are not known to be associated with any specific pathogens, but they are responsible for some allergic responses. For example, the hapten urushiol, a molecule found in the oil of plants that cause poison ivy, causes an immune response that can result in a severe rash (called contact dermatitis). Similarly, the hapten penicillin can cause allergic reactions to drugs in the penicillin class.

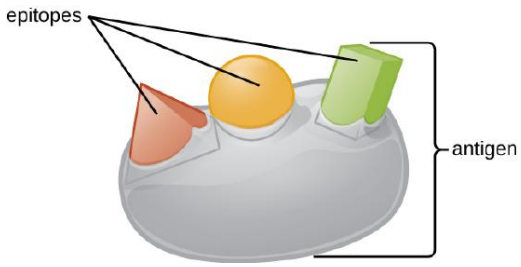


Figure **22.2.2**: An antigen is a macromolecule that reacts with components of the immune system. A given antigen may contain several motifs that are recognized by immune cells.

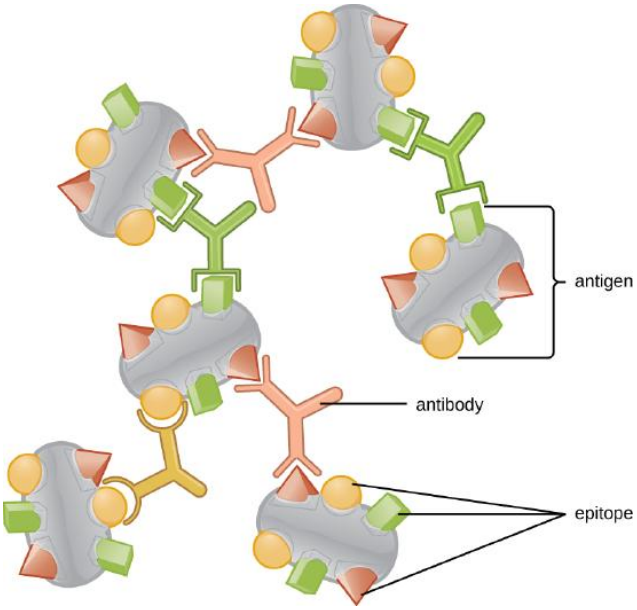


Figure **22.2.3**: A typical protein antigen has multiple epitopes, shown by the ability of three different antibodies to bind to different epitopes of the same antigen.

Key Concepts and Summary

- **Antigens**, also called **immunogens**, are molecules that activate adaptive immunity. A single antigen possesses smaller **epitopes**, each capable of inducing a specific adaptive immune response.
- An antigen's ability to stimulate an immune response depends on several factors, including its molecular class, molecular complexity, and size.

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22.3: Antibodies

Learning Objectives

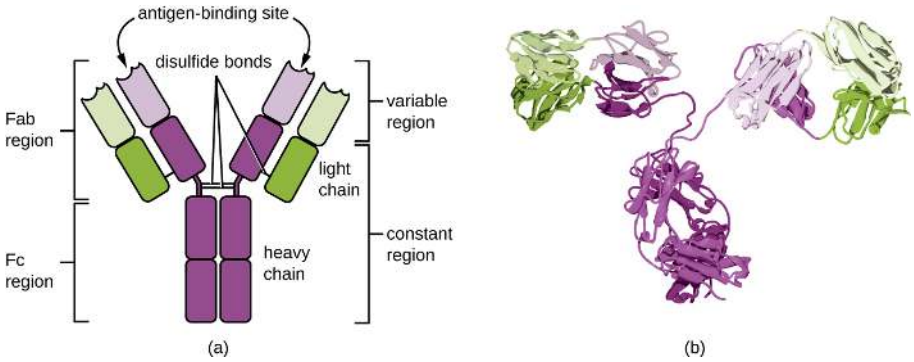
- Describe the structure and function of antibodies (immunoglobulins)
- Distinguish the functions of the Fab region and the Fc region
- Identify the five classes of antibodies
- Compare and contrast IgM and IgG in terms of their structure, function and abundance

Antibodies (Immunoglobulins)

Antibodies (also called **immunoglobulins**) are glycoproteins that are present in both the blood and tissue fluids. The basic structure of an antibody monomer consists of four protein chains held together by disulfide bonds (Figure **22.3.4**). A disulfide bond is a covalent bond between the sulfhydryl *R* groups found on two cysteine amino acids. The two largest chains are identical to each other and are called the **heavy chains**. The two smaller chains are also identical to each other and are called the **light chains**. Joined together, the heavy and light chains form a basic Y-shaped structure.

The two 'arms' of the Y-shaped antibody molecule are known as the **Fab region**, for "fragment of antigen binding." The far end of the Fab region is the **variable region**, which serves as the site of antigen binding. The amino acid sequence in the variable region dictates the three-dimensional structure, and thus the specific three-dimensional epitope to which the Fab region is capable of binding. Although the epitope specificity of the Fab regions is identical for each arm of a single antibody molecule, this region displays a high degree of variability between antibodies with different epitope specificities. Binding to the Fab region is necessary for neutralization of pathogens, agglutination or aggregation of pathogens, and antibody-dependent cell-mediated cytotoxicity.

The **constant region** of the antibody molecule includes the trunk of the Y and lower portion of each arm of the Y. The trunk of the Y is also called the **Fc region**, for "fragment of crystallization," and is the site of complement factor binding and binding to phagocytic cells during antibody-mediated opsonization.



Antibody Classes

The constant region of an antibody molecule determines its class, or isotype. The five classes of antibodies are IgG, IgM, IgA, IgD, and IgE. Each class possesses unique heavy chains designated by Greek letters γ, μ, α, δ, and ε, respectively. Antibody classes also exhibit important differences in abundance in serum, arrangement, body sites of action, functional roles, and size (Figure 22.3.5).

IgG is a monomer that is by far the most abundant antibody in human blood, accounting for about 80% of total serum antibody. IgG penetrates efficiently into tissue spaces, and is the only antibody class with the ability to cross the placental barrier, providing passive immunity to the developing fetus during pregnancy. IgG is also the most versatile antibody class in terms of its role in the body's defense against pathogens.

IgM is initially produced in a monomeric membrane-bound form that serves as an antigen-binding receptor on B cells. The secreted form of IgM assembles into a pentamer with five monomers of IgM bound together by a protein structure called the J chain. Although the location of the J chain relative to the Fc regions of the five monomers prevents IgM from performing some of the functions of IgG, the ten available Fab sites associated with a pentameric IgM make it an important antibody in the body's arsenal of defenses. IgM is the first antibody produced and secreted by B cells during the primary and secondary immune responses, making pathogen-specific IgM a valuable diagnostic marker during active or recent infections.

IgA accounts for about 13% of total serum antibody, and secretory IgA is the most common and abundant antibody class found in the mucus secretions that protect the mucous membranes. IgA can also be found in other secretions such as breast milk, tears, and saliva. Secretory IgA is assembled into a dimeric form with two monomers joined by a protein structure called the secretory component. One of the important functions of secretory IgA is to trap pathogens in mucus so that they can later be eliminated from the body.

Similar to IgM, **IgD** is a membrane-bound monomer found on the surface of B cells, where it serves as an antigen-binding receptor. However, IgD is not secreted by B cells, and only trace amounts are detected in serum. These trace amounts most likely come from the degradation of old B cells and the release of IgD molecules from their cytoplasmic membranes.

IgE is the least abundant antibody class in serum. Like IgG, it is secreted as a monomer, but its role in adaptive immunity is restricted to anti-parasitic defenses. The Fc region of IgE binds to basophils and mast cells. The Fab region of the bound IgE then interacts with specific antigen epitopes, causing the cells to release potent pro-inflammatory mediators. The inflammatory reaction resulting from the activation of mast cells and basophils aids in the defense against parasites, but this reaction is also central to allergic reactions (see Diseases of the Immune System.)

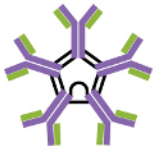
The Five Immunoglobulin (Ig) Classes					
Properties	**IgG monomer**	**IgM pentamer**	**Secretory IgA dimer**	**IgD monomer**	**IgE monomer**
Structure					
Heavy chains	γ	μ	α	δ	ε
Number of antigen-binding sites	2	10	4	2	2
Molecular weight (Daltons)	150,000	900,000	385,000	180,000	200,000
Percentage of total antibody in serum	80%	6%	13% (monomer)	<1%	<1%
Crosses placenta	yes	no	no	no	no
Fixes complement	yes	yes	no	no	no
Fc binds to	phagocytes				mast cells and basophils
Function	Neutralization, agglutination, complement activation, opsonization, and antibody-dependent cell-mediated cyotoxicity.	Neutralization, agglutination, and complement activation. The monomer form serves as the B-cell receptor.	Neutralization and trapping of pathogens in mucus.	B-cell receptor.	Activation of basophils and mast cells against parasites and allergens.

Figure **22.3.5**: The five Immunoglobulin classes

Key Concepts and Summary

- **Antibodies** (**immunoglobulins**) are Y-shaped glycoproteins with two Fab sites for binding antigens and an Fc portion involved in complement activation and opsonization.
- The five classes of antibody are **IgM**, **IgG**, **IgA**, **IgE**, and **IgD**, each differing in size, arrangement, location within the body, and function. The five primary functions of antibodies are neutralization, opsonization, agglutination, complement activation, and antibody-dependent cell-mediated cytotoxicity (ADCC).

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22.4: Antigen-Antibody Interactions

Learning Objectives

- Summarize the functions of antibodies
- Compare the mechanisms of opsonization and antibody-dependent cell-mediated cytotoxicity
- Relate the structure and location of antibodies to their functions

Antigen-Antibody Interactions

Different classes of antibody play important roles in the body's defense against pathogens. These functions include neutralization of pathogens, opsonization for phagocytosis, agglutination, complement activation, and antibody-dependent cell-mediated cytotoxicity. For most of these functions, antibodies also provide an important link between adaptive specific immunity and innate nonspecific immunity.

Neutralization

Neutralization involves the binding of certain antibodies (IgG, IgM, or IgA) to epitopes on the surface of pathogens or toxins, preventing their attachment to cells. For example, Secretory IgA can bind to specific pathogens and block initial attachment to intestinal mucosal cells. Similarly, specific antibodies can bind to certain toxins, blocking them from attaching to target cells and thus neutralizing their toxic effects. Viruses can be neutralized and prevented from infecting a cell by the same mechanism (Figure 22.4.6).

Opsonization

As described in Chemical Defenses, **opsonization** is the coating of a pathogen with molecules, such as complement factors, C-reactive protein, and serum amyloid A, to assist in phagocyte binding to facilitate phagocytosis. IgG antibodies also serve as excellent opsonins, binding their Fab sites to specific epitopes on the surface of pathogens. Phagocytic cells such as macrophages, dendritic cells, and neutrophils have receptors on their surfaces that recognize and bind to the Fc portion of the IgG molecules; thus, IgG helps such phagocytes attach to and engulf the pathogens they have bound (Figure 22.4.7).

Agglutination

Agglutination or **aggregation** involves the cross-linking of pathogens by antibodies to create large aggregates (Figure 22.4.8). IgG has two Fab antigen-binding sites, which can bind to two separate pathogen cells, clumping them together. When multiple IgG antibodies are involved, large aggregates can develop; these aggregates are easier for the kidneys and spleen to filter from the blood and easier for phagocytes to ingest for destruction. The pentameric structure of IgMprovides ten Fab binding sites per molecule, making it the most efficient antibody for agglutination.

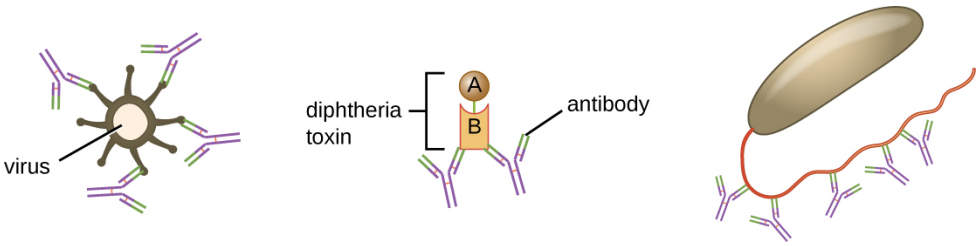


Figure 22.4.6: Neutralization involves the binding of specific antibodies to antigens found on bacteria, viruses, and toxins, preventing them from attaching to target cells.

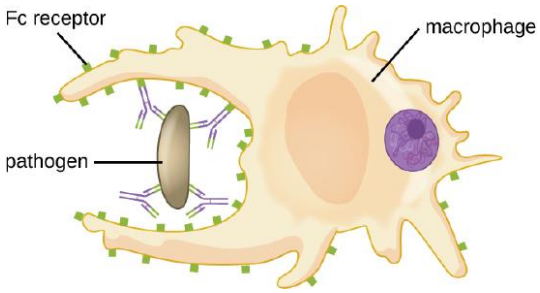


Figure **22.4.7**: Antibodies serve as opsonins and inhibit infection by tagging pathogens for destruction by macrophages, dendritic cells, and neutrophils. These phagocytic cells use Fc receptors to bind to IgG-opsonized pathogens and initiate the first step of attachment before phagocytosis.

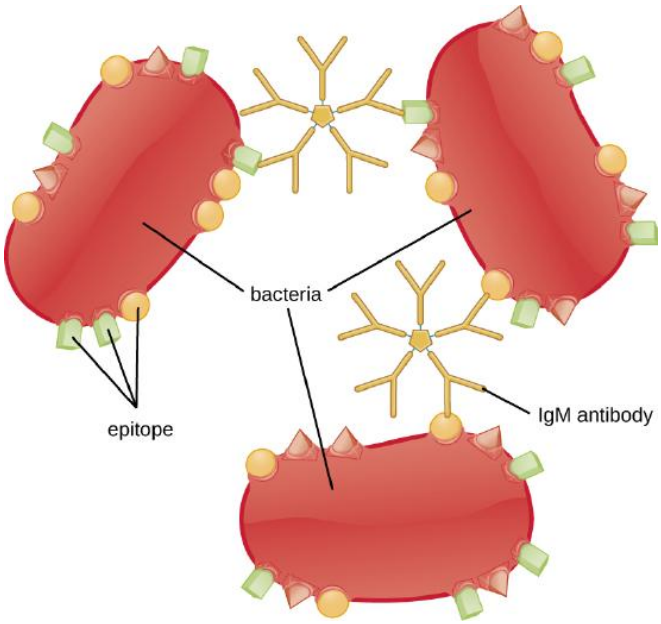


Figure **22.4.8**: Antibodies, especially IgM antibodies, agglutinate bacteria by binding to epitopes on two or more bacteria simultaneously. When multiple pathogens and antibodies are present, aggregates form when the binding sites of antibodies bind with separate pathogens.

Activation of the Complement System

Another important function of antibodies is **activation of the complement cascade**. As discussed in the previous chapter, the complement system is an important component of the innate defenses, promoting the inflammatory response, recruiting phagocytes to site of infection, enhancing phagocytosis by opsonization, and killing gram-negative bacterial pathogens with the membrane attack complex (MAC). Complement activation can occur through three different pathways (see Figure 17.2.2), but the most efficient is the classical pathway, which requires the initial binding of IgG or IgM antibodies to the surface of a pathogen cell, allowing for recruitment and activation of the C1 complex.

Antibody-Dependent Cell-mediated Cytotoxicity (ADCC)

Yet another important function of antibodies is **antibody-dependent cell-mediated cytotoxicity (ADCC)**, which enhances killing of pathogens that are too large to be phagocytosed. This process is best characterized for natural killer cells (NK cells), as shown in Figure **22.4.9**, but it can also involve macrophages and eosinophils. ADCC occurs when the Fab region of an IgG antibody binds to a large pathogen; Fc receptors on effector cells (e.g., NK cells) then bind to the Fc region of the antibody, bringing them into close proximity with the target pathogen. The effector cell then secretes powerful cytotoxins (e.g., perforin and granzymes) that kill the pathogen.

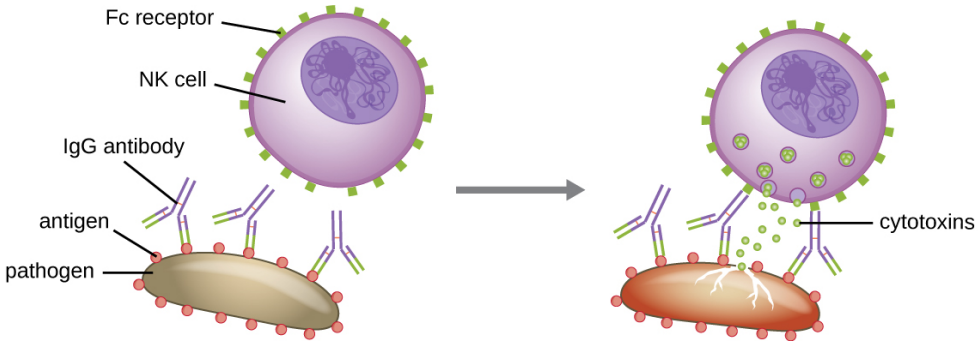


Figure **22.4.9**: In this example of ADCC, antibodies bind to a large pathogenic cell that is too big for phagocytosis and then bind to Fc receptors on the membrane of a natural killer cell. This interaction brings the NK cell into close proximity, where it can kill the pathogen through release of lethal extracellular cytotoxins.

Key Concepts and Summary

- The five classes of antibody are **IgM**, **IgG**, **IgA**, **IgE**, and **IgD**, each differing in size, arrangement, location within the body, and function. The five primary functions of antibodies are neutralization, opsonization, agglutination, complement activation, and antibody-dependent cell-mediated cytotoxicity (ADCC).

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22.5: Major Histocompatibility Complexes (MHC)

Learning Objectives

- Identify cells that express MHC I and/or MHC II molecules
- Describe the structures and cellular location of MHC I and MHC II molecules

As discussed in Cellular Defenses, major histocompatibility complex (MHC) molecules are expressed on the surface of healthy cells, identifying them as normal and "self" to natural killer (NK) cells. MHC molecules also play an important role in the presentation of foreign antigens, which is a critical step in the activation of T cells and thus an important mechanism of the adaptive immune system.

Major Histocompatibility Complex Molecules

The **major histocompatibility complex (MHC)** is a collection of genes coding for MHC molecules found on the surface of all nucleated cells of the body. In humans, the MHC genes are also referred to as **human leukocyte antigen (HLA)** genes. Mature red blood cells, which lack a nucleus, are the only cells that do not express MHC molecules on their surface.

There are two classes of MHC molecules involved in adaptive immunity, MHC I and MHC II (Figure 22.5.1). **MHC I** molecules are found on **all nucleated cells**; they present normal self-antigens as well as abnormal or nonself pathogens to the effector T cells involved in cellular immunity. In contrast, **MHC II** molecules are only found on **macrophages, dendritic cells, and B cells**; they present abnormal or nonself pathogen antigens for the initial activation of T cells.

Both types of MHC molecules are transmembrane glycoproteins that assemble as dimers in the cytoplasmic membrane of cells, but their structures are quite different. MHC I molecules are composed of a longer α protein chain coupled with a smaller β_2 microglobulin protein, and only the α chain spans the cytoplasmic membrane. The α chain of the MHC I molecule folds into three separate domains: α_1, α_2 and α_3. MHC II molecules are composed of two protein chains (an α and a β chain) that are approximately similar in length. Both chains of the MHC II molecule possess portions that span the plasma membrane, and each chain folds into two separate domains: α_1 and α_2, and β_1, and β_2. In order to present abnormal or non-self-antigens to T cells, MHC molecules have a cleft that serves as the antigen-binding site near the "top" (or outermost) portion of the MHC-I or MHC-II dimer. For MHC I, the antigen-binding cleft is formed by the α_1 and α_2 domains, whereas for MHC II, the cleft is formed by the α_1 and β_1 domains (Figure 22.5.1).

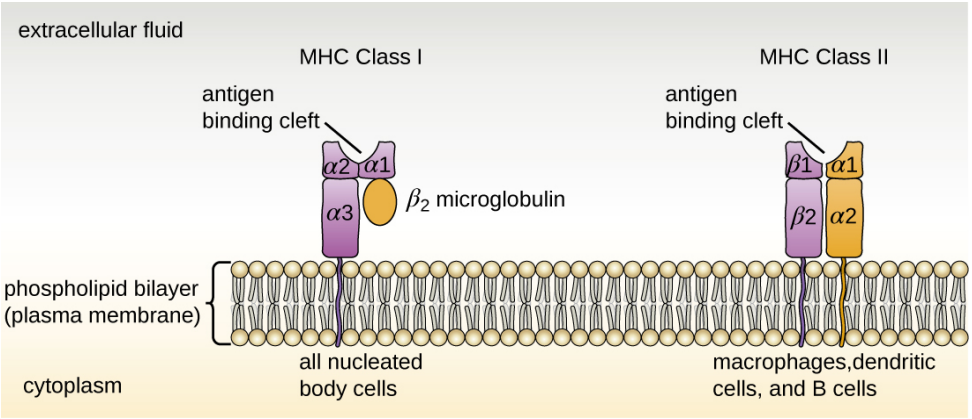


Figure 22.5.1: MHC I are found on all nucleated body cells, and MHC II are found on macrophages, dendritic cells, and B cells (along with MHC I). The antigen-binding cleft of MHC I is formed by domains α_1 and α_2. The antigen-binding cleft of MHC II is formed by domains α_1 and β_1.

Key Concepts and Summary

- **Major histocompatibility complex (MHC)** is a collection of genes coding for glycoprotein molecules expressed on the surface of all nucleated cells.

- **MHC I** molecules are expressed on all nucleated cells and are essential for presentation of normal “self” antigens. Cells that become infected by intracellular pathogens can present foreign antigens on MHC I as well, marking the infected cell for destruction.
- **MHC II** molecules are expressed only on the surface of **antigen-presenting cells** (macrophages, dendritic cells, and B cells). Antigen presentation with MHC II is essential for the activation of T cells.

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22.6: Antigen Presenting Cells (APC)

Learning Objectives

- Identify the cells that are antigen-presenting cells (APCs)
- Describe the process of antigen processing and presentation with MHC I and MHC II
- Explain the role of antigen-presentation in adaptive immunity

Antigen-Presenting Cells (APCs)

All nucleated cells in the body have mechanisms for processing and presenting antigens in association with MHC molecules. This signals the immune system, indicating whether the cell is normal and healthy or infected with an intracellular pathogen. However, only macrophages, dendritic cells, and B cells have the ability to present antigens specifically for the purpose of activating T cells; for this reason, these types of cells are sometimes referred to as **antigen-presenting cells (APCs)**.

While all APCs play a similar role in adaptive immunity, there are some important differences to consider. Macrophages and dendritic cells are phagocytes that ingest and kill pathogens that penetrate the first-line barriers (i.e., skin and mucous membranes). B cells, on the other hand, do not function as phagocytes but play a primary role in the production and secretion of antibodies. In addition, whereas macrophages and dendritic cells recognize pathogens through nonspecific receptor interactions (e.g., PAMPs, toll-like receptors, and receptors for opsonizing complement or antibody), B cells interact with foreign pathogens or their free antigens using antigen-specific immunoglobulin as receptors (monomeric IgD and IgM). When the immunoglobulin receptors bind to an antigen, the B cell internalizes the antigen by endocytosis before processing and presentting the antigen to T cells.

Antigen Presentation with MHC II Molecules

MHC II molecules are only found on the surface of APCs. Macrophages and dendritic cells use similar mechanisms for processing and presentation of antigens and their epitopes in association with MHC II; B cells use somewhat different mechanisms that will be described further in B Lymphocytes and Humoral Immunity. For now, we will focus on the steps of the process as they pertain to dendritic cells.

After a dendritic cell recognizes and attaches to a pathogen cell, the pathogen is internalized by phagocytosis and is initially contained within a phagosome. Lysosomes containing antimicrobial enzymes and chemicals fuse with the phagosome to create a phagolysosome, where degradation of the pathogen for antigen processing begins. Proteases (protein-degrading) are especially important in antigen processing because only protein antigen epitopes are presented to T cells by MHC II (Figure **22.6.2**).

APCs do not present all possible epitopes to T cells; only a selection of the most antigenic or immunodominantepitopes are presented. The mechanism by which epitopes are selected for processing and presentation by an APC is complicated and not well understood; however, once the most antigenic, immunodominant epitopes have been processed, they associate within the antigen-binding cleft of MHC II molecules and are translocated to the cell surface of the dendritic cell for presentation to T cells.

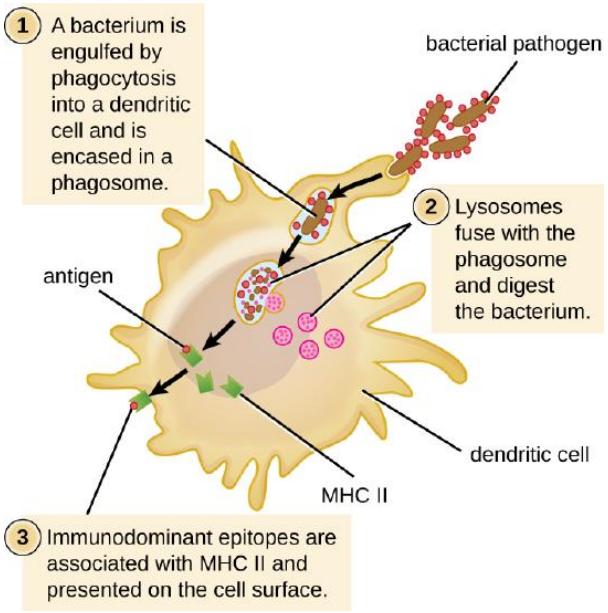


Figure 22.6.2: A dendritic cell phagocytoses a bacterial cell and brings it into a phagosome. Lysosomes fuse with the phagosome to create a phagolysosome, where antimicrobial chemicals and enzymes degrade the bacterial cell. Proteases process bacterial antigens, and the most antigenic epitopes are selected and presented on the cell's surface in conjunction with MHC II molecules. T cells recognize the presented antigens and are thus activated.

Antigen Presentation with MHC I Molecules

MHC I molecules, found on all normal, healthy, nucleated cells, signal to the immune system that the cell is a normal "self" cell. In a healthy cell, proteins normally found in the cytoplasm are degraded by proteasomes (enzyme complexes responsible for degradation and processing of proteins) and processed into self-antigen epitopes; these self-antigen epitopes bind within the MHC I antigen-binding cleft and are then presented on the cell surface. Immune cells, such as NK cells, recognize these self-antigens and do not target the cell for destruction. However, if a cell becomes infected with an intracellular pathogen (e.g., a virus), protein antigens specific to the pathogen are processed in the proteasomes and bind with MHC I molecules for presentation on the cell surface. This presentation of pathogen-specific antigens with MHC I signals that the infected cell must be targeted for destruction along with the pathogen.

Before elimination of infected cells can begin, APCs must first activate the T cells involved in cellular immunity. If an intracellular pathogen directly infects the cytoplasm of an APC, then the processing and presentation of antigens can occur as described (in proteasomes and on the cell surface with MHC I). However, if the intracellular pathogen does not directly infect APCs, an alternative strategy called **cross-presentation** is utilized. In cross-presentation, antigens are brought into the APC by mechanisms normally leading to presentation with MHC II (i.e., through phagocytosis), but the antigen is presented on an MHC I molecule for CD8 T cells. The exact mechanisms by which cross-presentation occur are not yet well understood, but it appears that cross-presentation is primarily a function of dendritic cells and not macrophages or B cells.

Key Concepts and Summary

- **Antigen-presenting cells (APCs)** primarily ingest pathogens by phagocytosis, destroy them in the phagolysosomes, process the protein antigens, and select the most antigenic/immunodominant epitopes with MHC II for presentation to T cells.
- **Cross-presentation** is a mechanism of antigen presentation and T-cell activation used by dendritic cells not directly infected by the pathogen; it involves phagocytosis of the pathogen but presentation on MHC I rather than MHC II.

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22.E: Adaptive Immunity I (Exercises)

22.1: Architecture of the Immune System

Adaptive immunity is defined by two important characteristics: specificity and memory. Specificity refers to the adaptive immune system's ability to target specific pathogens, and memory refers to its ability to quickly respond to pathogens to which it has previously been exposed. For example, when an individual recovers from chickenpox, the body develops a memory of the infection that will specifically protect it from the causative agent if it is exposed to the virus again later.

Multiple Choice

Antibodies are produced by _____.

A. plasma cells
B. T cells
C. bone marrow
D. B cells

Answer

A

Cellular adaptive immunity is carried out by _____.

A. B cells
B. T cells
C. bone marrow
D. neutrophils

Answer

B

A single antigen molecule may be composed of many individual _____.

A. T-cell receptors
B. B-cell receptors
C. MHC II
D. epitopes

Answer

D

Which class of molecules is the most antigenic?

A. polysaccharides
B. lipids
C. proteins
D. carbohydrates

Answer

C

Matching

Match the antibody class with its description.

___IgA	A. This class of antibody is the only one that can cross the placenta.
___IgD	B. This class of antibody is the first to appear after activation of B cells.
___IgE	C. This class of antibody is involved in the defense against parasitic infections and involved in allergic responses.
___IgG	D. This class of antibody is found in very large amounts in mucus secretions.
___IgM	E. This class of antibody is not secreted by B cells but is expressed on the surface of naïve B cells.

Answer

d, e, c, a, b

Fill in the Blank

There are two critically important aspects of adaptive immunity. The first is specificity, while the second is _____.

Answer

memory

_____ immunity involves the production of antibody molecules that bind to specific antigens.

Answer

Humoral

The heavy chains of an antibody molecule contain _____ region segments, which help to determine its class or isotype.

Answer

constant

The variable regions of the heavy and light chains form the _____ sites of an antibody.

Answer

antigen-binding

Short Answer

What is the difference between humoral and cellular adaptive immunity?

What is the difference between an antigen and a hapten?

Describe the mechanism of antibody-dependent cell-mediated cytotoxicity.

22.2: Antigens, Antigen Presenting Cells, and Major Histocompatibility Complexes

Major histocompatibility complex (MHC) molecules are expressed on the surface of healthy cells, identifying them as normal and “self” to natural killer (NK) cells. MHC molecules also play an important role in the presentation of foreign antigens, which is a critical step in the activation of T cells and thus an important mechanism of the adaptive immune system.

Multiple Choice

MHC I molecules present

A. processed foreign antigens from proteasomes.
B. processed self-antigens from phagolysosome.
C. antibodies.
D. T cell antigens.

Answer

A

MHC II molecules present

A. processed self-antigens from proteasomes.
B. processed foreign antigens from phagolysosomes.
C. antibodies.
D. T cell receptors.

Answer

B

Which type of antigen-presenting molecule is found on all nucleated cells?

A. MHC II
B. MHC I
C. antibodies
D. B-cell receptors

Answer

B

Which type of antigen-presenting molecule is found only on macrophages, dendritic cells, and B cells?

A. MHC I
B. MHC II
C. T-cell receptors
D. B-cell receptors

Answer

B

Fill in the Blank

MHC molecules are used for antigen _____ to T cells.

Answer

presentation

MHC II molecules are made up of two subunits (α and β) of approximately equal size, whereas MHC I molecules consist of a larger α subunit and a smaller subunit called _____.

Answer

β_2 microglobulin

Critical Thinking

Which mechanism of antigen presentation would be used to present antigens from a cell infected with a virus?

Which pathway of antigen presentation would be used to present antigens from an extracellular bacterial infection?

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CHAPTER OVERVIEW

23: Adaptive Immunity II

People living in developed nations and born in the 1960s or later may have difficulty understanding the once heavy burden of devastating infectious diseases. For example, smallpox, a deadly viral disease, once destroyed entire civilizations but has since been eradicated. Thanks to the vaccination efforts by multiple groups, including the World Health Organization, Rotary International, and the United Nations Children's Fund (UNICEF), smallpox has not been diagnosed in a patient since 1977. Polio is another excellent example. This crippling viral disease paralyzed patients, who were often kept alive in "iron lung wards" as recently as the 1950s (Figure 23.1). Today, vaccination against polio has nearly eradicated the disease. Vaccines have also reduced the prevalence of once-common infectious diseases such as chickenpox, German measles, measles, mumps, and whooping cough. The success of these and other vaccines is due to the very specific and adaptive host defenses that are the focus of this chapter.

Innate Nonspecific Host Defenses described innate immunity against microbial pathogens. Higher animals, such as humans, also possess an adaptive immune defense, which is highly specific for individual microbial pathogens. This specific adaptive immunity is acquired through active infection or vaccination and serves as an important defense against pathogens that evade the defenses of innate immunity.



Figure 23.1: Polio was once a common disease with potentially serious consequences, including paralysis. Vaccination has all but eliminated the disease from most countries around the world. An iron-lung ward, such as the one shown in this 1953 photograph, housed patients paralyzed from polio and unable to breathe for themselves.

23.1: T Lymphocytes - Overview

23.2: T-Cell Receptors (TCR) and Helper T Cells

23.3: Cytotoxic T Cells and Superantigens

23.4: B Lymphocytes and Antibodies

23.5: B Cell Activation

23.6: Vaccines

23.7: Variolation and Vaccination

23.8: Classes of Vaccines

23.E: Adaptive Immunity II (Exercises)

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23.1: T Lymphocytes - Overview

Learning Objectives

- Describe the process of T-cell production and maturation
- Summarize the steps involved in thymic selection
- Compare and contrast central tolerance versus peripheral tolerance
- Summarize the unique functions of the three classes of T cells

As explained in Overview of Specific Adaptive Immunity, the antibodies involved in humoral immunity often bind pathogens and toxins before they can attach to and invade host cells. Thus, **humoral immunity is primarily concerned with fighting pathogens in extracellular spaces.** However, pathogens that have already gained entry to host cells are largely protected from the humoral antibody-mediated defenses. **Cellular immunity, on the other hand, targets and eliminates intracellular pathogens through the actions of T lymphocytes, or T cells** (Figure 23.1.1). T cells also play a more central role in orchestrating the overall adaptive immune response (humoral as well as cellular) along with the cellular defenses of innate immunity.

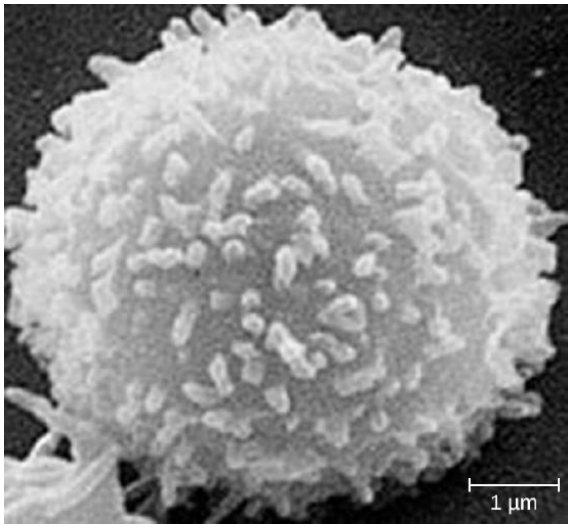


Figure 23.1.1: This scanning electron micrograph shows a T lymphocyte, which is responsible for the cell-mediated immune response. The spike-like membrane structures increase surface area, allowing for greater interaction with other cell types and their signals. (credit: modification of work by NCI)

T Cell Production and Maturation

T cells, like all other white blood cells involved in innate and adaptive immunity, are formed from **multipotent hematopoietic stem cells (HSCs)** in the bone marrow (see Figure 17.3.1). However, unlike the white blood cells of innate immunity, eventual T cells differentiate first into lymphoid stem cells that then become small, immature lymphocytes, sometimes called lymphoblasts. The first steps of differentiation occur in the red marrow of bones (Figure 23.1.2), after which immature T lymphocytes enter the bloodstream and travel to the thymus for the final steps of maturation (Figure 23.1.3). Once in the thymus, the immature T lymphocytes are referred to as thymocytes.

The maturation of thymocytes within the thymus can be divided into tree critical steps of positive and negative selection, collectively referred to as **thymic selection**. The first step of thymic selection occurs in the cortex of the thymus and involves the development of a functional T-cell receptor (TCR) that is required for activation by APCs. Thymocytes with defective TCRs are removed by negative selection through the induction of **apoptosis (programmed controlled cell death)**. The second step of thymic selection also occurs in the cortex and involves the positive selection of thymocytes that will interact appropriately with MHC molecules. Thymocytes that can interact appropriately with MHC molecules receive a positive stimulation that moves them further through the process of maturation, whereas thymocytes that do not interact appropriately are not stimulated and are eliminated by apoptosis. The third and final step of thymic selection occurs in both the cortex and medulla and involves negative selection to remove self-reacting thymocytes, those that react to self-antigens, by apoptosis. This final step is sometimes referred to

as **central tolerance** because it prevents self-reacting T cells from reaching the bloodstream and potentially causing autoimmune disease, which occurs when the immune system attacks healthy “self” cells.

Despite central tolerance, some self-reactive T cells generally escape the thymus and enter the peripheral bloodstream. Therefore, a second line of defense called peripheral tolerance is needed to protect against autoimmune disease. **Peripheral tolerance** involves mechanisms of **anergy** and inhibition of self-reactive T cells by **regulatory T cells**. Anergy refers to a state of nonresponsiveness to antigen stimulation. In the case of self-reactive T cells that escape the thymus, lack of an essential co-stimulatory signal required for activation causes anergy and prevents autoimmune activation. Regulatory T cells participate in peripheral tolerance by inhibiting the activation and function of self-reactive T cells and by secreting anti-inflammatory cytokines.

It is not completely understood what events specifically direct maturation of thymocytes into regulatory T cells. Current theories suggest the critical events may occur during the third step of thymic selection, when most self-reactive T cells are eliminated. Regulatory T cells may receive a unique signal that is below the threshold required to target them for negative selection and apoptosis. Consequently, these cells continue to mature and then exit the thymus, armed to inhibit the activation of self-reactive T cells.

It has been estimated that the three steps of thymic selection eliminate 98% of thymocytes. The remaining 2% that exit the thymus migrate through the bloodstream and lymphatic system to sites of secondary lymphoid organs/tissues, such as the lymph nodes, spleen, and tonsils (Figure **23.1.3**), where they await activation through the presentation of specific antigens by APCs. Until they are activated, they are known as **mature naïve T cells**.

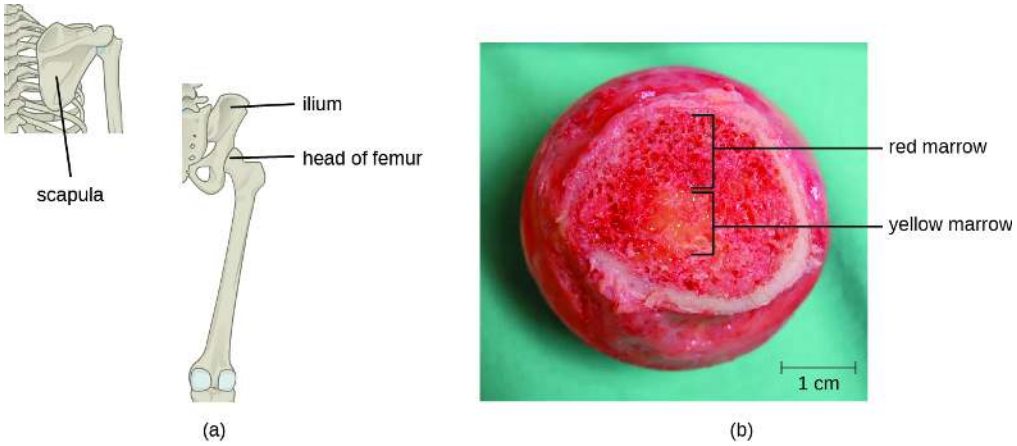


Figure **23.1.2**: (a) Red bone marrow can be found in the head of the femur (thighbone) and is also present in the flat bones of the body, such as the ilium and the scapula. (b) Red bone marrow is the site of production and differentiation of many formed elements of blood, including erythrocytes, leukocytes, and platelets. The yellow bone marrow is populated primarily with adipose cells.

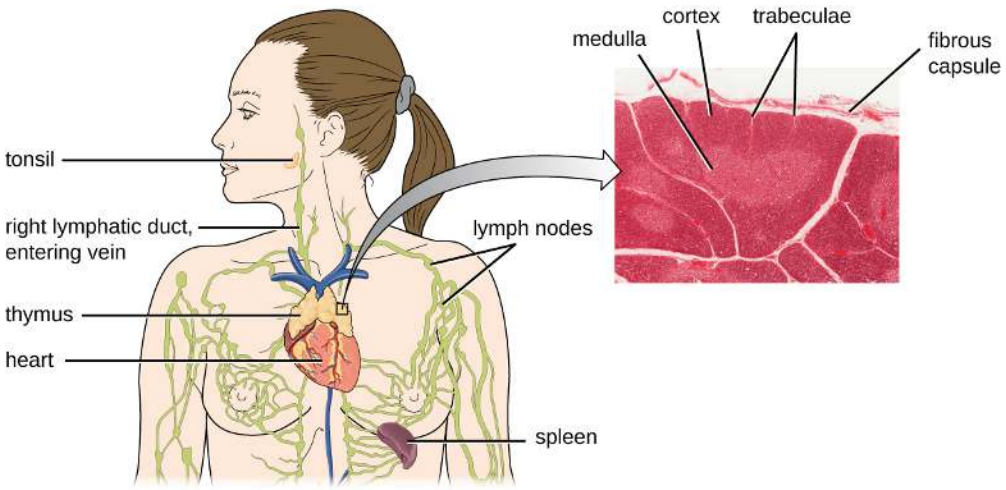


Figure **23.1.3**: The thymus is a bi-lobed, H-shaped glandular organ that is located just above the heart. It is surrounded by a fibrous capsule of connective tissue. The darkly staining cortex and the lighter staining medulla of individual lobules are clearly visible in the light micrograph of the thymus of a newborn (top right, LM × 100). (credit micrograph: modification of micrograph provided by the Regents of University of Michigan Medical School © 2012)

Classes of T Cells

T cells can be categorized into three distinct classes: **helper T cells, regulatory T cells, and cytotoxic T cells**. These classes are differentiated based on their expression of certain surface molecules, their mode of activation, and their functional roles in adaptive immunity (Table **23.1.1**).

All T cells produce **cluster of differentiation (CD)** molecules, cell surface glycoproteins that can be used to identify and distinguish between the various types of white blood cells. Although T cells can produce a variety of CD molecules, CD4and CD8 are the two most important used for differentiation of the classes. Helper T cells and regulatory T cells are characterized by the expression of CD4 on their surface, whereas cytotoxic T cells are characterized by the expression of CD8.

Classes of T cells can also be distinguished by the specific MHC molecules and APCs with which they interact for activation. Helper T cells and regulatory T cells can only be activated by APCs presenting antigens associated with MHC II. In contrast, cytotoxic T cells recognize antigens presented in association with MHC I, either by APCs or by nucleated cells infected with an intracellular pathogen.

The different classes of T cells also play different functional roles in the immune system. **Helper T cells** serve as the central orchestrators that help activate and direct functions of humoral and cellular immunity. In addition, helper T cells enhance the pathogen-killing functions of macrophages and NK cells of innate immunity. In contrast, the primary role of **regulatory T cells** is to prevent undesirable and potentially damaging immune responses. Their role in peripheral tolerance, for example, protects against autoimmune disorders, as discussed earlier. Finally, **cytotoxic T cells** are the primary effector cells for cellular immunity. They recognize and target cells that have been infected by intracellular pathogens, destroying infected cells along with the pathogens inside.

Table **23.1.1**: Classes of T Cells

Class	Surface CD Molecules	Activation	Functions
Helper T cells	CD4	APCs presenting antigens associated with MHC II	Orchestrate humoral and cellular immunity
			Involved in the activation of macrophages and NK cells
Regulatory T cells	CD4	APCs presenting antigens associated with MHC II	Involved in peripheral tolerance and prevention of autoimmune responses
Cytotoxic T cells	CD8	APCs or infected nucleated cells presenting antigens associated with MHC I	Destroy cells infected with intracellular pathogens

Key Concepts and Summary

- Immature T lymphocytes are produced in the red bone marrow and travel to the thymus for maturation.
- **Thymic selection** is a three-step process of negative and positive selection that determines which T cells will mature and exit the thymus into the peripheral bloodstream.
- **Central tolerance** involves negative selection of self-reactive T cells in the thymus, and **peripheral tolerance**involves **anergy** and **regulatory T cells** that prevent self-reactive immune responses and autoimmunity.

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23.2: T-Cell Receptors (TCR) and Helper T Cells

Learning Objectives

- Explain the genetic events that lead to diversity of T-cell receptors
- Recognize how the genetic rearrangement (VDJ recombination) process can produce millions of unique TCR binding sites
- Compare and contrast the structures of TCR and antibodies / immunoglobulins (Ab / Ig)
- Describe the activation of helper T cells
- Identify the subtypes of helper T cells and their functions

T-Cell Receptors

For both helper T cells and cytotoxic T cells, activation is a complex process that requires the interactions of multiple molecules and exposure to cytokines. The **T-cell receptor (TCR)** is involved in the first step of pathogen epitope recognition during the activation process.

The TCR comes from the same receptor family as the antibodies IgD and IgM, the antigen receptors on the B cell membrane surface, and thus shares common structural elements. Similar to antibodies, the TCR has a **variable region** and a **constant region**, and the variable region provides the antigen-binding site (Figure **23.2.4**). However, the structure of TCR is smaller and less complex than the immunoglobulin molecules (Figure 18.1.4). Whereas immunoglobulins have four peptide chains and Y-shaped structures, the TCR consists of just two peptide chains (α and β chains), both of which span the cytoplasmic membrane of the T cell.

TCRs are epitope-specific, and it has been estimated that 25 million T cells with unique epitope-binding TCRs are required to protect an individual against a wide range of microbial pathogens. Because the human genome only contains about 25,000 genes, we know that each specific TCR cannot be encoded by its own set of genes. This raises the question of how such a vast population of T cells with millions of specific TCRs can be achieved. The answer is a process called **genetic rearrangement**, which occurs in the thymus during the first step of thymic selection.

The genes that code for the variable regions of the TCR are divided into distinct gene segments called variable (V), diversity (D), and joining (J) segments. The genes segments associated with the α chain of the TCR consist 70 or more different V_α segments and 61 different J_α segments. The gene segments associated with the β chain of the TCR consist of 52 different V_β segments, two different D_β segments, and 13 different J_β segments. During the development of the functional TCR in the thymus, genetic rearrangement in a T cell brings together one V_α segment and one J_α segment to code for the variable region of the α chain. Similarly, genetic rearrangement brings one of the V_β segments together with one of the D_β segments and one of theJ_β segments to code for the variable region of the β chain. All the possible combinations of rearrangements between different segments of V, D, and J provide the genetic diversity required to produce millions of TCRs with unique epitope-specific variable regions.

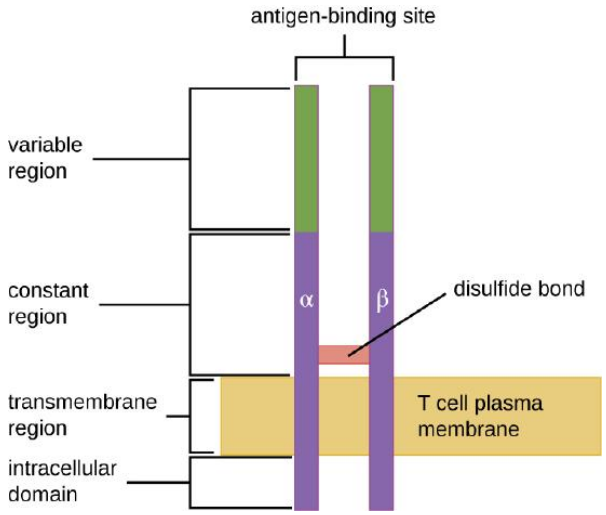


Figure **23.2.4**: A T-cell receptor spans the cytoplasmic membrane and projects variable binding regions into the extracellular space to bind processed antigens associated with MHC I or MHC II molecules.

Activation and Differentiation of Helper T Cells

Helper T Cell Activation

Helper T cells can only be activated by APCs presenting processed foreign epitopes in association with MHC II. The first step in the activation process is TCR recognition of the specific foreign epitope presented within the MHC II antigen-binding cleft. The second step involves the interaction of CD4 on the helper T cell with a region of the MHC II molecule separate from the antigen-binding cleft. This second interaction anchors the MHC II-TCR complex and ensures that the helper T cell is recognizing both the foreign ("nonself") epitope and "self" antigen of the APC; both recognitions are required for activation of the cell. In the third step, the APC and T cell secrete cytokines that activate the helper T cell.

Clonal Expansion and Clonal Differentiation

The activated helper T cell then proliferates, dividing by **mitosis** (a phenomenon called **clonal expansion**) to produce clonal naïve helper T cells. These naïve helper T cells then differentiate into subtypes with different functions (Figure **23.2.5**).

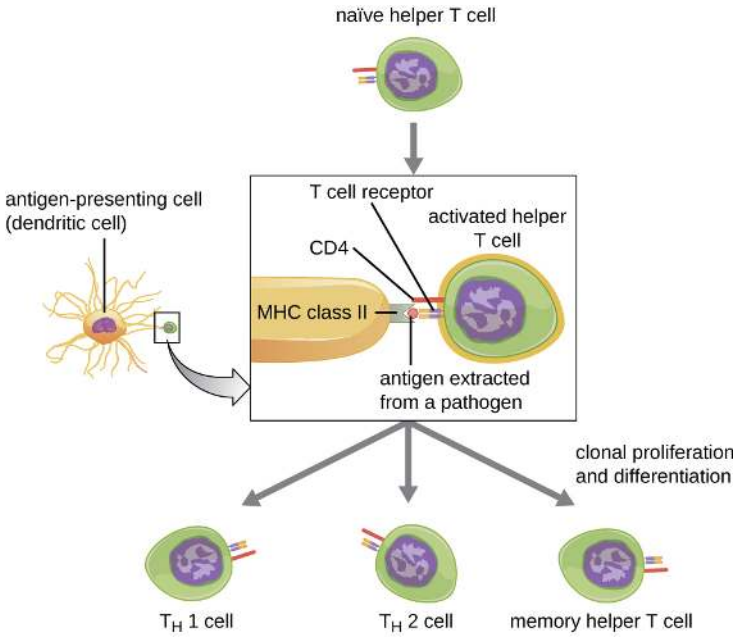


Figure **23.2.5**: This illustration depicts the activation of a naïve (unactivated) helper T cell by an antigen-presenting cell and the subsequent proliferation and differentiation of the activated T cell into different subtypes.

Subtypes of Helper T Cells

Activated helper T cells can differentiate into one of **four distinct subtypes**, summarized in Table **23.2.2**. The differentiation process is directed by APC-secreted cytokines. Depending on which APC-secreted cytokines interact with an activated helper T cell, the cell may differentiate into a T helper 1 (T_H1) cell, a T helper 2 (T_H2) cell, or a memory helper T cell. The two types of helper T cells are relatively short-lived effector cells, meaning that they perform various functions of the immediate immune response. In contrast, memory helper T cells are relatively long lived; they are programmed to "remember" a specific antigen or epitope in order to mount a rapid, strong, secondary response to subsequent exposures.

T_H1 cells secrete their own cytokines that are involved in stimulating and orchestrating other cells involved in adaptive and innate immunity. For example, they stimulate cytotoxic T cells, enhancing their killing of infected cells and promoting differentiation into memory cytotoxic T cells. T_H1 cells also stimulate macrophages and neutrophils to become more effective in their killing of intracellular bacteria. They can also stimulate NK cells to become more effective at killing target cells.

T_H2 cells play an important role in orchestrating the humoral immune response through their secretion of cytokines that activate B cells and direct B cell differentiation and antibody production. Various cytokines produced by T_H2 cells orchestrate antibody class

switching, which allows B cells to switch between the production of IgM, IgG, IgA, and IgE as needed to carry out specific antibody functions and to provide pathogen-specific humoral immune responses.

A third subtype of helper T cells called T_H17 cells was discovered through observations that immunity to some infections is not associated with T_H1 or T_H2 cells. T_H17 cells and the cytokines they produce appear to be specifically responsible for the body's defense against chronic mucocutaneous infections. Patients who lack sufficient T_H17 cells in the mucosa (e.g., HIV patients) may be more susceptible to bacteremia and gastrointestinal infections.[1]

Table **23.2.2**: Subtypes of Helper T Cells

Subtype	Functions
T_H1 cells	Stimulate cytotoxic T cells and produce memory cytotoxic T cells
	Stimulate macrophages and neutrophils (PMNs) for more effective intracellular killing of pathogens
	Stimulate NK cells to kill more effectively
T_H2 cells	Stimulate B cell activation and differentiation into plasma cells and memory B cells
	Direct antibody class switching in B cells
T_H17 cells	Stimulate immunity to specific infections such as chronic mucocutaneous infections
Memory helper T cells	"Remember" a specific pathogen and mount a strong, rapid secondary response upon re-exposure

Key Concepts and Summary

- The **TCR** is similar in structure to immunoglobulins, but less complex. Millions of unique epitope-binding TCRs are encoded through a process of genetic rearrangement of V, D, and J gene segments.
- T cells can be divided into three classes—**helper T cells, cytotoxic T cells,** and **regulatory T cells—**based on their expression of CD4 or CD8, the MHC molecules with which they interact for activation, and their respective functions.
- Activated helper T cells differentiate into **T_H1, T_H2, T_H17**, or **memory T cell subtypes**. Differentiation is directed by the specific cytokines to which they are exposed. T_H1, T_H2, and T_H17 perform different functions related to stimulation of adaptive and innate immune defenses. Memory T cells are long-lived cells that can respond quickly to secondary exposures.

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23.3: Cytotoxic T Cells and Superantigens

Learning Objectives

- Compare and contrast the activation of helper T cells and cytotoxic T cells (CTLs)
- Describe the mechanism of CTL-mediated destruction of infected cells
- Define superantigens and give some examples

Activation and Differentiation of Cytotoxic T Cells

Cytotoxic T cells (also referred to as **cytotoxic T lymphocytes, or CTLs**) are activated by APCs in a three-step process similar to that of helper T cells. The key difference is that the activation of cytotoxic T cells involves recognition of an antigen presented with MHC I (as opposed to MHC II) and interaction of CD8 (as opposed to CD4) with the receptor complex. After the successful co-recognition of foreign epitope and self-antigen, the production of cytokines by the APC and the cytotoxic T cell activate clonal proliferation and differentiation. Activated cytotoxic T cells can differentiate into effector cytotoxic T cells that target pathogens for destruction or memory cells that are ready to respond to subsequent exposures.

As noted, proliferation and differentiation of cytotoxic T cells is also stimulated by cytokines secreted from T_H1 cells activated by the same foreign epitope. The co-stimulation that comes from these T_H1 cells is provided by secreted cytokines. Although it is possible for activation of cytotoxic T cells to occur without stimulation from T_H1 cells, the activation is not as effective or long-lasting.

Once activated, cytotoxic T cells serve as the effector cells of cellular immunity, recognizing and kill cells infected with intracellular pathogens through a mechanism very similar to that of NK cells. However, whereas NK cells recognize nonspecific signals of cell stress or abnormality, cytotoxic T cells recognize infected cells through antigen presentation of pathogen-specific epitopes associated with MHC I. Once an infected cell is recognized, the TCR of the cytotoxic T cell binds to the epitope and releases perforin and granzymes that destroy the infected cell (Figure **23.3.6**). Perforin is a protein that creates pores in the target cell, and granzymes are proteases that enter the pores and induce apoptosis. This mechanism of programmed cell death is a controlled and efficient means of destroying and removing infected cells without releasing the pathogens inside to infect neighboring cells, as might occur if the infected cells were simply lysed.

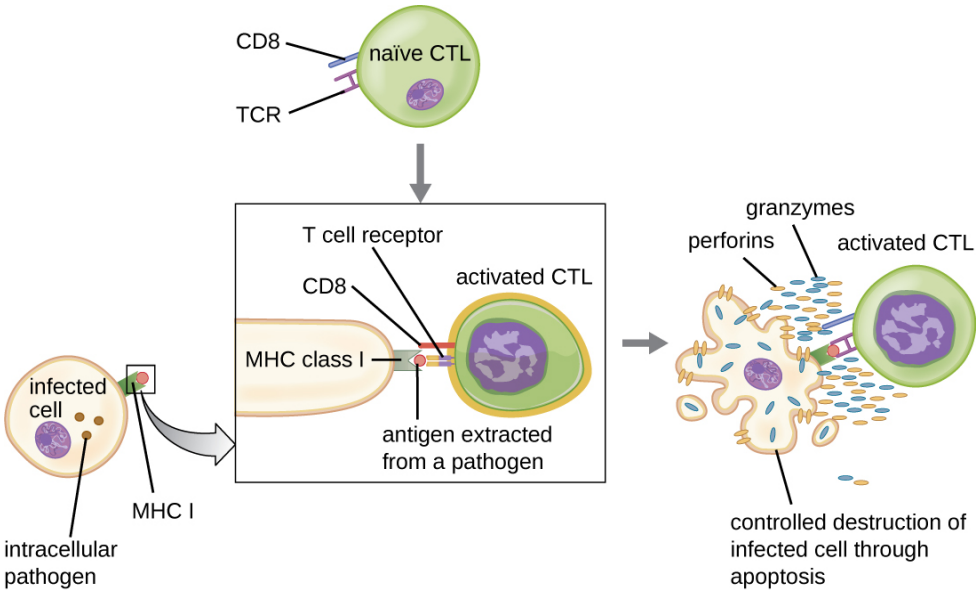


Figure **23.3.6**: This figure illustrates the activation of a naïve (unactivated) cytotoxic T cell (CTL) by an antigen-presenting MHC I molecule on an infected body cell. Once activated, the CTL releases perforin and granzymes that invade the infected cell and induce controlled cell death, or apoptosis.

Superantigens and Unregulated Activation of T Cells

When T cell activation is controlled and regulated, the result is a protective response that is effective in combating infections. However, if T cell activation is unregulated and excessive, the result can be a life-threatening. Certain bacterial and viral pathogens produce toxins known as **superantigens** (see Virulence Factors of Bacterial and Viral Pathogens) that can trigger such an unregulated response. Known bacterial superantigens include toxic shock syndrome toxin (TSST), staphylococcal enterotoxins, streptococcal pyrogenic toxins, streptococcal superantigen, and the streptococcal mitogenic exotoxin. Viruses known to produce superantigens include Epstein-Barr virus (human herpesvirus 4), cytomegalovirus (human herpesvirus 5), and others.

The mechanism of T cell activation by superantigens involves their simultaneous binding to MHC II molecules of APCs and the variable region of the TCR β chain. This binding occurs outside of the antigen-binding cleft of MHC II, so the superantigen will bridge together and activate MHC II and TCR without specific foreign epitope recognition (Figure **23.3.7**). The result is an excessive, uncontrolled release of cytokines, often called a cytokine storm, which stimulates an excessive inflammatory response. This can lead to a dangerous decrease in blood pressure, shock, multi-organ failure, and potentially, death.

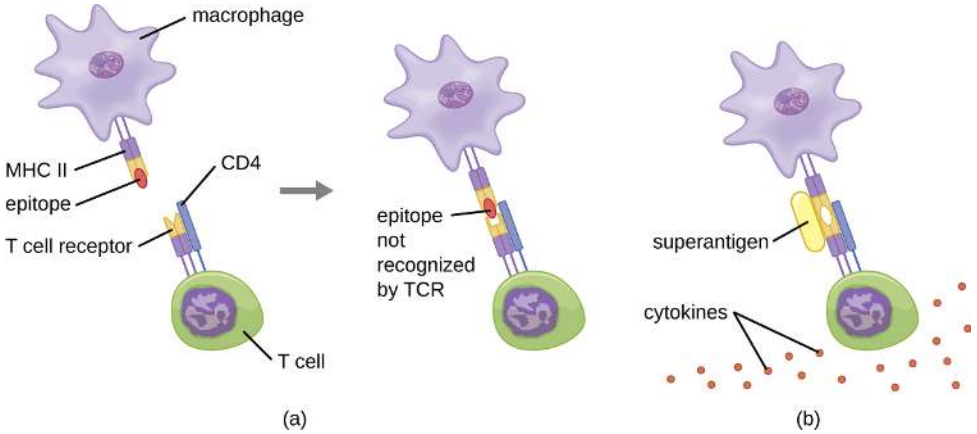


Figure **23.3.7**: (a) The macrophage in this figure is presenting a foreign epitope that does not match the TCR of the T cell. Because the T cell does not recognize the epitope, it is not activated. (b) The macrophage in this figure is presenting a superantigen that is not recognized by the TCR of the T cell, yet the superantigen still is able to bridge and bind the MHC II and TCR molecules. This nonspecific, uncontrolled activation of the T cell results in an excessive release of cytokines that activate other T cells and cause excessive inflammation. (credit: modification of work by "Microbiotic"/YouTube)

Case in Point: Superantigens

Melissa, an otherwise healthy 22-year-old woman, is brought to the emergency room by her concerned boyfriend. She complains of a sudden onset of high fever, vomiting, diarrhea, and muscle aches. In her initial interview, she tells the attending physician that she is on hormonal birth control and also is two days into the menstruation portion of her cycle. She is on no other medications and is not abusing any drugs or alcohol. She is not a smoker. She is not diabetic and does not currently have an infection of any kind to her knowledge.

While waiting in the emergency room, Melissa's blood pressure begins to drop dramatically and her mental state deteriorates to general confusion. The physician believes she is likely suffering from toxic shock syndrome (TSS). TSS is caused by the toxin TSST-1, a superantigen associated with *Staphylococcus aureus*, and improper tampon use is a common cause of infections leading to TSS. The superantigen inappropriately stimulates widespread T cell activation and excessive cytokine release, resulting in a massive and systemic inflammatory response that can be fatal.

Vaginal or cervical swabs may be taken to confirm the presence of the microbe, but these tests are not critical to perform based on Melissa's symptoms and medical history. The physician prescribes rehydration, supportive therapy, and antibiotics to stem the bacterial infection. She also prescribes drugs to increase Melissa's blood pressure. Melissa spends three days in the hospital undergoing treatment; in addition, her kidney function is monitored because of the high risk of kidney failure associated with TSS. After 72 hours, Melissa is well enough to be discharged to continue her recovery at home.

> **Exercise 23.3.6**
>
> In what way would antibiotic therapy help to combat a superantigen?

Key Concepts and Summary

- Once activated, cytotoxic T cells target and kill cells infected with intracellular pathogens. Killing requires recognition of specific pathogen epitopes presented on the cell surface using MHC I molecules. Killing is mediated by **perforin** and **granzymes** that induce apoptosis.
- **Superantigens** are bacterial or viral proteins that cause a nonspecific activation of helper T cells, leading to an excessive release of cytokines (**cytokine storm**) and a systemic, potentially fatal inflammatory response.

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23.4: B Lymphocytes and Antibodies

Learning Objectives

- Describe the production and maturation of B cells
- Compare the structure of B-cell receptors and T-cell receptors
- Compare the maturation of B cells with the maturation of T cells

Humoral immunity refers to mechanisms of the adaptive immune defenses that are mediated by antibodies secreted by B lymphocytes, or B cells. This section will focus on B cells and discuss their production and maturation, receptors, and mechanisms of activation.

B Cell Production and Maturation

Like T cells, B cells are formed from multipotent hematopoietic stem cells (HSCs) in the bone marrow and follow a pathway through lymphoid stem cell and lymphoblast (see Figure 17.3.1). Unlike T cells, however, lymphoblasts destined to become B cells do not leave the bone marrow and travel to the thymus for maturation. Rather, eventual B cells continue to mature in the bone marrow.

The first step of B cell maturation is an assessment of the functionality of their antigen-binding receptors. This occurs through positive selection for B cells with normal functional receptors. A mechanism of negative selection is then used to eliminate self-reacting B cells and minimize the risk of autoimmunity. Negative selection of self-reacting B cells can involve elimination by apoptosis, editing or modification of the receptors so they are no longer self-reactive, or induction of anergy in the B cell. Immature B cells that pass the selection in the bone marrow then travel to the spleenfor their final stages of maturation. There they become **naïve mature B cells**, i.e., mature B cells that have not yet been activated.

B Cell Receptor (BCR)

Like T cells, B cells possess antigen-specific receptors with diverse specificities. Although they rely on T cells for optimum function, B cells can be activated without help from T cells. B-cell receptors (BCRs) for naïve mature B cells are membrane-bound monomeric forms of IgD and IgM. They have two identical heavy chains and two identical light chains connected by disulfide bonds into a basic “Y” shape (Figure **23.4.1**). The trunk of the Y-shaped molecule, the constant region of the two heavy chains, spans the B cell membrane. The two antigen-binding sites exposed to the exterior of the B cell are involved in the binding of specific pathogen epitopes to initiate the activation process. It is estimated that each naïve mature B cell has upwards of 100,000 BCRs on its membrane, and each of these BCRs has an identical epitope-binding specificity.

In order to be prepared to react to a wide range of microbial epitopes, B cells, like T cells, use genetic rearrangementof hundreds of gene segments to provide the necessary diversity of receptor specificities. The variable region of the BCR heavy chain is made up of V, D, and J segments, similar to the β chain of the TCR. The variable region of the BCR light chain is made up of V and J segments, similar to the α chain of the TCR. Genetic rearrangement of all possible combinations of V-J-D (heavy chain) and V-J (light chain) provides for millions of unique antigen-binding sites for the BCR and for the antibodies secreted after activation.

One important difference between BCRs and TCRs is the way they can interact with antigenic epitopes. Whereas TCRs can only interact with antigenic epitopes that are presented within the antigen-binding cleft of MHC I or MHC II, BCRs do not require antigen presentation with MHC; they can interact with epitopes on free antigens or with epitopesdisplayed on the surface of intact pathogens. Another important difference is that TCRs only recognize protein epitopes, whereas BCRs can recognize epitopes associated with different molecular classes (e.g., proteins, polysaccharides, lipopolysaccharides).

Activation of B cells occurs through different mechanisms depending on the molecular class of the antigen. Activation of a B cell by a protein antigen requires the B cell to function as an APC, presenting the protein epitopes with MHC II to helper T cells. Because of their dependence on T cells for activation of B cells, protein antigens are classified as **T-dependent antigens**. In contrast, polysaccharides, lipopolysaccharides, and other nonprotein antigens are considered **T-independent antigens** because they can activate B cells without antigen processing and presentation to T cells.

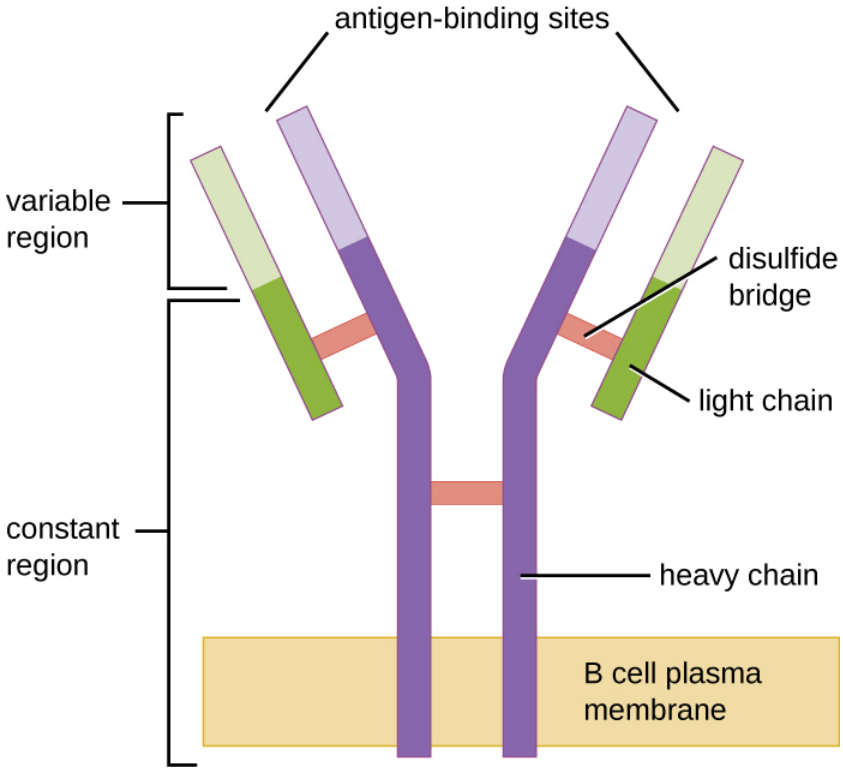


Figure **23.4.1**: B-cell receptors are embedded in the membranes of B cells. The variable regions of all of the receptors on a single cell bind the same specific antigen.

Key Concepts and Summary

- **B lymphocytes** or **B cells** produce antibodies involved in humoral immunity. B cells are produced in the bone marrow, where the initial stages of maturation occur, and travel to the spleen for final steps of maturation into naïve mature B cells.
- **B-cell receptors (BCRs)** are membrane-bound monomeric forms of IgD and IgM that bind specific antigen epitopes with their Fab antigen-binding regions. Diversity of antigen binding specificity is created by genetic rearrangement of V, D, and J segments similar to the mechanism used for TCR diversity.
- Protein antigens are called **T-dependent antigens** because they can only activate B cells with the cooperation of helper T cells. Other molecule classes do not require T cell cooperation and are called **T-independent antigens**.

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23.5: B Cell Activation

Learning Objectives

- Compare T-dependent and T-independent activation of B cells
- Identify the two signals required for T-cell independent activation of B cells
- Describe class switching and its importance
- Compare the primary and secondary antibody responses

T Cell-Independent Activation of B cells

Activation of B cells without the cooperation of helper T cells is referred to as T cell-independent activation and occurs when BCRs interact with T-independent antigens. T-independent antigens (e.g., polysaccharide capsules, lipopolysaccharide) have repetitive epitope units within their structure, and this repetition allows for the cross-linkageof multiple BCRs, providing the first signal for activation (Figure **23.5.2**). Because T cells are not involved, the second signal has to come from other sources, such as interactions of toll-like receptors with PAMPs or interactions with factors from the complement system.

Once a B cell is activated, it undergoes **clonal proliferation (clonal expansion)** and daughter cells differentiate into **plasma cells**. Plasma cells are antibody factories that secrete large quantities of antibodies. After differentiation, the surface BCRs disappear and the plasma cell secretes pentameric IgM molecules that have the same antigen specificity as the BCRs (Figure **23.5.2**).

The T cell-independent response is short-lived and does not result in the production of memory B cells. Thus it will not result in a secondary response to subsequent exposures to T-independent antigens.

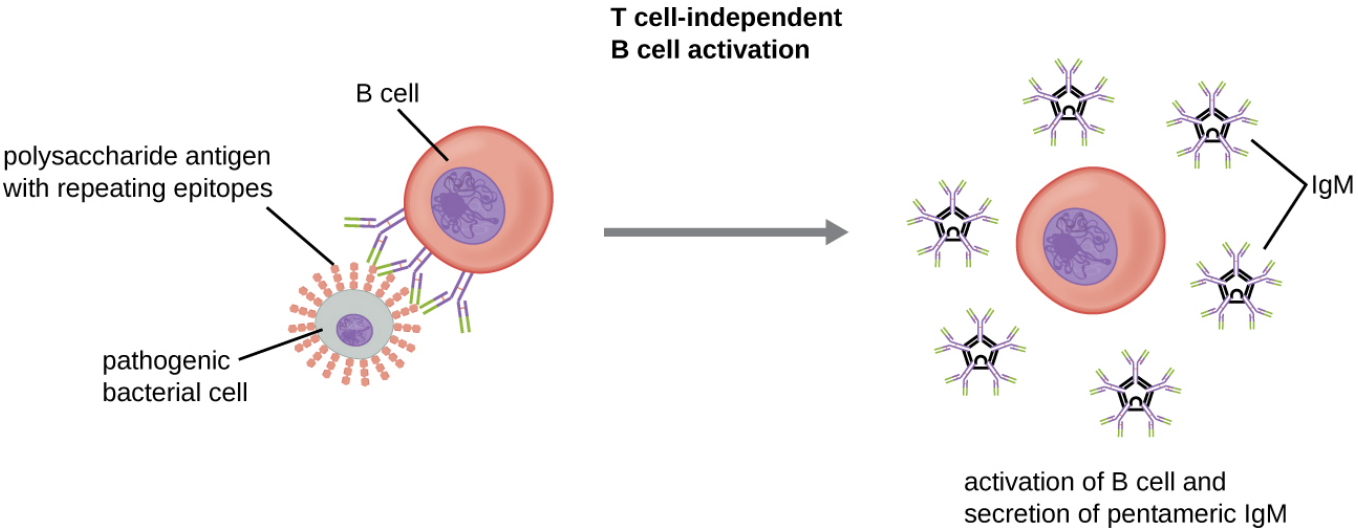


Figure **23.5.2**: T-independent antigens have repeating epitopes that can induce B cell recognition and activation without involvement from T cells. A second signal, such as interaction of TLRs with PAMPs (not shown), is also required for activation of the B cell. Once activated, the B cell proliferates and differentiates into antibody-secreting plasma cells.

T Cell-Dependent Activation of B cells

T cell-dependent activation of B cells is more complex than T cell-independent activation, but the resulting immune response is stronger and develops memory. T cell-dependent activation can occur either in response to free protein antigens or to protein antigens associated with an intact pathogen. Interaction between the BCRs on a naïve mature B cell and a free protein antigen stimulate internalization of the antigen, whereas interaction with antigens associated with an intact pathogen initiates the extraction of the antigen from the pathogen before internalization. Once internalized inside the B cell, the protein antigen is processed and presented with MHC II. The presented antigen is then recognized by helper T cells specific to the same antigen. The TCR of the

helper T cell recognizes the foreign antigen, and the T cell's CD4 molecule interacts with MHC II on the B cell. The coordination between B cells and helper T cells that are specific to the same antigen is referred to as **linked recognition**.

Once activated by linked recognition, T_H2 cells produce and secrete cytokines that activate the B cell and cause proliferation into clonal daughter cells. After several rounds of proliferation, additional cytokines provided by the T_H2 cells stimulate the differentiation of activated B cell clones into **memory B cells**, which will quickly respond to subsequent exposures to the same protein epitope, and plasma cells that lose their membrane BCRs and initially secrete pentameric IgM (Figure **23.5.3**).

After initial secretion of IgM, cytokines secreted by T_H2 cells stimulate the plasma cells to switch from IgM production to production of IgG, IgA, or IgE. This process, called **class switching** or **isotype switching**, allows plasma cellscloned from the same activated B cell to produce a variety of antibody classes with the same epitope specificity. Class switching is accomplished by genetic rearrangement of gene segments encoding the constant region, which determines an antibody's class. The variable region is not changed, so the new class of antibody retains the original epitope specificity.

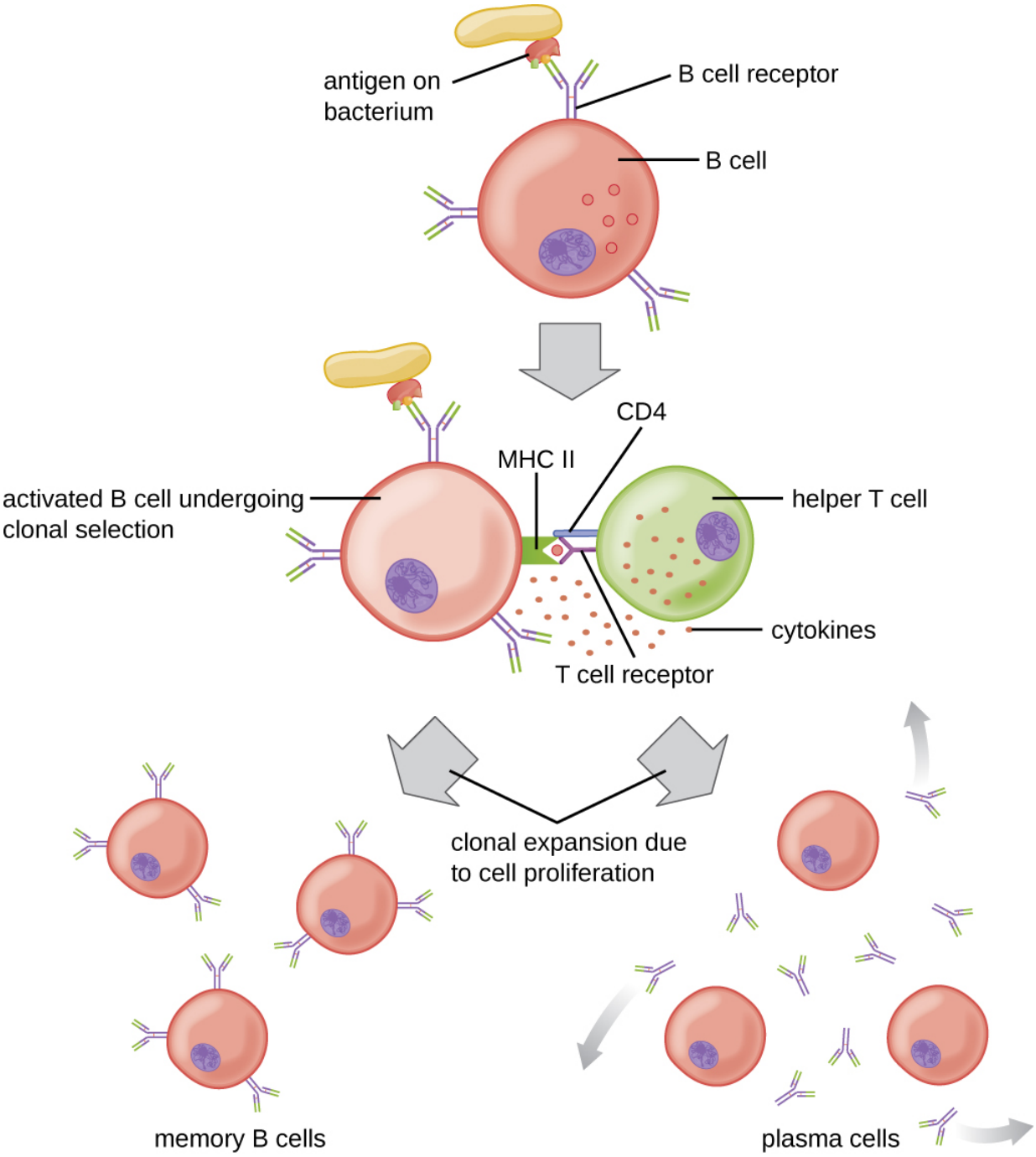


Figure **23.5.3**: In T cell-dependent activation of B cells, the B cell recognizes and internalizes an antigen and presents it to a helper T cell that is specific to the same antigen. The helper T cell interacts with the antigen presented by the B cell, which activates the T cell and stimulates the release of cytokines that then activate the B cell. Activation of the B cell triggers proliferation and differentiation into B cells and plasma cells.

Primary and Secondary Responses

T cell-dependent activation of B cells plays an important role in both the primary and secondary responses associated with adaptive immunity. With the first exposure to a protein antigen, a T cell-dependent primary antibody responseoccurs. The initial stage of the primary response is a **lag period**, or **latent period**, of approximately 10 days, during which no antibody can be detected in serum. This lag period is the time required for all of the steps of the primary response, including naïve mature B cell binding of antigen with BCRs, antigen processing and presentation, helper T cell activation, B cell activation, and clonal proliferation. The end of the lag period is characterized by a rise in IgM levels in the serum, as T_H2 cells stimulate B cell differentiation into plasma cells. IgM

levels reach their peak around 14 days after primary antigen exposure; at about this same time, T_H2 stimulates antibody class switching, and IgM levels in serum begin to decline. Meanwhile, levels of IgG increase until they reach a peak about three weeks into the primary response (Figure **23.5.4**).

During the primary response, some of the cloned B cells are differentiated into **memory B cells** programmed to respond to subsequent exposures. This secondary response occurs more quickly and forcefully than the primary response. The lag period is decreased to only a few days and the production of IgG is significantly higher than observed for the primary response (Figure **23.5.4**). In addition, the antibodies produced during the secondary response are more effective and bind with higher affinity to the targeted epitopes. Plasma cells produced during secondary responses live longer than those produced during the primary response, so levels of specific antibody remain elevated for a longer period of time.

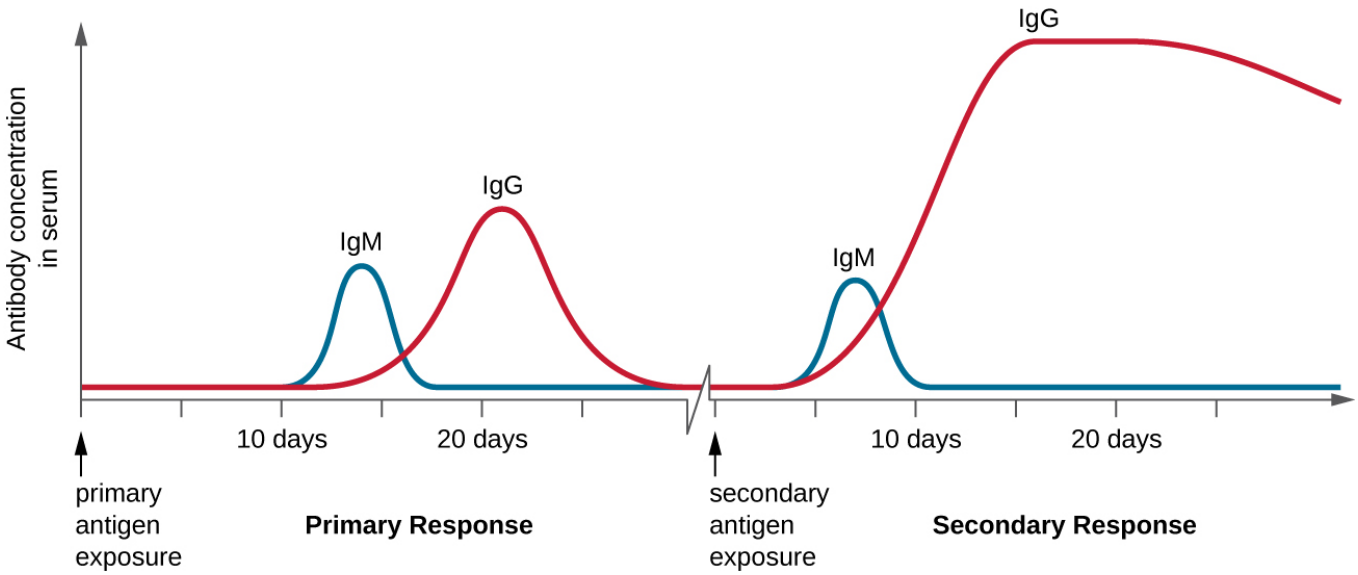


Figure **23.5.4**: Compared to the primary response, the secondary antibody response occurs more quickly and produces antibody levels that are higher and more sustained. The secondary response mostly involves IgG.

Key Concepts and Summary

- **T cell-independent activation** of B cells involves cross-linkage of BCRs by repetitive nonprotein antigen epitopes. It is characterized by the production of IgM by **plasma cells** and does not produce memory B cells.
- **T cell-dependent activation** of B cells involves processing and presentation of protein antigens to helper T cells, activation of the B cells by cytokines secreted from activated T_H2 cells, and plasma cells that produce different classes of antibodies as a result of **class switching**. **Memory B cells** are also produced.
- Secondary exposures to T-dependent antigens result in a secondary antibody response initiated by memory B cells. The secondary response develops more quickly and produces higher and more sustained levels of antibody with higher affinity for the specific antigen.

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23.6: Vaccines

Learning Objectives

- Compare the various kinds of artificial immunity
- Differentiate between active and passive immunity

For many diseases, prevention is the best form of treatment, and few strategies for disease prevention are as effective as vaccination. Vaccination is a form of artificial immunity. By artificially stimulating the adaptive immune defenses, a vaccine triggers memory cell production similar to that which would occur during a primary response. In so doing, the patient is able to mount a strong secondary response upon exposure to the pathogen—but without having to first suffer through an initial infection. In this section, we will explore several different kinds of artificial immunity along with various types of vaccines and the mechanisms by which they induce artificial immunity.

Classifications of Adaptive Immunity

All forms of adaptive immunity can be described as either active or passive. Active immunity refers to the activation of an individual's own adaptive immune defenses, whereas passive immunity refers to the transfer of adaptive immune defenses from another individual or animal. Active and passive immunity can be further subdivided based on whether the protection is acquired naturally or artificially.

Natural active immunity is adaptive immunity that develops after natural exposure to a pathogen (Figure **23.6.1**). Examples would include the lifelong immunity that develops after recovery from a chickenpox or measles infection (although an acute infection is not always necessary to activate adaptive immunity). The length of time that an individual is protected can vary substantially depending upon the pathogen and antigens involved. For example, activation of adaptive immunity by protein spike structures during an intracellular viral infection can activate lifelong immunity, whereas activation by carbohydrate capsule antigens during an extracellular bacterial infection may activate shorter-term immunity.

Natural passive immunity involves the natural passage of antibodies from a mother to her child before and after birth. IgG is the only antibody class that can cross the placenta from mother's blood to the fetal blood supply. Placental transfer of IgG is an important passive immune defense for the infant, lasting up to six months after birth. Secretory IgA can also be transferred from mother to infant through breast milk.

Artificial passive immunity refers to the transfer of antibodies produced by a donor (human or animal) to another individual. This transfer of antibodies may be done as a prophylactic measure (i.e., to prevent disease after exposure to a pathogen) or as a strategy for treating an active infection. For example, artificial passive immunity is commonly used for post-exposure prophylaxis against rabies, hepatitis A, hepatitis B, and chickenpox (in high risk individuals). Active infections treated by artificial passive immunity include cytomegalovirus infections in immunocompromised patients and Ebola virus infections. In 1995, eight patients in the Democratic Republic of the Congo with active Ebola infections were treated with blood transfusions from patients who were recovering from Ebola. Only one of the eight patients died (a 12.5% mortality rate), which was much lower than the expected 80% mortality rate for Ebola in untreated patients.[1] Artificial passive immunity is also used for the treatment of diseases caused by bacterial toxins, including tetanus, botulism, and diphtheria.

Artificial active immunity is the foundation for vaccination. It involves the activation of adaptive immunity through the deliberate exposure of an individual to weakened or inactivated pathogens, or preparations consisting of key pathogen antigens.

Mechanisms of Acquisition of Immunity		
	Natural acquired	**Artificial acquired**
Passive	Immunity acquired from antibodies passed in breast milk or through placenta 	Immunity gained through antibodies harvested from another person or an animal 
Active	Immunity gained through illness and recovery 	Immunity acquired through a vaccine 

Figure **23.6.1**: The four classifications of immunity. (credit top left photo: modification of work by USDA; credit top right photo: modification of work by "Michaelberry"/Wikimedia; credit bottom left photo: modification of work by Centers for Disease Control and Prevention; credit bottom right photo: modification of work by Friskila Silitonga, Indonesia, Centers for Disease Control and Prevention)

Herd Immunity

The four kinds of immunity just described result from an individual's adaptive immune system. For any given disease, an individual may be considered immune or susceptible depending on his or her ability to mount an effective immune response upon exposure. Thus, any given population is likely to have some individuals who are immune and other individuals who are susceptible. If a population has very few susceptible individuals, even those susceptible individuals will be protected by a phenomenon called **herd immunity**. Herd immunity has nothing to do with an individual's ability to mount an effective immune response; rather, it occurs because there are too few susceptible individuals in a population for the disease to spread effectively.

Vaccination programs create herd immunity by greatly reducing the number of susceptible individuals in a population. Even if some individuals in the population are not vaccinated, as long as a certain percentage is immune (either naturally or artificially), the few susceptible individuals are unlikely to be exposed to the pathogen. However, because new individuals are constantly entering populations (for example, through birth or relocation), vaccination programs are necessary to maintain herd immunity.

Vaccination: Obligation or Choice

A growing number of parents are choosing not to vaccinate their children. They are dubbed "antivaxxers," and the majority of them believe that vaccines are a cause of autism (or other disease conditions), a link that has now been thoroughly disproven. Others object to vaccines on religious or moral grounds (e.g., the argument that Gardasil vaccination against HPV may

promote sexual promiscuity), on personal ethical grounds (e.g., a conscientious objection to any medical intervention), or on political grounds (e.g., the notion that mandatory vaccinations are a violation of individual liberties).[2]

It is believed that this growing number of unvaccinated individuals has led to new outbreaks of whooping cough and measles. We would expect that herd immunity would protect those unvaccinated in our population, but herd immunity can only be maintained if enough individuals are being vaccinated.

Vaccination is clearly beneficial for public health. But from the individual parent's perspective the view can be murkier. Vaccines, like all medical interventions, have associated risks, and while the risks of vaccination may be extremely low compared to the risks of infection, parents may not always understand or accept the consensus of the medical community. Do such parents have a right to withhold vaccination from their children? Should they be allowed to put their children—and society at large—at risk?

Many governments insist on childhood vaccinations as a condition for entering public school, but it has become easy in most states to opt out of the requirement or to keep children out of the public system. Since the 1970s, West Virginia and Mississippi have had in place a stringent requirement for childhood vaccination, without exceptions, and neither state has had a case of measles since the early 1990s. California lawmakers recently passed a similar law in response to a measles outbreak in 2015, making it much more difficult for parents to opt out of vaccines if their children are attending public schools. Given this track record and renewed legislative efforts, should other states adopt similarly strict requirements?

What role should health-care providers play in promoting or enforcing universal vaccination? Studies have shown that many parents' minds can be changed in response to information delivered by health-care workers, but is it the place of health-care workers to try to persuade parents to have their children vaccinated? Some health-care providers are understandably reluctant to treat unvaccinated patients. Do they have the right to refuse service to patients who decline vaccines? Do insurance companies have the right to deny coverage to antivaxxers? These are all ethical questions that policymakers may be forced to address as more parents skirt vaccination norms.

Key Concepts and Summary

- Adaptive immunity can be divided into four distinct classifications: **natural active immunity, natural passive immunity, artificial passive immunity,** and **artificial active immunity.**
- Artificial active immunity is the foundation for **vaccination** and vaccine development. Vaccination programs not only confer artificial immunity on individuals, but also foster **herd immunity** in populations.

Footnotes

1. K. Mupapa, M. Massamba, K. Kibadi, K. Kivula, A. Bwaka, M. Kipasa, R. Colebunders, J. J. Muyembe-Tamfum. "Treatment of Ebola Hemorrhagic Fever with Blood Transfusions from Convalescent Patients." *Journal of Infectious Diseases* 179 Suppl. (1999): S18–S23.
2. Elizabeth Yale. "Why Anti-Vaccination Movements Can Never Be Tamed." *Religion & Politics*, July 22, 2014. religionandpolitics.org/2014/...never-be-tamed.
3. N. J. Willis. "Edward Jenner and the Eradication of Smallpox." *Scottish Medical Journal* 42 (1997): 118–121.

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23.7: Variolation and Vaccination

Learning Objectives

- Differentiate between variolation and vaccination
- Explain why vaccination is less risky than variolation

Variolation and Vaccination

Thousands of years ago, it was first recognized that individuals who survived a smallpox infection were immune to subsequent infections. The practice of inoculating individuals to actively protect them from smallpox appears to have originated in the 10th century in China, when the practice of **variolation** was described (Figure **23.7.2**). Variolation refers to the deliberate inoculation of individuals with infectious material from scabs or pustules of smallpox victims. Infectious materials were either injected into the skin or introduced through the nasal route. The infection that developed was usually milder than naturally acquired smallpox, and recovery from the milder infection provided protection against the more serious disease.

Although the majority of individuals treated by variolation developed only mild infections, the practice was not without risks. More serious and sometimes fatal infections did occur, and because smallpox was contagious, infections resulting from variolation could lead to epidemics. Even so, the practice of variolation for smallpox prevention spread to other regions, including India, Africa, and Europe.



Figure **23.7.2**: Variolation for smallpox originated in the Far East and the practice later spread to Europe and Africa. This Japanese relief depicts a patient receiving a smallpox variolation from the physician Ogata Shunsaku (1748–1810).

Although variolation had been practiced for centuries, the English physician Edward Jenner (1749–1823) is generally credited with developing the modern process of vaccination. Jenner observed that milkmaids who developed cowpox, a disease similar to smallpox but milder, were immune to the more serious smallpox. This led Jenner to hypothesize that exposure to a less virulent pathogen could provide immune protection against a more virulent pathogen, providing a safer alternative to variolation. In 1796, Jenner tested his hypothesis by obtaining infectious samples from a milkmaid's active cowpox lesion and injecting the materials into a young boy (Figure **23.7.3**). The boy developed a mild infection that included a low-grade fever, discomfort in his axillae (armpit) and loss of appetite. When the boy was later infected with infectious samples from smallpox lesions, he did not contract smallpox.[3] This new approach was termed **vaccination**, a name deriving from the use of cowpox (Latin *vacca* meaning "cow") to protect against smallpox. Today, we know that Jenner's vaccine worked because the cowpox virus is genetically and antigenically related to the *Variola* viruses that caused smallpox. Exposure to cowpox antigens resulted in a primary response and the production of memory cells that identical or related epitopes of Variola virus upon a later exposure to smallpox.

The success of Jenner's smallpox vaccination led other scientists to develop vaccines for other diseases. Perhaps the most notable was Louis Pasteur, who developed vaccines for rabies, cholera, and anthrax. During the 20th and 21st centuries, effective vaccines were developed to prevent a wide range of diseases caused by viruses (e.g., chickenpox and shingles, hepatitis, measles, mumps, polio, and yellow fever) and bacteria (e.g., diphtheria, pneumococcal pneumonia, tetanus, and whooping cough).



Figure **23.7.3**: (a) A painting of Edward Jenner depicts a cow and a milkmaid in the background. (b) Lesions on a patient infected with cowpox, a zoonotic disease caused by a virus closely related to the one that causes smallpox. (credit b: modification of work by the Centers for Disease Control and Prevention)

Key Concepts and Summary

- **Variolation** against smallpox originated in the 10th century in China, but the procedure was risky because it could cause the disease it was intended to prevent. Modern vaccination was developed by Edward Jenner, who developed the practice of inoculating patients with infectious materials from cowpox lesions to prevent smallpox.

Footnotes

1. M. Alonso and J. C. Leong. "Licensed DNA Vaccines Against Infectious Hematopoietic Necrosis Virus (IHNV)." *Recent Patents on DNA & Gene Sequences (Discontinued)* 7 no. 1 (2013): 62–65, issn 1872-2156/2212-3431. doi 10.2174/1872215611307010009.

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23.8: Classes of Vaccines

Learning Objectives

- Describe different types of vaccines
- Explain their respective advantages and disadvantages
- Identify the risk associated with a live attenuated vaccine

Classes of Vaccines

For a vaccine to provide protection against a disease, it must expose an individual to pathogen-specific antigens that will stimulate a protective adaptive immune response. By its very nature, this entails some risk. As with any pharmaceutical drug, vaccines have the potential to cause adverse effects. However, the ideal vaccine causes no severe adverse effects and poses no risk of contracting the disease that it is intended to prevent. Various types of vaccines have been developed with these goals in mind. These different classes of vaccines are described in the next section and summarized in Table **23.8.1**.

Live Attenuated Vaccines

Live attenuated vaccines expose an individual to a weakened strain of a pathogen with the goal of establishing a subclinical infection that will activate the adaptive immune defenses. Pathogens are attenuated to decrease their virulence using methods such as genetic manipulation (to eliminate key virulence factors) or long-term culturing in an unnatural host or environment (to promote mutations and decrease virulence).

By establishing an active infection, live attenuated vaccines stimulate a more comprehensive immune response than some other types of vaccines. Live attenuated vaccines activate both cellular and humoral immunity and stimulate the development of memory for long-lasting immunity. In some cases, vaccination of one individual with a live attenuated pathogen can even lead to natural transmission of the attenuated pathogen to other individuals. This can cause the other individuals to also develop an active, subclinical infection that activates their adaptive immune defenses.

Disadvantages associated with live attenuated vaccines include the challenges associated with long-term storage and transport as well as the potential for a patient to develop signs and symptoms of disease during the active infection (particularly in immunocompromised patients). There is also a risk of the attenuated pathogen reverting back to full virulence. Table **23.8.1** lists examples of live attenuated vaccines.

Inactivated Vaccines

Inactivated vaccines contain whole pathogens that have been killed or inactivated with heat, chemicals, or radiation. For inactivated vaccines to be effective, the inactivation process must not affect the structure of key antigens on the pathogen.

Because the pathogen is killed or inactive, inactivated vaccines do not produce an active infection, and the resulting immune response is weaker and less comprehensive than that provoked by a live attenuated vaccine. Typically the response involves only humoral immunity, and the pathogen cannot be transmitted to other individuals. In addition, inactivated vaccines usually require higher doses and multiple boosters, possibly causing inflammatory reactions at the site of injection.

Despite these disadvantages, inactivated vaccines do have the advantages of long-term storage stability and ease of transport. Also, there is no risk of causing severe active infections. However, inactivated vaccines are not without their side effects. Table **23.8.1** lists examples of inactivated vaccines.

Subunit Vaccines

Whereas live attenuated and inactive vaccines expose an individual to a weakened or dead pathogen, **subunit vaccines** only expose the patient to the key antigens of a pathogen—not whole cells or viruses. Subunit vaccines can be produced either by chemically degrading a pathogen and isolating its key antigens or by producing the antigens through genetic engineering. Because these vaccines contain only the essential antigens of a pathogen, the risk of side effects is relatively low. Table **23.8.1** lists examples of subunit vaccines.

Toxoid Vaccines

Like subunit vaccines, **toxoid vaccines** do not introduce a whole pathogen to the patient; they contain inactivated bacterial toxins, called toxoids. Toxoid vaccines are used to prevent diseases in which bacterial toxins play an important role in pathogenesis. These vaccines activate humoral immunity that neutralizes the toxins. Table **23.8.1** lists examples of toxoid vaccines.

Conjugate Vaccines

A **conjugate vaccine** is a type of subunit vaccine that consists of a protein conjugated to a capsule polysaccharide. Conjugate vaccines have been developed to enhance the efficacy of subunit vaccines against pathogens that have protective polysaccharide capsules that help them evade phagocytosis, causing invasive infections that can lead to meningitis and other serious conditions. The subunit vaccines against these pathogens introduce T-independent capsular polysaccharide antigens that result in the production of antibodies that can opsonize the capsule and thus combat the infection; however, children under the age of two years do not respond effectively to these vaccines. Children do respond effectively when vaccinated with the conjugate vaccine, in which a protein with T-dependent antigens is conjugated to the capsule polysaccharide. The conjugated protein-polysaccharide antigen stimulates production of antibodies against both the protein and the capsule polysaccharide. Table **23.8.1** lists examples of conjugate vaccines.

Table **23.8.1**: Classes of Vaccines

Class	Description	Advantages	Disadvantages	Examples
Live attenuated	Weakened strain of whole pathogen	Cellular and humoral immunity	Difficult to store and transport	Chickenpox, German measles, measles, mumps, tuberculosis, typhoid fever, yellow fever
		Long-lasting immunity	Risk of infection in immunocompromised patients	
		Transmission to contacts	Risk of reversion	
Inactivated	Whole pathogen killed or inactivated with heat, chemicals, or radiation	Ease of storage and transport	Weaker immunity (humoral only)	Cholera, hepatitis A, influenza, plague, rabies
		No risk of severe active infection	Higher doses and more boosters required	
Subunit	Immunogenic antigens	Lower risk of side effects	Limited longevity	Anthrax, hepatitis B, influenza, meningitis, papillomavirus, pneumococcal pneumonia, whooping cough
			Multiple doses required	
			No protection against antigenic variation	
Toxoid	Inactivated bacterial toxin	Humoral immunity to neutralize toxin	Does not prevent infection	Botulism, diphtheria, pertussis, tetanus
Conjugate	Capsule polysaccharide conjugated to protein	T-dependent response to capsule	Costly to produce	Meningitis (*Haemophilus influenzae*, *Streptococcus pneumoniae*, *Neisseria meningitides*)
			No protection against antigenic variation	
		Better response in young children	May interfere with other vaccines	

DNA Vaccines

DNA vaccines represent a relatively new and promising approach to vaccination. A DNA vaccine is produced by incorporating genes for antigens into a recombinant plasmid vaccine. Introduction of the DNA vaccine into a patient leads to uptake of the

recombinant plasmid by some of the patient's cells, followed by transcription and translation of antigens and presentation of these antigens with MHC I to activate adaptive immunity. This results in the stimulation of both humoral and cellular immunity without the risk of active disease associated with live attenuated vaccines.

Although most DNA vaccines for humans are still in development, it is likely that they will become more prevalent in the near future as researchers are working on engineering DNA vaccines that will activate adaptive immunity against several different pathogens at once. First-generation DNA vaccines tested in the 1990s looked promising in animal models but were disappointing when tested in human subjects. Poor cellular uptake of the DNA plasmids was one of the major problems impacting their efficacy. Trials of second-generation DNA vaccines have been more promising thanks to new techniques for enhancing cellular uptake and optimizing antigens. DNA vaccines for various cancers and viral pathogens such as HIV, HPV, and hepatitis B and C are currently in development.

Some DNA vaccines are already in use. In 2005, a DNA vaccine against West Nile virus was approved for use in horses in the United States. Canada has also approved a DNA vaccine to protect fish from infectious hematopoietic necrosis virus.[4] A DNA vaccine against Japanese encephalitis virus was approved for use in humans in 2010 in Australia.

Key Concepts and Summary

- **Live attenuated vaccines** and **inactivated vaccines** contain whole pathogens that are weak, killed, or inactivated. **Subunit vaccines, toxoid vaccines,** and **conjugate vaccines** contain acellular components with antigens that stimulate an immune response.

Footnotes

1. S.B. Halstead and S. J. Thomas. "New Japanese Encephalitis Vaccines: Alternatives to Production in Mouse Brain." *Expert Review of Vaccines* 10 no. 3 (2011): 355–64.

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23.E: Adaptive Immunity II (Exercises)

23.1: B Lymphocytes and Antibodies

Humoral immunity refers to mechanisms of the adaptive immune defenses that are mediated by antibodies secreted by B lymphocytes, or B cells. This section focuses on B cells and discusses their production and maturation, receptors, and mechanisms of activation.

Multiple Choice

Which of the following would be a T-dependent antigen?

A. lipopolysaccharide
B. glycolipid
C. protein
D. carbohydrate

Answer

C

Which of the following would be a BCR?

A. CD4
B. MHC II
C. MHC I
D. IgD

Answer

D

Which of the following does not occur during the lag period of the primary antibody response?

A. activation of helper T cells
B. class switching to IgG
C. presentation of antigen with MHC II
D. binding of antigen to BCRs

Answer

B

Fill in the Blank

_____ antigens can stimulate B cells to become activated but require cytokine assistance delivered by helper T cells.

Answer

T-dependent

T-independent antigens can stimulate B cells to become activated and secrete antibodies without assistance from helper T cells. These antigens possess _____ antigenic epitopes that cross-link BCRs.

Answer

repetitive

Critical Thinking

A patient lacks the ability to make functioning T cells because of a genetic disorder. Would this patient's B cells be able to produce antibodies in response to an infection? Explain your answer.

23.2: Vaccines

By artificially stimulating the adaptive immune defenses, a vaccine triggers memory cell production similar to that which would occur during a primary response. In so doing, the patient is able to mount a strong secondary response upon exposure to the pathogen—but without having to first suffer through an initial infection. In this section, we explore several different kinds of artificial immunity along with various types of vaccines and their mechanisms for inducing artificial immunity.

Multiple Choice

A patient is bitten by a dog with confirmed rabies infection. After treating the bite wound, the physician injects the patient with antibodies that are specific for the rabies virus to prevent the development of an active infection. This is an example of:

A. Natural active immunity
B. Artificial active immunity
C. Natural passive immunity
D. Artificial passive immunity

Answer

D

A patient gets a cold, and recovers a few days later. The patient's classmates come down with the same cold roughly a week later, but the original patient does not get the same cold again. This is an example of:

A. Natural active immunity
B. Artificial active immunity
C. Natural passive immunity
D. Artificial passive immunity

Answer

A

Matching

Match each type of vaccine with the corresponding example.

___inactivated vaccine	A. Weakened influenza virions that can only replicate in the slightly lower temperatures of the nasal passages are sprayed into the nose. They do not cause serious flu symptoms, but still produce an active infection that induces a protective adaptive immune response.
___live attenuated vaccine	B. Tetanus toxin molecules are harvested and chemically treated to render them harmless. They are then injected into a patient's arm.
___toxoid vaccine	C. Influenza virus particles grown in chicken eggs are harvested and chemically treated to render them noninfectious. These immunogenic particles are then purified and packaged and administered as an injection.
___subunit vaccine	D. The gene for hepatitis B virus surface antigen is inserted into a yeast genome. The modified yeast is grown and the virus protein is produced, harvested, purified, and used in a vaccine.

Answer

C, A, B, D

Fill in the Blank

A(n) _____ pathogen is in a weakened state; it is still capable of stimulating an immune response but does not cause a disease.

Answer

attenuated

_____ immunity occurs when antibodies from one individual are harvested and given to another to protect against disease or treat active disease.

Answer

Artificial passive

In the practice of _____, scabs from smallpox victims were used to immunize susceptible individuals against smallpox.

Answer

variolation

Short answer

Briefly compare the pros and cons of inactivated versus live attenuated vaccines.

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CHAPTER OVERVIEW

24: Hypersensitivites

An allergic reaction is an immune response to a type of antigen called an allergen. Allergens can be found in many different items, from peanuts and insect stings to latex and some drugs. Unlike other kinds of antigens, allergens are not necessarily associated with pathogenic microbes, and many allergens provoke no immune response at all in most people.

Allergic responses vary in severity. Some are mild and localized, like hay fever or hives, but others can result in systemic, life-threatening reactions. Anaphylaxis, for example, is a rapidly developing allergic reaction that can cause a dangerous drop in blood pressure and severe swelling of the throat that may close off the airway.

Allergies are just one example of how the immune system—the system normally responsible for preventing disease—can actually cause or mediate disease symptoms. In this chapter, we will further explore allergies and other disorders of the immune system, including hypersensitivity reactions, autoimmune diseases, transplant rejection, and diseases associated with immunodeficiency.



Figure **24.1**: Bee stings and other allergens can cause life-threatening, systemic allergic reactions. Sensitive individuals may need to carry an epinephrine auto-injector (e.g., EpiPen) in case of a sting. A bee-sting allergy is an example of an immune response that is harmful to the host rather than protective; epinephrine counteracts the severe drop in blood pressure that can result from the immune response. (credit right: modification of work by Carol Bleistine)

24.1: Type I Hypersensitivities
24.2: Type II Hypersensitivities
24.3: Type III Hypersensitivities
24.4: Type IV Hypersensitivities
24.5: Hypersensitivity Pneumonitis and Summary
24.6: Diagnosis and Treatment of Hypersensitivities
24.E: Diseases of the Immune System (Exercises)

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24.1: Type I Hypersensitivities

Learning Objectives

- Compare the distinguishing characteristics, mechanisms, and major examples of type I, II, III, and IV hypersensitivities
- Identify the cells that cause a type I hypersensitivity reaction
- Describe the differences between immediate and late-phase type I hypersensitivity reactions
- List the signs and symptoms of anaphylaxis

In Adaptive Specific Host Defenses, we discussed the mechanisms by which adaptive immune defenses, both humoral and cellular, protect us from infectious diseases. However, these same protective immune defenses can also be responsible for undesirable reactions called **hypersensitivity** reactions. Hypersensitivity reactions are classified by their immune mechanism.

- Type I hypersensitivity reactions involve immunoglobulin E (IgE) antibody against soluble antigen, triggering mast cell degranulation.
- Type II hypersensitivity reactions involve IgG and IgM antibodies directed against cellular antigens, leading to cell damage mediated by other immune system effectors.
- Type III hypersensitivity reactions involve the interactions of IgG, IgM, and, occasionally, IgA[1] antibodies with antigen to form immune complexes. Accumulation of immune complexes in tissue leads to tissue damage mediated by other immune system effectors.
- Type IV hypersensitivity reactions are T-cell–mediated reactions that can involve tissue damage mediated by activated macrophages and cytotoxic T cells.

Type I Hypersensitivities

When a presensitized individual is exposed to an **allergen**, it can lead to a rapid immune response that occurs almost immediately. Such a response is called an **allergy** and is classified as a **type I hypersensitivity**. Allergens may be seemingly harmless substances such as animal dander, molds, or pollen. Allergens may also be substances considered innately more hazardous, such as insect venom or therapeutic drugs. Food intolerances can also yield allergic reactions as individuals become sensitized to foods such as peanuts or shellfish (Figure **24.1.1**). Regardless of the allergen, the first exposure activates a primary IgE antibody response that sensitizes an individual to type I hypersensitivity reaction upon subsequent exposure.



Figure **24.1.1**: (a) Allergens in plant pollen, shown here in a colorized electron micrograph, may trigger allergic rhinitis or hay fever in sensitive individuals. (b) Skin rashes are often associated with allergic reactions. (c) Peanuts can be eaten safely by most people but can provoke severe allergic reactions in sensitive individuals.

Mechanism of Type I Hypersensitivity

For susceptible individuals, a first exposure to an allergen activates a strong T_H2 cell response (Figure **24.1.2**). Cytokines interleukin (IL)-4 and IL-13 from the T_H2 cells activate B cells specific to the same allergen, resulting in clonal proliferation, differentiation into plasma cells, and antibody-class switch from production of IgM to production of IgE. The fragment

crystallizable (Fc) regions of the IgE antibodies bind to specific receptors on the surface of mast cellsthroughout the body. It is estimated that each mast cell can bind up to 500,000 IgE molecules, with each IgE molecule having two allergen-specific fragment antigen-binding (Fab) sites available for binding allergen on subsequent exposures. By the time this occurs, the allergen is often no longer present and there is no allergic reaction, but the mast cells are primed for a subsequent exposure and the individual is sensitized to the allergen.

On subsequent exposure, allergens bind to multiple IgE molecules on mast cells, cross-linking the IgE molecules. Within minutes, this cross-linking of IgE activates the mast cells and triggers **degranulation**, a reaction in which the contents of the granules in the mast cell are released into the extracellular environment. Preformed components that are released from granules include histamine, serotonin, and bradykinin (Table **24.1.1**). The activated mast cells also release newly formed lipid mediators (leukotrienes and prostaglandins from membrane arachadonic acid metabolism) and cytokines such as tumor necrosis factor (Table **24.1.2**).

Table **24.1.1**: Selected Preformed Components of Mast Cell Granules

Granule Component	Activity
Heparin	Stimulates the generation of bradykinin, which causes increased vascular permeability, vasodilation, bronchiole constriction, and increased mucus secretion
Histamine	Causes smooth-muscle contraction, increases vascular permeability, increases mucus and tear formation
Serotonin	Increases vascular permeability, causes vasodilation and smooth-muscle contraction

The chemical mediators released by mast cells collectively cause the inflammation and signs and symptoms associated with type I hypersensitivity reactions. Histamine stimulates mucus secretion in nasal passages and tear formation from lacrimal glands, promoting the runny nose and watery eyes of allergies. Interaction of histamine with nerve endings causes itching and sneezing. The vasodilation caused by several of the mediators can result in hives, headaches, angioedema (swelling that often affects the lips, throat, and tongue), and hypotension (low blood pressure). Bronchiole constriction caused by some of the chemical mediators leads to wheezing, dyspnea (difficulty breathing), coughing, and, in more severe cases, cyanosis (bluish color to the skin or mucous membranes). Vomiting can result from stimulation of the vomiting center in the cerebellum by histamine and serotonin. Histamine can also cause relaxation of intestinal smooth muscles and diarrhea.

Table **24.1.2**: Selected Newly Formed Chemical Mediators of Inflammation and Allergic Response

Chemical Mediator	**Activity**
Leukotriene	Causes smooth-muscle contraction and mucus secretion, increases vascular permeability
Prostaglandin	Causes smooth-muscle contraction and vasodilation
TNF-α (cytokine)	Causes inflammation and stimulates cytokine production by other cell types

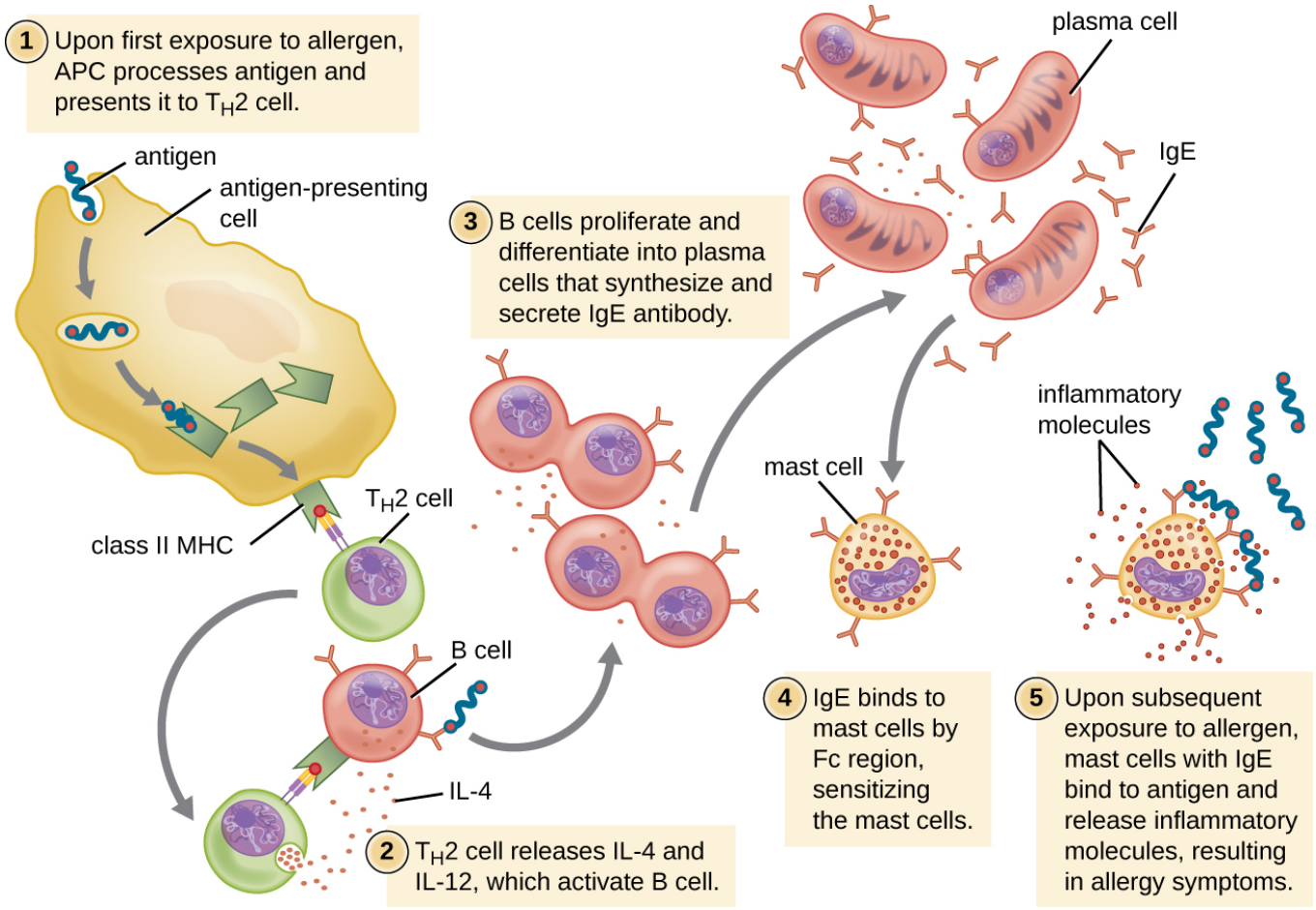


Figure **24.1.2**: On first exposure to an allergen in a susceptible individual, antigen-presenting cells process and present allergen epitopes with major histocompatibility complex (MHC) II to T helper cells. B cells also process and present the same allergen epitope to T_H2 cells, which release cytokines IL-4 and IL-13 to stimulate proliferation and differentiation into IgE-secreting plasma cells. The IgE molecules bind to mast cells with their Fc region, sensitizing the mast cells for activation with subsequent exposure to the allergen. With each subsequent exposure, the allergen cross-links IgE molecules on the mast cells, activating the mast cells and causing the release of preformed chemical mediators from granules (degranulation), as well as newly formed chemical mediators that collectively cause the signs and symptoms of type I hypersensitivity reactions.

Types of Type I Hypersensitivity

Type I hypersensitivity reactions can be either localized or systemic. **Localized type I hypersensitivity** reactions include hay fever rhinitis, hives, and asthma (Table **24.1.3**). **Systemic type I hypersensitivity** reactions are referred to as **anaphylaxis** or **anaphylactic shock**. Although anaphylaxis shares many symptoms common with the localized type I hypersensitivity reactions, the swelling of the tongue and trachea, blockage of airways, dangerous drop in blood pressure, and development of shock can make anaphylaxis especially severe and life-threatening. In fact, death can occur within minutes of onset of signs and symptoms.

Late-phase reactions in type I hypersensitivities may develop 4–12 hours after the early phase and are mediated by eosinophils, neutrophils, and lymphocytes that have been recruited by chemotactic factors released from mast cells. Activation of these recruited cells leads to the release of more chemical mediators that cause tissue damage and late-phase symptoms of swelling and redness of the skin, coughing, wheezing, and nasal discharge.

Individuals who possess genes for maladaptive traits, such as intense type I hypersensitivity reactions to otherwise harmless components of the environment, would be expected to suffer reduced reproductive success. With this kind of evolutionary selective pressure, such traits would not be expected to persist in a population. This suggests that type I hypersensitivities may have an adaptive function. There is evidence that the IgE produced during type I hypersensitivity reactions is actually meant to counter helminth infections[2]. Helminths are one of few organisms that possess proteins that are targeted by IgE. In addition, there is

evidence that helminth infections at a young age reduce the likelihood of type I hypersensitivities to innocuous substances later in life. Thus it may be that allergies are an unfortunate consequence of strong selection in the mammalian lineage or earlier for a defense against parasitic worms.

Table **24.1.3**: Type I Hypersensitivities

Common Name	Cause	Signs and Symptoms
Allergy-induced asthma	Inhalation of allergens	Constriction of bronchi, labored breathing, coughing, chills, body aches
Anaphylaxis	Systemic reaction to allergens	Hives, itching, swelling of tongue and throat, nausea, vomiting, low blood pressure, shock
Hay fever	Inhalation of mold or pollen	Runny nose, watery eyes, sneezing
Hives (urticaria)	Food or drug allergens, insect stings	Raised, bumpy skin rash with itching; bumps may converge into large raised areas

The Hygiene Hypothesis

In most modern societies, good hygiene is associated with regular bathing, and good health with cleanliness. But some recent studies suggest that the association between health and clean living may be a faulty one. Some go so far as to suggest that children should be encouraged to play in the dirt—or even eat dirt[3]—for the benefit of their health. This recommendation is based on the so-called hygiene hypothesis, which proposes that childhood exposure to antigens from a diverse range of microbes leads to a better-functioning immune system later in life.

The hygiene hypothesis was first suggested in 1989 by David Strachan[4], who observed an inverse relationship between the number of older children in a family and the incidence of hay fever. Although hay fever in children had increased dramatically during the mid-20th century, incidence was significantly lower in families with more children. Strachan proposed that the lower incidence of allergies in large families could be linked to infections acquired from older siblings, suggesting that these infections made children less susceptible to allergies. Strachan also argued that trends toward smaller families and a greater emphasis on cleanliness in the 20th century had decreased exposure to pathogens and thus led to higher overall rates of allergies, asthma, and other immune disorders.

Other researchers have observed an inverse relationship between the incidence of immune disorders and infectious diseases that are now rare in industrialized countries but still common in less industrialized countries.[5] In developed nations, children under the age of 5 years are not exposed to many of the microbes, molecules, and antigens they almost certainly would have encountered a century ago. The lack of early challenges to the immune system by organisms with which humans and their ancestors evolved may result in failures in immune system functioning later in life.

Key Concepts and Summary

- An **allergy** is an adaptive immune response, sometimes life-threatening, to an **allergen**.
- **Type I hypersensitivity** requires sensitization of mast cells with IgE, involving an initial IgE antibody response and IgE attachment to mast cells. On second exposure to an allergen, cross-linking of IgE molecules on mast cells triggers degranulation and release of preformed and newly formed chemical mediators of inflammation. Type I hypersensitivity may be localized and relatively minor (hives and hay fever) or system-wide and dangerous (systemic **anaphylaxis**).

Footnotes

1. 1 D.S. Strayer et al (eds). *Rubin's Pathology: Clinicopathologic Foundations of Medicine*. 7th ed. 2Philadelphia, PA: Lippincott, Williams & Wilkins, 2014.

2. 2 C.M. Fitzsimmons et al. “Helminth Allergens, Parasite-Specific IgE, and Its Protective Role in Human Immunity.” *Frontier in Immunology* 5 (2015):47.
3. 3 S.T. Weiss. “Eat Dirt—The Hygiene Hypothesis and Allergic Diseases.” *New England Journal of Medicine* 347 no. 12 (2002):930–931.
4. D.P. Strachan “Hay Fever, Hygiene, and Household Size.” *British Medical Journal* 299 no. 6710 (1989):1259.
5. H. Okada et al. “The ‘Hygiene Hypothesis’ for Autoimmune and Allergic Diseases: An Update.” *Clinical & Experimental Immunology* 160 no. 1 (2010):1–9.

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24.2: Type II Hypersensitivities

Learning Objectives

- Summarize the ABO blood group system
- Explain how hemolytic transfusion reaction (HTR) occurs
- Describe hemolytic disease of the newborn (HDN)
- Explain how HDN can be prevented

Type II (Cytotoxic) Hypersensitivities

Immune reactions categorized as **type II hypersensitivities**, or **cytotoxic hypersensitivities**, are mediated by IgG and IgM antibodies binding to cell-surface antigens or matrix-associated antigens on basement membranes. These antibodies can either activate complement, resulting in an inflammatory response and lysis of the targeted cells, or they can be involved in antibody-dependent cell-mediated cytotoxicity (ADCC) with cytotoxic T cells.

In some cases, the antigen may be a self-antigen, in which case the reaction would also be described as an autoimmune disease. (Autoimmune diseases are described in Autoimmune Disorders). In other cases, antibodies may bind to naturally occurring, but exogenous, cell-surface molecules such as antigens associated with blood typing found on red blood cells (RBCs). This leads to the coating of the RBCs by antibodies, activation of the complement cascade, and complement-mediated lysis of RBCs, as well as opsonization of RBCs for phagocytosis. Two examples of type II hypersensitivity reactions involving RBCs are hemolytic transfusion reaction (HTR) and hemolytic disease of the newborn (HDN). These type II hypersensitivity reactions, which will be discussed in greater detail, are summarized in Table **24.2.4**.

Immunohematology is the study of blood and blood-forming tissue in relation to the immune response. Antibody-initiated responses against blood cells are type II hypersensitivities, thus falling into the field of immunohematology. For students first learning about immunohematology, understanding the immunological mechanisms involved is made even more challenging by the complex nomenclature system used to identify different blood-group antigens, often called blood types. The first blood-group antigens either used alphabetical names or were named for the first person known to produce antibodies to the red blood cell antigen (e.g., Kell, Duffy, or Diego). However, in 1980, the International Society of Blood Transfusion (ISBT) Working Party on Terminology created a standard for blood-group terminology in an attempt to more consistently identify newly discovered blood group antigens. New antigens are now given a number and assigned to a blood-group system, collection, or series. However, even with this effort, blood-group nomenclature is still inconsistent.

Table **24.2.4**: Common Type II Hypersensitivities

Common Name	Cause	Signs and Symptoms
Hemolytic disease of the newborn (HDN)	IgG from mother crosses the placenta, targeting the fetus' RBCs for destruction	Anemia, edema, enlarged liver or spleen, hydrops (fluid in body cavity), leading to death of newborn in severe cases
Hemolytic transfusion reactions (HTR)	IgG and IgM bind to antigens on transfused RBCs, targeting donor RBCs for destruction	Fever, jaundice, hypotension, disseminated intravascular coagulation, possibly leading to kidney failure and death

ABO Blood Group Incompatibility

The recognition that individuals have different blood types was first described by Karl Landsteiner (1868–1943) in the early 1900s, based on his observation that serum from one person could cause a clumping of RBCs from another. These studies led Landsteiner to the identification of four distinct blood types. Subsequent research by other scientists determined that the four blood types were based on the presence or absence of surface glycoproteins "A" and "B," and this provided the foundation for the **ABO blood group system** that is still in use today (Figure **24.2.3**). The functions of these antigens are unknown, but some have been associated with normal biochemical functions of the cell. Furthermore, ABO blood types are inherited as alleles (one from each parent), and they display patterns of dominant and codominant inheritance. The alleles for A and B blood types are codominant to each other, and

both are dominant over blood type O. Therefore, individuals with genotypes of AA or AO have type A blood and express the A glycoprotein antigen on the surface of their RBCs. People with genotypes of BB or BO have type B blood and express the B glycoprotein antigen on the surface of their RBCs. Those with a genotype of AB have type AB blood and express both A and B glycoprotein antigens on the surface of their RBCs. Finally, individuals with a genotype of OO have type O blood and lack A and B glycoproteins on the surface of their RBCs.

It is important to note that the RBCs of all four ABO blood types share a common protein receptor molecule, and it is the addition of specific carbohydrates to the protein receptors that determines A, B, and AB blood types. The genes that are inherited for the A, B, and AB blood types encode enzymes that add the carbohydrate component to the protein receptor. Individuals with O blood type still have the protein receptor but lack the enzymes that would add carbohydrates that would make their red blood cell type A, B, or AB.

IgM antibodies in plasma that cross-react with blood group antigens not present on an individual's own RBCs are called **isohemagglutinins** (Figure **24.2.3**). Isohemagglutinins are produced within the first few weeks after birth and persist throughout life. These antibodies are produced in response to exposure to environmental antigens from food and microorganisms. A person with type A blood has A antigens on the surface of their RBCs and will produce anti-B antibodies to environmental antigens that resemble the carbohydrate component of B antigens. A person with type B blood has B antigens on the surface of their RBCs and will produce anti-A antibodies to environmental antigens that are similar to the carbohydrate component of A antigens. People with blood type O lack both A and B antigens on their RBCs and, therefore, produce both anti-A and anti-B antibodies. Conversely, people with AB blood type have both A and B antigens on their RBCs and, therefore, lack anti-A and anti-B antibodies.

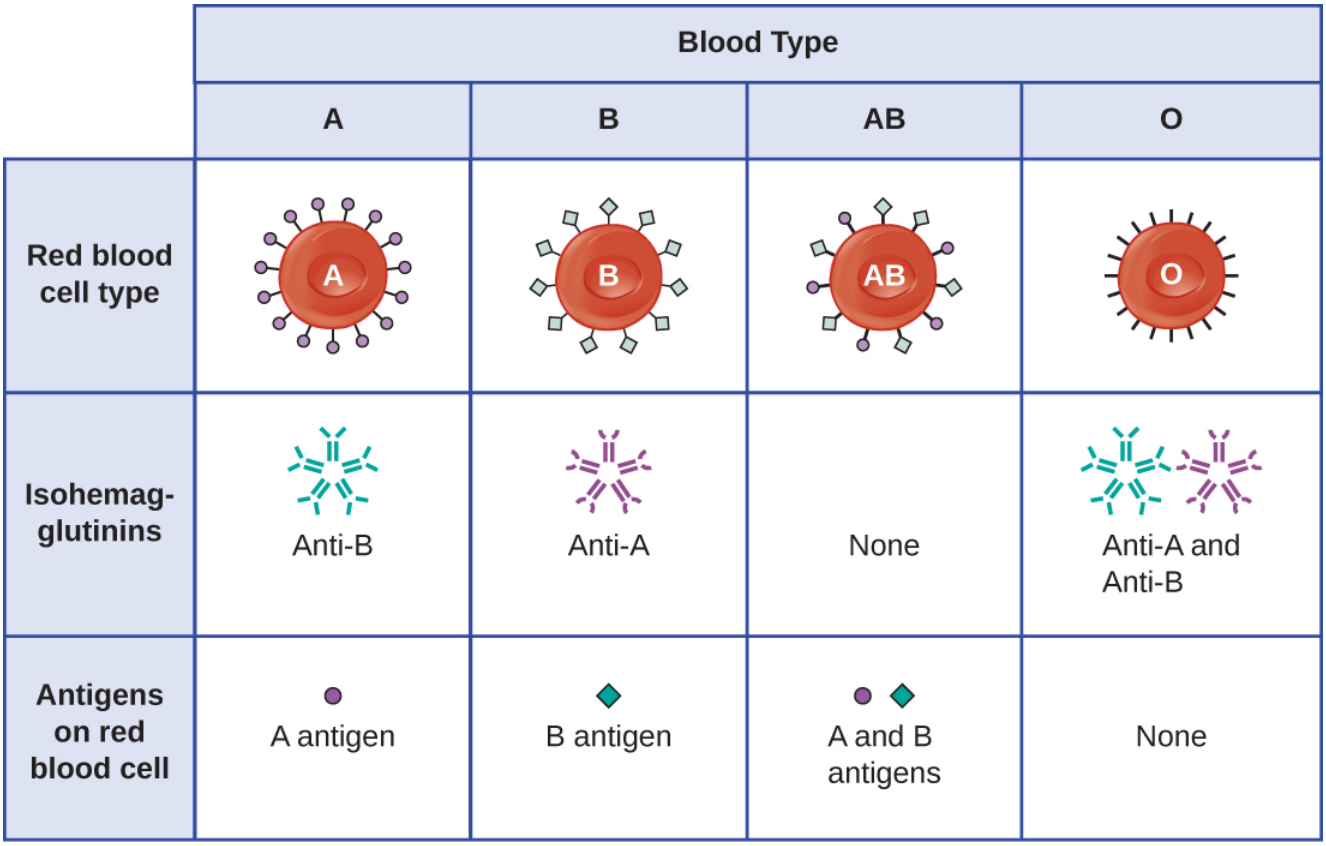


Figure **24.2.3**: Isohemagglutinins

A patient may require a blood transfusion because they lack sufficient RBCs (anemia) or because they have experienced significant loss of blood volume through trauma or disease. Although the blood transfusion is given to help the patient, it is essential that the patient receive a transfusion with matching ABO blood type. A transfusion with an incompatible ABO blood type may lead to a strong, potentially lethal type II hypersensitivity cytotoxic response called **hemolytic transfusion reaction (HTR)** (Figure **24.2.4**).

For instance, if a person with type B blood receives a transfusion of type A blood, their anti-A antibodies will bind to and agglutinate the transfused RBCs. In addition, activation of the classical complement cascade will lead to a strong inflammatory response, and the complement membrane attack complex (MAC) will mediate massive hemolysis of the transfused RBCs. The debris from damaged and destroyed RBCs can occlude blood vessels in the alveoli of the lungs and the glomeruli of the kidneys. Within 1 to 24 hours of an incompatible transfusion, the patient experiences fever, chills, pruritus (itching), urticaria (hives), dyspnea, hemoglobinuria (hemoglobin in the urine), and hypotension (low blood pressure). In the most serious reactions, dangerously low blood pressure can lead to shock, multi-organ failure, and death of the patient.

Hospitals, medical centers, and associated clinical laboratories typically use hemovigilance systems to minimize the risk of HTRs due to clerical error. Hemovigilance systems are procedures that track transfusion information from the donor source and blood products obtained to the follow-up of recipient patients. Hemovigilance systems used in many countries identify HTRs and their outcomes through mandatory reporting (e.g., to the Food and Drug Administration in the United States), and this information is valuable to help prevent such occurrences in the future. For example, if an HTR is found to be the result of laboratory or clerical error, additional blood products collected from the donor at that time can be located and labeled correctly to avoid additional HTRs. As a result of these measures, HTR-associated deaths in the United States occur in about one per 2 million transfused units.[6]

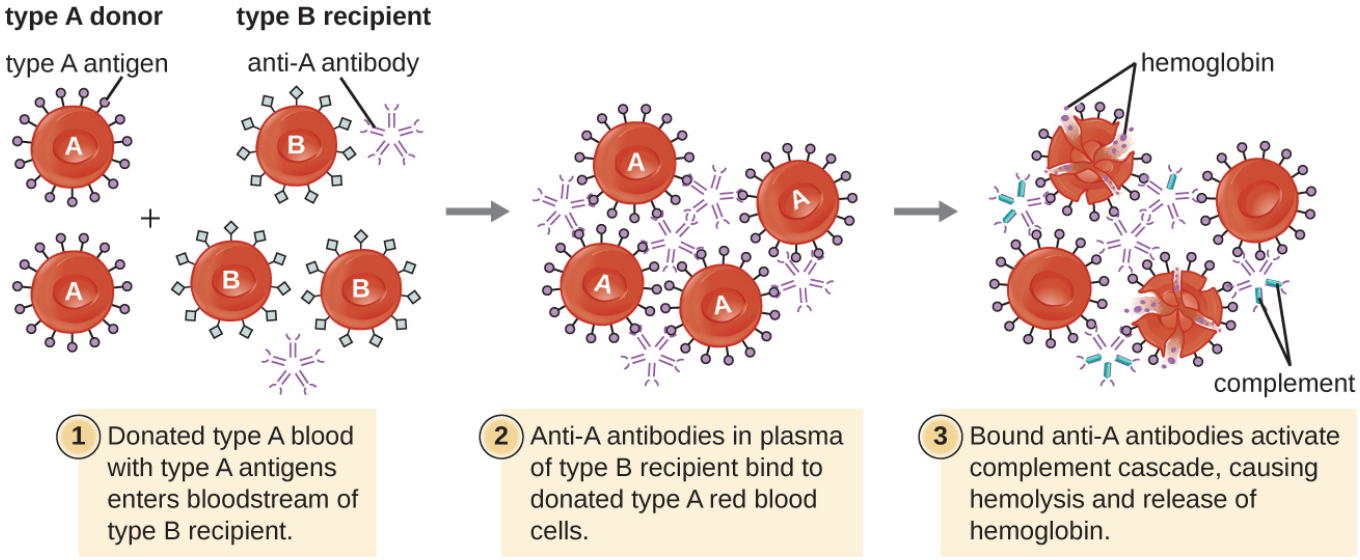


Figure **24.2.4**: A type II hypersensitivity hemolytic transfusion reaction (HTR) leading to hemolytic anemia. Blood from a type A donor is administered to a patient with type B blood. The anti-A isohemagglutinin IgM antibodies in the recipient bind to and agglutinate the incoming donor type A red blood cells. The bound anti-A antibodies activate the classical complement cascade, resulting in destruction of the donor red blood cells.

Rh Factors

Many different types of erythrocyte antigens have been discovered since the description of the ABO red cell antigens. The second most frequently described RBC antigens are **Rh factors**, named after the rhesus macaque (*Macaca mulatta*) factors identified by Karl Landsteiner and Alexander Weiner in 1940. The Rh system of RBC antigens is the most complex and immunogenic blood group system, with more than 50 specificities identified to date. Of all the Rh antigens, the one designated Rho (Weiner) or D (Fisher-Race) is the most immunogenic. Cells are classified as Rh positive (Rh+) if the Rho/D antigen is present or as Rh negative (Rh−) if the Rho/D antigen is absent. In contrast to the carbohydrate molecules that distinguish the ABO blood groups and are the targets of IgM isohemagglutinins in HTRs, the Rh factor antigens are proteins. As discussed in B Lymphocytes and Humoral Immunity, protein antigens activate B cells and antibody production through a T-cell–dependent mechanism, and the T_H2 cells stimulate class switching from IgM to other antibody classes. In the case of Rh factor antigens, T_H2 cells stimulate class switching to IgG, and this has important implications for the mechanism of HDN.

Like ABO incompatibilities, blood transfusions from a donor with the wrong Rh factor antigens can cause a type II hypersensitivity HTR. However, in contrast to the IgM isohemagglutinins produced early in life through exposure to environmental antigens, production of anti-Rh factor antibodies requires the exposure of an individual with Rh− blood to Rh+ positive RBCs and activation of a primary antibody response. Although this primary antibody response can cause an HTR in the transfusion patient, the

hemolytic reaction would be delayed up to 2 weeks during the extended lag period of a primary antibody response (B Lymphocytes and Humoral Immunity). However, if the patient receives a subsequent transfusion with Rh+ RBCs, a more rapid HTR would occur with anti-Rh factor antibody already present in the blood. Furthermore, the rapid secondary antibody response would provide even more anti-Rh factor antibodies for the HTR.

Rh factor incompatibility between mother and fetus can also cause a type II hypersensitivity hemolytic reaction, referred to as **hemolytic disease of the newborn (HDN)** (Figure 24.2.5). If an Rh− woman carries an Rh+ baby to term, the mother's immune system can be exposed to Rh+ fetal red blood cells. This exposure will usually occur during the last trimester of pregnancy and during the delivery process. If this exposure occurs, the Rh+ fetal RBCs will activate a primary adaptive immune response in the mother, and anti-Rh factor IgG antibodies will be produced. IgG antibodies are the only class of antibody that can cross the placenta from mother to fetus; however, in most cases, the first Rh+ baby is unaffected by these antibodies because the first exposure typically occurs late enough in the pregnancy that the mother does not have time to mount a sufficient primary antibody response before the baby is born.

If a subsequent pregnancy with an Rh+ fetus occurs, however, the mother's second exposure to the Rh factor antigens causes a strong secondary antibody response that produces larger quantities of anti-Rh factor IgG. These antibodies can cross the placenta from mother to fetus and cause HDN, a potentially lethal condition for the baby (Figure 24.2.5).

Prior to the development of techniques for diagnosis and prevention, Rh factor incompatibility was the most common cause of HDN, resulting in thousands of infant deaths each year worldwide.[7] For this reason, the Rh factors of prospective parents are regularly screened, and treatments have been developed to prevent HDN caused by Rh incompatibility. To prevent Rh factor-mediated HDN, human Rho(D) immune globulin (e.g., RhoGAM) is injected intravenously or intramuscularly into the mother during the 28th week of pregnancy and within 72 hours after delivery. Additional doses may be administered after events that may result in transplacental hemorrhage (e.g., umbilical blood sampling, chorionic villus sampling, abdominal trauma, amniocentesis). This treatment is initiated during the first pregnancy with an Rh+ fetus. The anti-Rh antibodies in Rho(D) immune globulin will bind to the Rh factor of any fetal RBCs that gain access to the mother's bloodstream, preventing these Rh+ cells from activating the mother's primary antibody response. Without a primary anti-Rh factor antibody response, the next pregnancy with an Rh+ will have minimal risk of HDN. However, the mother will need to be retreated with Rho(D) immune globulin during that pregnancy to prevent a primary anti-Rh antibody response that could threaten subsequent pregnancies.

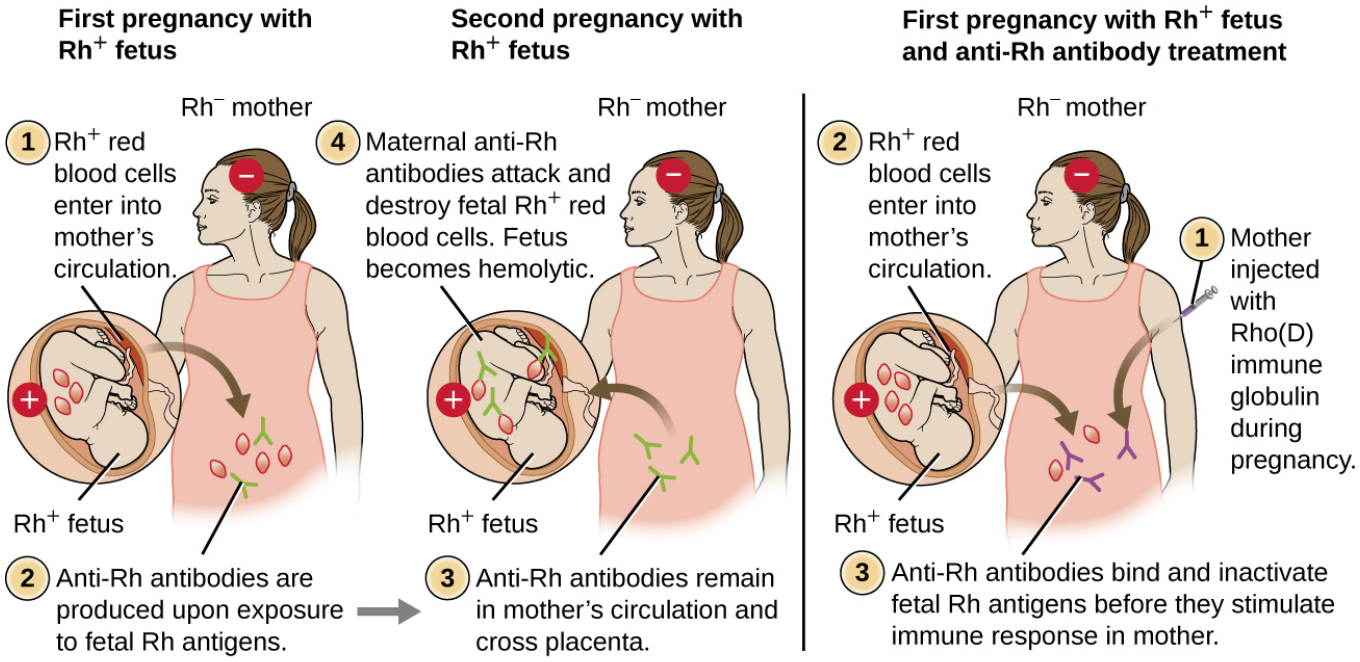


Figure 24.2.5: (a) When an Rh− mother has an Rh+ fetus, fetal erythrocytes are introduced into the mother's circulatory system before or during birth, leading to production of anti-Rh IgG antibodies. These antibodies remain in the mother and, if she becomes pregnant with a second Rh+ baby, they can cross the placenta and attach to fetal Rh+ erythrocytes. Complement-mediated hemolysis of fetal erythrocytes results in a lack of sufficient cells for proper oxygenation of the fetus. (b) HDN can be prevented by administering Rho(D) immune globulin during and after each pregnancy with an Rh+ fetus. The immune globulin binds fetal Rh+ RBCs that gain access to the mother's bloodstream, preventing activation of her primary immune response.

Link to Learning

Use this interactive Blood Typing Game to reinforce your knowledge of blood typing.

Key Concepts and Summary

- **Type II hypersensitivities** result from antibodies binding to antigens on cells and initiating cytotoxic responses. Examples include **hemolytic transfusion reaction** and **hemolytic disease of the newborn**

Footnotes

1. E.C. Vamvakas, M.A. Blajchman. "Transfusion-Related Mortality: The Ongoing Risks of Allogeneic Blood Transfusion and the Available Strategies for Their Prevention." *Blood* 113 no. 15 (2009):3406–3417.
2. G. Reali. "Forty Years of Anti-D Immunoprophylaxis." *Blood Transfusion* 5 no. 1 (2007):3–6.

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24.3: Type III Hypersensitivities

Learning Objectives

- Describe type III hypersensitivity and the different types
- Explain why antibody excess is important in type III hypersensitivity
- Compare and contrast Arthus reaction versus serium sickness

Type III Hypersensitivities

Type III hypersensitivities are immune-complex reactions that were first characterized by Nicolas Maurice Arthus (1862–1945) in 1903. To produce antibodies for experimental procedures, Arthus immunized rabbits by injecting them with serum from horses. However, while immunizing rabbits repeatedly with horse serum, Arthus noticed a previously unreported and unexpected localized subcutaneous hemorrhage with edema at the site of injection. This reaction developed within 3 to10 hours after injection. This localized reaction to non-self serum proteins was called an **Arthus reaction**. An Arthus reaction occurs when soluble antigens bind with IgG in a ratio that results in the accumulation of antigen-antibody aggregates called **immune complexes**.

A unique characteristic of type III hypersensitivity is antibody excess (primarily IgG), coupled with a relatively low concentration of antigen, resulting in the formation of small immune complexes that deposit on the surface of the epithelial cells lining the inner lumen of small blood vessels or on the surfaces of tissues (Figure **24.3.6**). This immune complex accumulation leads to a cascade of inflammatory events that include the following:

1. IgG binding to antibody receptors on localized mast cells, resulting in mast-cell degranulation
2. Complement activation with production of pro-inflammatory C3a and C5a (see Chemical Defenses)
3. Increased blood-vessel permeability with chemotactic recruitment of neutrophils and macrophages

Because these immune complexes are not an optimal size and are deposited on cell surfaces, they cannot be phagocytosed in the usual way by neutrophils and macrophages, which, in turn, are often described as “frustrated.” Although phagocytosis does not occur, neutrophil degranulation results in the release of lysosomal enzymes that cause extracellular destruction of the immune complex, damaging localized cells in the process. Activation of coagulation pathways also occurs, resulting in thrombi (blood clots) that occlude blood vessels and cause ischemia that can lead to vascular necrosis and localized hemorrhage.

Systemic type III hypersensitivity (serum sickness) occurs when immune complexes deposit in various body sites, resulting in a more generalized systemic inflammatory response. These immune complexes involve non-self proteins such as antibodies produced in animals for artificial passive immunity (see Vaccines), certain drugs, or microbial antigens that are continuously released over time during chronic infections (e.g., subacute bacterial endocarditis, chronic viral hepatitis). The mechanisms of serum sickness are similar to those described in localized type III hypersensitivity but involve widespread activation of mast cells, complement, neutrophils, and macrophages, which causes tissue destruction in areas such as the kidneys, joints, and blood vessels. As a result of tissue destruction, symptoms of serum sickness include chills, fever, rash, vasculitis, and arthritis. Development of glomerulonephritis or hepatitis is also possible.

Autoimmune diseases such as systemic lupus erythematosus (SLE) and rheumatoid arthritis can also involve damaging type III hypersensitivity reactions when auto-antibodies form immune complexes with self antigens. These conditions are discussed in Autoimmune Disorders.

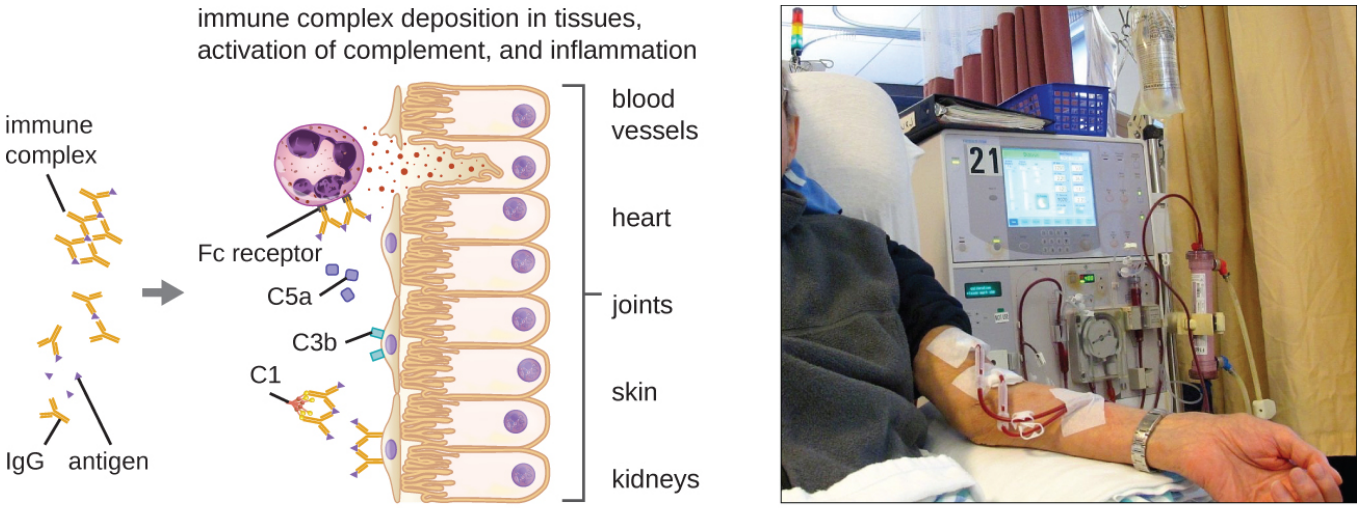


Figure **24.3.6**: Type III hypersensitivities and the systems they affect. (a) Immune complexes form and deposit in tissue. Complement activation, stimulation of an inflammatory response, and recruitment and activation of neutrophils result in damage to blood vessels, heart tissue, joints, skin, and/or kidneys. (b) If the kidneys are damaged by a type III hypersensitivity reaction, dialysis may be required.

Diphtheria Antitoxin

Antibacterial sera are much less commonly used now than in the past, having been replaced by toxoid vaccines. However, a diphtheria antitoxin produced in horses is one example of such a treatment that is still used in some parts of the world. Although it is not licensed by the FDA for use in the United States, diphtheria antitoxin can be used to treat cases of diphtheria, which are caused by the bacterium *Corynebacterium diphtheriae*.[1] The treatment is not without risks, however. Serum sickness can occur when the patient develops an immune response to non-self horse proteins. Immune complexes are formed between the horse proteins and circulating antibodies when the two exist in certain proportions. These immune complexes can deposit in organs, causing damage such as arthritis, nephritis, rash, and fever. Serum sickness is usually transient with no permanent damage unless the patient is chronically exposed to the antigen, which can then result in irreversible damage to body sites such as joints and kidneys. Over time, phagocytic cells such as macrophages are able to clear the horse serum antigens, which results in improvement of the patient's condition and a decrease in symptoms as the immune response dissipates.

Key Concepts and Summary

- **Type III hypersensitivities** result from formation and accumulation of **immune complexes** in tissues, stimulating damaging inflammatory responses.

Footnotes

1. Centers for Disease Control and Prevention. "Diphtheria Antitoxin." http://www.cdc.gov/diphtheria/dat.html. Accessed March 25, 2016.

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24.4: Type IV Hypersensitivities

Learning Objectives

- Describe the three subtypes of type IV hypersensitivity
- Explain how T cells contribute to tissue damage in type IV hypersensitivity

Type IV Hypersensitivities

Type IV hypersensitivities are not mediated by antibodies like the other three types of hypersensitivities. Rather, type IV hypersensitivities are regulated by **T cells** and involve the action of effector cells. These types of hypersensitivities can be organized into three subcategories based on T-cell subtype, type of antigen, and the resulting effector mechanism (Table **24.4.5**).

In the first type IV subcategory, CD4 T_H1-mediated reactions are described as delayed-type hypersensitivities (DTH). The sensitization step involves the introduction of antigen into the skin and phagocytosis by local antigen presenting cells (APCs). The APCs activate helper T cells, stimulating clonal proliferation and differentiation into memory T_H1 cells. Upon subsequent exposure to the antigen, these sensitized memory T_H1 cells release cytokines that activate macrophages, and activated macrophages are responsible for much of the tissue damage. Examples of this T_H1-mediated hypersensitivity are observed in tuberculin the Mantoux skin test and **contact dermatitis**, such as occurs in latex allergy reactions.

In the second type IV subcategory, CD4 T_H2-mediated reactions result in chronic asthma or chronic allergic rhinitis. In these cases, the soluble antigen is first inhaled, resulting in eosinophil recruitment and activation with the release of cytokines and inflammatory mediators.

In the third type IV subcategory, CD8 cytotoxic T lymphocyte (CTL)-mediated reactions are associated with tissue transplant rejection and contact dermatitis (Figure **24.4.7**). For this form of cell-mediated hypersensitivity, APCs process and present the antigen with MHC I to naïve CD8 T cells. When these naïve CD8 T cells are activated, they proliferate and differentiate into CTLs. Activated T_H1 cells can also enhance the activation of the CTLs. The activated CTLs then target and induce granzyme-mediated apoptosis in cells presenting the same antigen with MHC I. These target cells could be “self” cells that have absorbed the foreign antigen (such as with contact dermatitis due to poison ivy), or they could be transplanted tissue cells displaying foreign antigen from the donor.

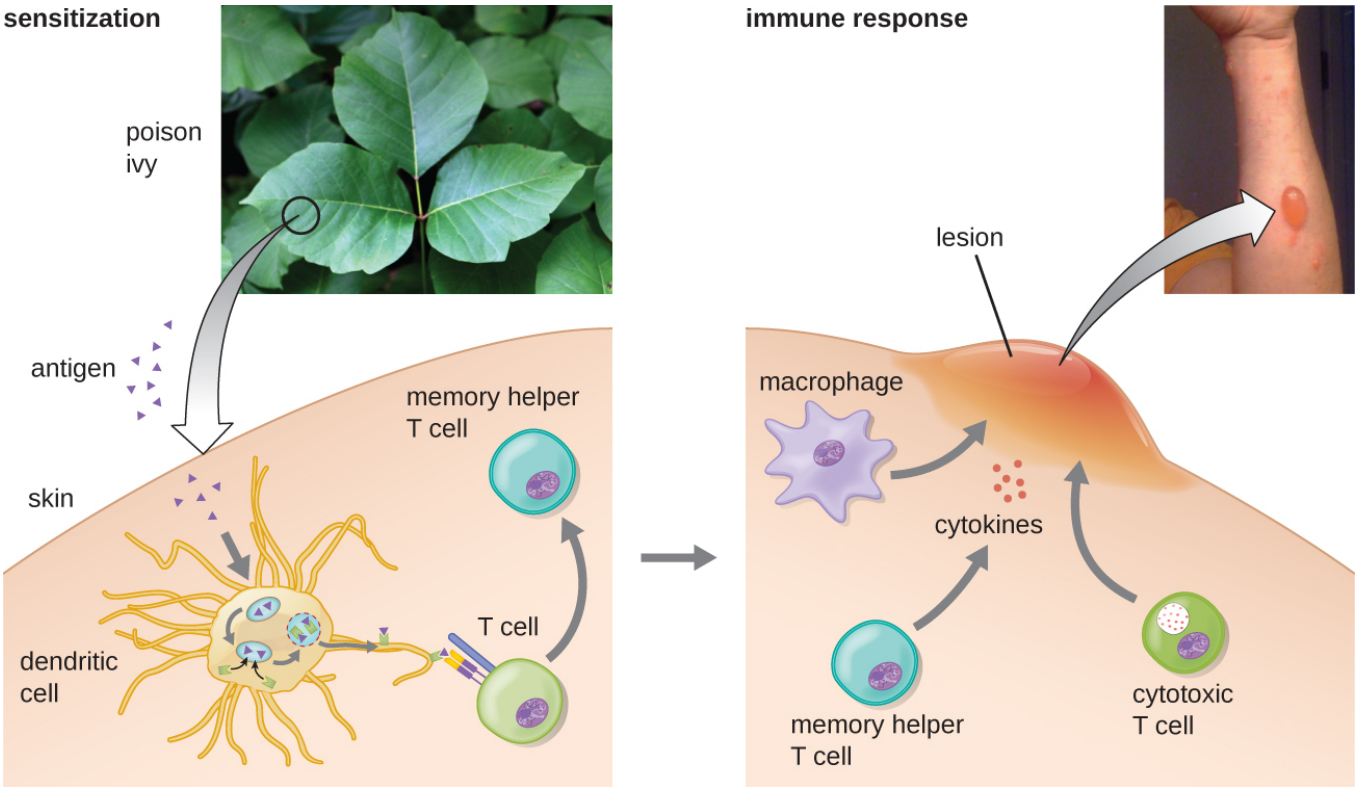


Figure **24.4.7**: Exposure to hapten antigens in poison ivy can cause contact dermatitis, a type IV hypersensitivity. (a) The first exposure to poison ivy does not result in a reaction. However, sensitization stimulates helper T cells, leading to production of memory helper T cells that can become reactivated on future exposures. (b) Upon secondary exposure, the memory helper T cells become reactivated, producing inflammatory cytokines that stimulate macrophages and cytotoxic T cells to induce an inflammatory lesion at the exposed site. This lesion, which will persist until the allergen is removed, can inflict significant tissue damage if it continues long enough.

Table **24.4.5**: Type IV Hypersensitivities

Subcategory	Antigen	Effector Mechanism	Examples
1	Soluble antigen	Activated macrophages damage tissue and promote inflammatory response	Contact dermatitis (e.g., exposure to latex) and delayed-type hypersensitivity (e.g., tuberculin reaction)
2	Soluble antigen	Eosinophil recruitment and activation release cytokines and pro-inflammatory chemicals	Chronic asthma and chronic allergic rhinitis
3	Cell-associated antigen	CTL-mediated cytotoxicity	Contact dermatitis (e.g., contact with poison ivy) and tissue-transplant rejection

Using Delayed Hypersensitivity to Test for TB

Austrian pediatrician Clemans von Pirquet (1874–1929) first described allergy mechanisms, including type III serum sickness. His interest led to the development of a test for tuberculosis (TB), using the tuberculin antigen, based on earlier work identifying the TB pathogen performed by Robert Koch. Pirquet's method involved scarification, which results in simultaneous multiple punctures, using a device with an array of needles to break the skin numerous times in a small area. The device Pirquet used was similar to the tine testdevice with four needles seen in Figure **24.4.8**.

The tips of all the needles in the array are coated with tuberculin, a protein extract of TB bacteria, effectively introducing the tuberculin into the skin. One to 3 days later, the area can be examined for a delayed hypersensitivity reaction, signs of which include swelling and redness.

As you can imagine, scarification was not a pleasant experience,[1] and the numerous skin punctures put the patient at risk of developing bacterial infection of the skin. Mantoux modified Pirquet's test to use a single subcutaneous injection of purified tuberculin material. A positive test, which is indicated by a delayed localized swelling at the injection site, does not necessarily mean that the patient is currently infected with active TB. Because type IV (delayed-type) hypersensitivity is mediated by reactivation of memory T cells, such cells may have been created recently (due to an active current infection) or years prior (if a patient had TB and had spontaneously cleared it, or if it had gone into latency). However, the test can be used to confirm infection in cases in which symptoms in the patient or findings on a radiograph suggest its presence.



Figure **24.4.8**: The modern version of Pirquet's scarification is the tine test, which uses devices like this to administer tuberculin antigen into the skin, usually on the inside of the forearm. The tine test is considered less reliable than the Mantoux test. (credit: modification of work by the Centers for Disease Control and Prevention)

Key Concepts and Summary

- **Type IV hypersensitivities** are not mediated by antibodies, but by helper T-cell activation of macrophages, eosinophils, and cytotoxic T cells.

Footnotes

1. B. Huber "100 Jahre Allergie: Clemens von Pirquet–sein Allergiebegriff und das ihm zugrunde liegende Krankheitsverständnis." *Wiener Klinische Wochenschrift* 118 no. 19–20 (2006):573–579.
2. C.A. Stewart. "The Pirquet Test: Comparison of the Scarification and the Puncture Methods of Application." *Archives of Pediatrics & Adolescent Medicine* 35 no. 3 (1928):388–391.

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24.5: Hypersensitivity Pneumonitis and Summary

Learning Objectives

- Explain why hypersensitivity pneumonitis is considered an occupational disease.
- Summarize the similarities and differences among different types of hypersensitivity.

Hypersensitivity Pneumonitis

Some disease caused by hypersensitivities are not caused exclusively by one type. For example, hypersensitivity pneumonitis (HP), which is often an occupational or environmental disease, occurs when the lungs become inflamed due to an allergic reaction to inhaled dust, endospores, bird feathers, bird droppings, molds, or chemicals. HP goes by many different names associated with various forms of exposure (Figure **24.5.9**). HP associated with bird droppings is sometimes called pigeon fancier's lung or poultry worker's lung—both common in bird breeders and handlers. Cheese handler's disease, farmer's lung, sauna takers' disease, and hot-tub lung are other names for HP associated with exposure to molds in various environments.

Pathology associated with HP can be due to both type III (mediated by immune complexes) and type IV (mediated by T_H1 cells and macrophages) hypersensitivities. Repeated exposure to allergens can cause alveolitis due to the formation of immune complexes in the alveolar wall of the lung accompanied by fluid accumulation, and the formation of granulomas and other lesions in the lung as a result of T_H1-mediated macrophage activation. Alveolitis with fluid and granuloma formation results in poor oxygen perfusion in the alveoli, which, in turn, can cause symptoms such as coughing, dyspnea, chills, fever, sweating, myalgias, headache, and nausea. Symptoms may occur as quickly as 2 hours after exposure and can persist for weeks if left untreated.



Figure **24.5.9**: Occupational exposure to dust, mold, and other allergens can result in hypersensitivity pneumonitis. (a) People exposed daily to large numbers of birds may be susceptible to poultry worker's lung. (b) Workers in a cheese factory may become sensitized to different types of molds and develop cheese handler's disease. (credit a: modification of work by The Global Orphan Project)

Figure **24.5.10** summarizes the mechanisms and effects of each type of hypersensitivity discussed in this section.

	Hypersensitivity Types and Their Mechanisms			
	Type I	**Type II**	**Type III**	**Type IV**
Immune reactant	IgE	IgG or IgM	IgG and IgM	T cells
Antigen form	Soluble antigen	Cell-bound antigen	Soluble antigen	Soluble or cell-bound antigen
Mechanism of activation	Allergen-specific IgE antibodies bind to mast cells via their Fc receptor. When the specific allergen binds to the IgE, cross-linking of IgE induces degranulation of mast cells.	IgG or IgM antibody binds to cellular antigen, leading to complement activation and cell lysis. IgG can also mediate ADCC with cytotoxic T cells, natural killer cells, macrophages, and neutrophils.	Antigen-antibody complexes are deposited in tissues. Complement activation provides inflammatory mediators and recruits neutrophils. Enzymes released from neutrophils damage tissue.	T_H1 cells secrete cytokines, which activate macrophages and cytotoxic T cells.
Examples of hypersensitivity reactions	Local and systemic anaphylaxis, seasonal hay fever, food allergies, and drug allergies	Red blood cell destruction after transfusion with mismatched blood types or during hemolytic disease of the newborn.	Post-streptococcal glomerulonephritis, rheumatoid arthritis, and systemic lupus erythematosus	Contact dermatitis, type I diabetes mellitus, and multiple sclerosis

Figure **24.5.10**: Components of the immune system cause four types of hypersensitivities. Notice that types I–III are B-cell/antibody-mediated hypersensitivities, whereas type IV hypersensitivity is exclusively a T-cell phenomenon.

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24.6: Diagnosis and Treatment of Hypersensitivities

Learning Objectives

- Describe the prick puncture skin test
- Explain why type III hypersensitivities can be difficult to diagnose
- Describe desensitization therapy
- Explain the role of epinephrine in treatment of hypersensitivity reactions

Diagnosis of Hypersensitivities

Diagnosis of type I hypersensitivities is a complex process requiring several diagnostic tests in addition to a well-documented patient history. Serum IgE levels can be measured, but elevated IgE alone does not confirm allergic disease. As part of the process to identify the antigens responsible for a type I reaction allergy, testing through a prick puncture skin test (PPST) or an intradermal test can be performed. PPST is carried out with the introduction of allergens in a series of superficial skin pricks on the patient's back or arms (Figure **24.6.11**). PPSTs are considered to be the most convenient and least expensive way to diagnose allergies, according to the US Joint Council of Allergy and the European Academy of Allergy and Immunology. The second type of testing, the intradermal test, requires injection into the dermis with a small needle. This needle, also known as a tuberculin needle, is attached to a syringe containing a small amount of allergen. Both the PPST and the intradermal tests are observed for 15–20 minutes for a wheal-flare reaction to the allergens. Measurement of any wheal (a raised, itchy bump) and flare (redness) within minutes indicates a type I hypersensitivity, and the larger the wheal-flare reaction, the greater the patient's sensitivity to the allergen.

Type III hypersensitivities can often be misdiagnosed because of their nonspecific inflammatory nature. The symptoms are easily visible, but they may be associated with any of a number of other diseases. A strong, comprehensive patient history is crucial to proper and accurate diagnosis. Tests used to establish the diagnosis of hypersensitivity pneumonitis (resulting from type III hypersensitivity) include bronchoalveolar lavage (BAL), pulmonary function tests, and high-resolution computed tomography (HRCT).

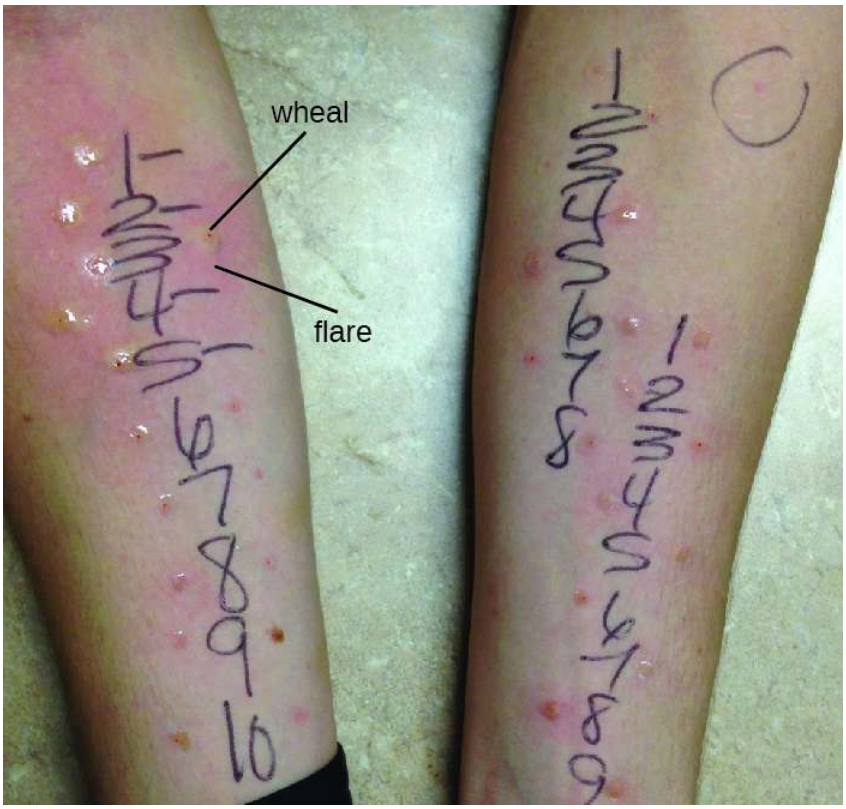


Figure **24.6.11**: Results of an allergy skin-prick test to test for type I hypersensitivity to a group of potential allergens. A positive result is indicated by a raised area (wheal) and surrounding redness (flare). (credit: modification of work by "OakleyOriginals"/Flickr)

Treatments of Hypersensitivities

Allergic reactions can be treated in various ways. Prevention of allergic reactions can be achieved by desensitization(hyposensitization) therapy, which can be used to reduce the hypersensitivity reaction through repeated injections of allergens. Extremely dilute concentrations of known allergens (determined from the allergen tests) are injected into the patient at prescribed intervals (e.g., weekly). The quantity of allergen delivered by the shots is slowly increased over a buildup period until an effective dose is determined and that dose is maintained for the duration of treatment, which can last years. Patients are usually encouraged to remain in the doctor's office for 30 minutes after receiving the injection in case the allergens administered cause a severe systemic reaction. Doctors' offices that administer desensitization therapy must be prepared to provide resuscitation and drug treatment in the case of such an event.

Desensitization therapy is used for insect sting allergies and environmental allergies. The allergy shots elicit the production of different interleukins and IgG antibody responses instead of IgE. When excess allergen-specific IgG antibodies are produced and bind to the allergen, they can act as blocking antibodies to neutralize the allergen before it can bind IgE on mast cells. There are early studies using oral therapy for desensitization of food allergies that are promising.[1112] These studies involve feeding children who have allergies tiny amounts of the allergen (e.g., peanut flour) or related proteins over time. Many of the subjects show reduced severity of reaction to the food allergen after the therapy.

There are also therapies designed to treat severe allergic reactions. Emergency systemic anaphylaxis is treated initially with an epinephrine injection, which can counteract the drop in blood pressure. Individuals with known severe allergies often carry a self-administering auto-injector that can be used in case of exposure to the allergen (e.g., an insect sting or accidental ingestion of a food that causes a severe reaction). By self-administering an epinephrine shot (or sometimes two), the patient can stem the reaction long enough to seek medical attention. Follow-up treatment generally involves giving the patient antihistamines and slow-acting corticosteroids for several days after the reaction to prevent potential late-phase reactions. However, the effects of antihistamine and corticosteroid treatment are not well studied and are used based on theoretical considerations.

Treatment of milder allergic reactions typically involves antihistamines and other anti-inflammatory drugs. A variety of antihistamine drugs are available, in both prescription and over-the-counter strengths. There are also antileukotriene and antiprostaglandin drugs that can be used in tandem with antihistamine drugs in a combined (and more effective) therapy regime.

Treatments of type III hypersensitivities include preventing further exposure to the antigen and the use of anti-inflammatory drugs. Some conditions can be resolved when exposure to the antigen is prevented. Anti-inflammatory corticosteroid inhalers can also be used to diminish inflammation to allow lung lesions to heal. Systemic corticosteroid treatment, oral or intravenous, is also common for type III hypersensitivities affecting body systems. Treatment of hypersensitivity pneumonitis includes avoiding the allergen, along with the possible addition of prescription steroids such as prednisone to reduce inflammation.

Treatment of type IV hypersensitivities includes antihistamines, anti-inflammatory drugs, analgesics, and, if possible, eliminating further exposure to the antigen.

Key Concepts and Summary

- An **allergy** is an adaptive immune response, sometimes life-threatening, to an **allergen**.
- **Type I hypersensitivity** requires sensitization of mast cells with IgE, involving an initial IgE antibody response and IgE attachment to mast cells. On second exposure to an allergen, cross-linking of IgE molecules on mast cells triggers degranulation and release of preformed and newly formed chemical mediators of inflammation. Type I hypersensitivity may be localized and relatively minor (hives and hay fever) or system-wide and dangerous (systemic **anaphylaxis**).
- **Type II hypersensitivities** result from antibodies binding to antigens on cells and initiating cytotoxic responses. Examples include **hemolytic transfusion reaction** and **hemolytic disease of the newborn**.
- **Type III hypersensitivities** result from formation and accumulation of **immune complexes** in tissues, stimulating damaging inflammatory responses.
- **Type IV hypersensitivities** are not mediated by antibodies, but by helper T-cell activation of macrophages, eosinophils, and cytotoxic T cells.

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24.E: Diseases of the Immune System (Exercises)

24.1: Hypersensitivities

An allergy is an adaptive immune response, sometimes life-threatening, to an allergen. Hypersensitivity reactions are classified by their immune mechanism.

Multiple Choice

Which of the following is the type of cell largely responsible for type I hypersensitivity responses?

A. erythrocyte
B. mast cell
C. T lymphocyte
D. antibody

Answer

B

Type I hypersensitivities require which of the following initial priming events to occur?

A. sensitization
B. secondary immune response
C. cellular trauma
D. degranulation

Answer

A

Which of the following are the main mediators/initiators of type II hypersensitivity reactions?

A. antibodies
B. mast cells
C. erythrocytes
D. histamines

Answer

A

Inflammatory molecules are released by mast cells in type I hypersensitivities; type II hypersensitivities, however, are characterized by which of the following?

A. cell lysis (cytotoxicity)
B. strong antibody reactions against antigens
C. leukotriene release upon stimulation
D. localized tissue reactions, such as hives

Answer

A

An immune complex is an aggregate of which of the following?

A. antibody molecules
B. antigen molecules
C. antibody and antigen molecules
D. histamine molecules

Answer

C

Which of the following is a common treatment for type III hypersensitivity reactions?

A. anti-inflammatory steroid treatments
B. antihistamine treatments
C. hyposensitization injections of allergens
D. RhoGAM injections

Answer

A

Which of the following induces a type III hypersensitivity?

A. release of inflammatory molecules from mast cells
B. accumulation of immune complexes in tissues and small blood vessels
C. destruction of cells bound by antigens
D. destruction of cells bound by antibodies

Answer

B

Which one of the following is not an example of a type IV hypersensitivity?

A. latex allergy
B. Contact dermatitis (e.g., contact with poison ivy)
C. a positive tuberculin skin test
D. hemolytic disease of the newborn

Answer

D

Fill in the Blank

Antibodies involved in type I hypersensitivities are of the _____ class.

Answer

IgE

Allergy shots work by shifting antibody responses to produce _____ antibodies.

Answer

IgG

A person who is blood type A would have IgM hemagglutinin antibodies against type _____ red blood cells in their plasma.

Answer

B

The itchy and blistering rash that develops with contact to poison ivy is caused by a type _____ hypersensitivity reaction.

Answer

IV

Short Answer

Although both type I and type II hypersensitivities involve antibodies as immune effectors, different mechanisms are involved with these different hypersensitivities. Differentiate the two.

What types of antibodies are most common in type III hypersensitivities, and why?

Critical Thinking

Patients are frequently given instructions to avoid allergy medications for a period of time prior to allergy testing. Why would this be important?

In some areas of the world, a tuberculosis vaccine known as bacillus Calmette-Guérin (BCG) is used. It is not used in the United States. Every person who has received this vaccine and mounted a protective response will have a positive reaction in a tuberculin skin test. Why? What does this mean for the usefulness of this skin test in those countries where this vaccine is used?

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CHAPTER OVERVIEW

25: Diseases of the Immune System

An allergic reaction is an immune response to a type of antigen called an allergen. Allergens can be found in many different items, from peanuts and insect stings to latex and some drugs. Unlike other kinds of antigens, allergens are not necessarily associated with pathogenic microbes, and many allergens provoke no immune response at all in most people.

Allergic responses vary in severity. Some are mild and localized, like hay fever or hives, but others can result in systemic, life-threatening reactions. Anaphylaxis, for example, is a rapidly developing allergic reaction that can cause a dangerous drop in blood pressure and severe swelling of the throat that may close off the airway.

Allergies are just one example of how the immune system—the system normally responsible for preventing disease—can actually cause or mediate disease symptoms. In this chapter, we will further explore allergies and other disorders of the immune system, including hypersensitivity reactions, autoimmune diseases, transplant rejection, and diseases associated with immunodeficiency.



Figure **25.1**: Bee stings and other allergens can cause life-threatening, systemic allergic reactions. Sensitive individuals may need to carry an epinephrine auto-injector (e.g., EpiPen) in case of a sting. A bee-sting allergy is an example of an immune response that is harmful to the host rather than protective; epinephrine counteracts the severe drop in blood pressure that can result from the immune response. (credit right: modification of work by Carol Bleistine)

25.1: Organ-Specific Autoimmune Disorders
25.2: Systemic Autoimmune Disorders
25.3: Organ Transplantation and Rejection
25.4: Immunodeficiency
25.E: Diseases of the Immune System (Exercises)

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25.1: Organ-Specific Autoimmune Disorders

Learning Objectives

- Explain why autoimmune disorders develop
- Name the organ-specific autoimmune disorders and the organs that they affect
- Describe how the mechanisms of Graves disease and Hashimoto thyroiditis differ
- Name the cells that are destroyed in type 1 diabetes melitus and describe the result

In 1970, artist Walt Kelly developed a poster promoting Earth Day, featuring a character from *Pogo*, his daily newspaper comic strip. In the poster, Pogo looks out across a litter-strewn forest and says wryly, “We have met the enemy and he is us.” Pogo was not talking about the human immune system, but he very well could have been. Although the immune system protects the body by attacking invading “enemies” (pathogens), in some cases, the immune system can mistakenly identify the body’s own cells as the enemy, resulting in **autoimmune disease**.

Autoimmune diseases are those in which the body is attacked by its own specific adaptive immune response. In normal, healthy states, the immune system induces **tolerance**, which is a lack of an anti-self immune response. However, with autoimmunity, there is a loss of immune tolerance, and the mechanisms responsible for autoimmune diseases include type II, III, and IV hypersensitivity reactions. Autoimmune diseases can have a variety of mixed symptoms that flare up and disappear, making diagnosis difficult.

The causes of autoimmune disease are a combination of the individual's genetic makeup and the effect of environmental influences, such as sunlight, infections, medications, and environmental chemicals. However, the vagueness of this list reflects our poor understanding of the etiology of these diseases. Except in a very few specific diseases, the initiation event(s) of most autoimmune states has not been fully characterized.

There are several possible causes for the origin of autoimmune diseases and autoimmunity is likely due to several factors. Evidence now suggests that regulatory T and B cells play an essential role in the maintenance of tolerance and prevention of autoimmune responses. The regulatory T cells are especially important for inhibiting autoreactive T cells that are not eliminated during thymic selection and escape the thymus (see T Lymphocytes and Cellular Immunity). In addition, antigen mimicry between pathogen antigens and our own self antigens can lead to cross-reactivity and autoimmunity. Hidden self-antigens may become exposed because of trauma, drug interactions, or disease states, and trigger an autoimmune response. All of these factors could contribute to autoimmunity. Ultimately, damage to tissues and organs in the autoimmune disease state comes as a result of inflammatory responses that are inappropriate; therefore, treatment often includes immunosuppressive drugs and corticosteroids.

Organ-Specific Autoimmune Diseases

Some autoimmune diseases are considered organ specific, meaning that the immune system targets specific organs or tissues. Examples of organ-specific autoimmune diseases include celiac disease, Graves disease, Hashimoto thyroiditis, type I diabetes mellitus, and Addison disease.

Celiac Disease

Celiac disease is largely a disease of the small intestine, although other organs may be affected. People in their 30s and 40s, and children are most commonly affected, but celiac disease can begin at any age. It results from a reaction to proteins, commonly called gluten, found mainly in wheat, barley, rye, and some other grains. The disease has several genetic causes (predispositions) and poorly understood environmental influences. On exposure to gluten, the body produces various autoantibodies and an inflammatory response. The inflammatory response in the small intestine leads to a reduction in the depth of the microvilli of the mucosa, which hinders absorption and can lead to weight loss and anemia. The disease is also characterized by diarrhea and abdominal pain, symptoms that are often misdiagnosed as irritable bowel syndrome.

Diagnosis of celiac disease is accomplished from serological tests for the presence of primarily IgA antibodies to components of gluten, the transglutinaminase enzyme, and autoantibodies to endomysium, a connective tissue surrounding muscle fibers. Serological tests are typically followed up with endoscopy and biopsy of the duodenal mucosa. Serological screening surveys have

found about 1% of individuals in the United Kingdom are positive even though they do not all display symptoms.[1] This early recognition allows for more careful monitoring and prevention of severe disease.

Celiac disease is treated with complete removal of gluten-containing foods from the diet, which results in improved symptoms and reduced risk of complications. Other theoretical approaches include breeding grains that do not contain the immunologically reactive components or developing dietary supplements that contain enzymes that break down the protein components that cause the immune response.[2]

Disorders of the Thyroid

Graves disease is the most common cause of hyperthyroidism in the United States. Symptoms of Graves disease result from the production of thyroid-stimulating immunoglobulin (TSI) also called TSH-receptor antibody. TSI targets and binds to the receptor for thyroid stimulating hormone (TSH), which is naturally produced by the pituitary gland. TSI may cause conflicting symptoms because it may stimulate the thyroid to make too much thyroid hormone or block thyroid hormone production entirely, making diagnosis more difficult. Signs and symptoms of Graves disease include heat intolerance, rapid and irregular heartbeat, weight loss, goiter (a swollen thyroid gland, protruding under the skin of the throat [Figure **25.1.1**]) and exophthalmia (bulging eyes) often referred to as Graves ophthalmopathy (Figure **25.1.2**).



Figure **25.1.1**: Goiter, a hypertrophy of the thyroid, is a symptom of Graves disease and Hashimoto thyroiditis.

The most common cause of hypothyroidism in the United States is **Hashimoto thyroiditis**, also called chronic lymphocytic thyroiditis. Patients with Hashimoto thyroiditis often develop a spectrum of different diseases because they are more likely to develop additional autoimmune diseases such as Addison disease (discussed later in this section), type 1 diabetes, rheumatoid arthritis, and celiac disease. Hashimoto thyroiditis is a T_H1 cell-mediated disease that occurs when the thyroid gland is attacked by cytotoxic lymphocytes, macrophages, and autoantibodies. This autoimmune response leads to numerous symptoms that include goiter (Figure **25.1.1**), cold intolerance, muscle weakness, painful and stiff joints, depression, and memory loss.

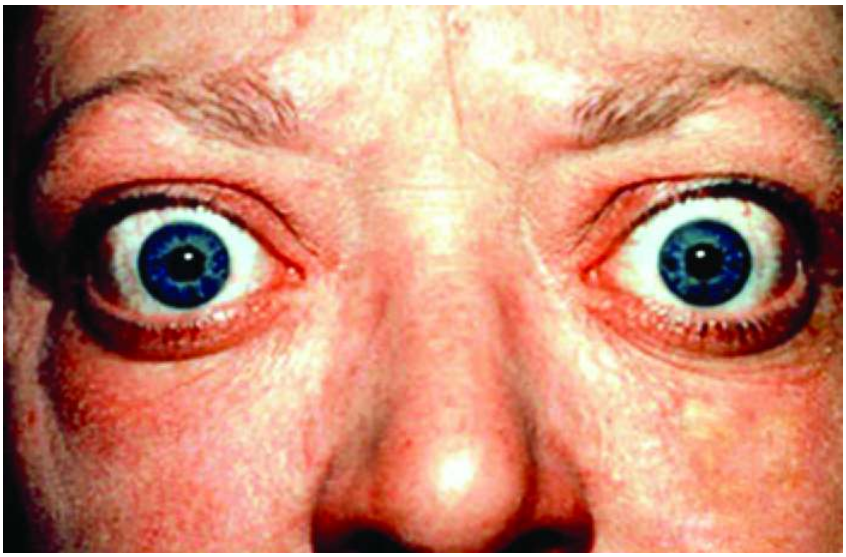


Figure **25.1.2**: Exophthalmia, or Graves ophthalmopathy, is a sign of Graves disease. (credit: modification of work by Jonathan Trobe, University of Michigan Kellogg Eye Center)

Type 1 Diabetes

Juvenile diabetes, or **type 1 diabetes mellitus**, is usually diagnosed in children and young adults. It is a T-cell-dependent autoimmune disease characterized by the selective destruction of the β cells of the islets of Langerhans in the pancreas by CD4 T_H1-mediated CD8 T cells, anti-β-cell antibodies, and macrophage activity. There is also evidence that viral infections can have either a potentiating or inhibitory role in the development of type 1 diabetes (T1D) mellitus. The destruction of the β cells causes a lack of insulin production by the pancreas. In T1D, β-cell destruction may take place over several years, but symptoms of hyperglycemia, extreme increase in thirst and urination, weight loss, and extreme fatigue usually have a sudden onset, and diagnosis usually does not occur until most β cells have already been destroyed.

Autoimmune Addison Disease

Destruction of the adrenal glands (the glands lying above the kidneys that produce glucocorticoids, mineralocorticoids, and sex steroids) is the cause of **Addison disease**, also called primary adrenal insufficiency (PAI). Today, up to 80% of Addison disease cases are diagnosed as autoimmune Addison disease (AAD), which is caused by an autoimmune response to adrenal tissues disrupting adrenal function. Disruption of adrenal function causes impaired metabolic processes that require normal steroid hormone levels, causing signs and symptoms throughout the body. There is evidence that both humoral and CD4 T_H1-driven CD8 T-cell–mediated immune mechanisms are directed at the adrenal cortex in AAD. There is also evidence that the autoimmune response is associated with autoimmune destruction of other endocrine glands as well, such as the pancreas and thyroid, conditions collectively referred to as autoimmune polyendocrine syndromes (APS). In up to 80% of patients with AAD, antibodies are produced to three enzymes involved in steroid synthesis: 21-hydroxylase (21-OH), 17α-hydroxylase, and cholesterol side-chain–cleaving enzyme.[3]The most common autoantibody found in AAD is to 21-OH, and antibodies to any of the key enzymes for steroid production are diagnostic for AAD. The adrenal cortex cells are targeted, destroyed, and replaced with fibrous tissue by immune-mediated inflammation. In some patients, at least 90% of the adrenal cortex is destroyed before symptoms become diagnostic.

Symptoms of AAD include weakness, nausea, decreased appetite, weight loss, hyperpigmentation (Figure **25.1.3**), hyperkalemia (elevated blood potassium levels), hyponatremia (decreased blood sodium levels), hypoglycemia(decreased levels of blood sugar), hypotension (decreased blood pressure), anemia, lymphocytosis (decreased levels of white blood cells), and fatigue. Under extreme stress, such as surgery, accidental trauma, or infection, patients with AAD may experience an adrenal crisis that causes the patient to vomit, experience abdominal pain, back or leg cramps, and even severe hypotension leading to shock.



Figure **25.1.3**: Hyperpigmentation is a sign of Addison disease. (credit: modification of work by Petros Perros)

Key Concepts and Summary

- **Autoimmune diseases** result from a breakdown in immunological tolerance. The actual induction event(s) for autoimmune states are largely unknown.
- Some autoimmune diseases attack specific organs, whereas others are more systemic.
- Organ-specific autoimmune diseases include **celiac disease**, **Graves disease**, **Hashimoto thyroiditis**, **type I diabetes mellitus,** and **Addison disease**.

Footnotes

1. 1 D.A. Van Heel, J. West. “Recent Advances in Coeliac Disease.” *Gut* 55 no. 7 (2006):1037—1046.
2. 2 ibid.
3. 3 P. Martorell et al. “Autoimmunity in Addison’s Disease.” *Netherlands Journal of Medicine* 60 no. 7 (2002):269—275.

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25.2: Systemic Autoimmune Disorders

Learning Objectives

- List the ways antibodies contribute to the pathogenesis of myasthenia gravis
- Explain why rheumatoid arthritis is considered a type III hypersensitivity
- Describe the symptoms of systemic lupus erythematosus and explain why they affect so many different parts of the body

Systemic Autoimmune Diseases

Whereas organ-specific autoimmune diseases target specific organs or tissues, systemic autoimmune diseases are more generalized, targeting multiple organs or tissues throughout the body. Examples of **systemic autoimmune diseases** include multiple sclerosis, myasthenia gravis, psoriasis, rheumatoid arthritis, and systemic lupus erythematosus.

Multiple Sclerosis

Multiple sclerosis (MS) is an autoimmune central nervous system disease that affects the brain and spinal cord. Lesions in multiple locations within the central nervous system are a hallmark of multiple sclerosis and are caused by infiltration of immune cells across the blood-brain barrier. The immune cells include T cells that promote inflammation, demyelination, and neuron degeneration, all of which disrupt neuronal signaling. Symptoms of MS include visual disturbances; muscle weakness; difficulty with coordination and balance; sensations such as numbness, prickling, or “pins and needles”; and cognitive and memory problems.

Myasthenia Gravis

Autoantibodies directed against acetylcholine receptors (AChRs) in the synaptic cleft of neuromuscular junctions lead to **myasthenia gravis** (Figure **25.2.4**). Anti-AChR antibodies are high-affinity IgGs and their synthesis requires activated CD4 T cells to interact with and stimulate B cells. Once produced, the anti-AChR antibodies affect neuromuscular transmission by at least three mechanisms:

- Complement binding and activation at the neuromuscular junction
- Accelerated AChR endocytosis of molecules cross-linked by antibodies
- Functional AChR blocking, which prevents normal acetylcholine attachment to, and activation of, AChR

Regardless of the mechanism, the effect of anti-AChR is extreme muscle weakness and potentially death through respiratory arrest in severe cases.

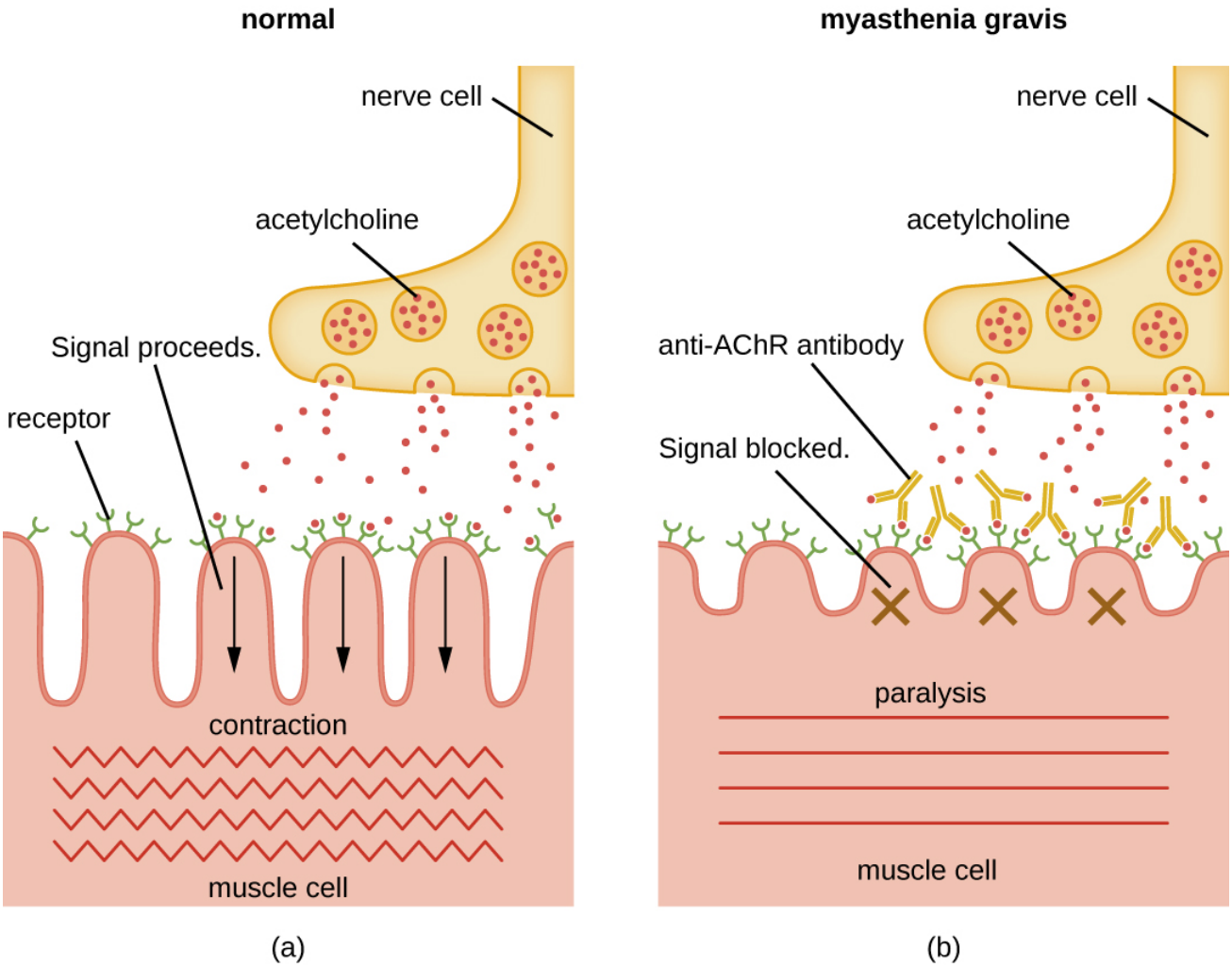


Figure **25.2.4**: Myasthenia gravis and impaired muscle contraction. (a) Normal release of the neurotransmitter acetylcholine stimulates muscle contraction. (b) In myasthenia gravis, autoantibodies block the receptors for acetylcholine (AChr) on muscle cells, resulting in paralysis.

Psoriasis

Psoriasis is a skin disease that causes itchy or sore patches of thick, red skin with silvery scales on elbows, knees, scalp, back, face, palms, feet, and sometimes other areas. Some individuals with **psoriasis** also get a form of arthritis called psoriatic arthritis, in which the joints can become inflamed. Psoriasis results from the complex interplay between keratinocytes, dendritic cells, and T cells, and the cytokines produced by these various cells. In a process called cell turnover, skin cells that grow deep in the skin rise to the surface. Normally, this process takes a month. In psoriasis, as a result of cytokine activation, cell turnover happens in just a few days. The thick inflamed patches of skin that are characteristic of psoriasis develop because the skin cells rise too fast.

Rheumatoid Arthritis

The most common chronic inflammatory joint disease is **rheumatoid arthritis (RA)** (Figure **25.2.5**) and it is still a major medical challenge because of unsolved questions related to the environmental and genetic causes of the disease. RA involves type III hypersensitivity reactions and the activation of CD4 T cells, resulting in chronic release of the inflammatory cytokines IL-1, IL-6, and tumor necrosis factor-α (TNF-α). The activated CD4 T cells also stimulate the production of rheumatoid factor (RF) antibodies and anticyclic citrullinated peptide antibodies (anti-CCP) that form immune complexes. Increased levels of acute-phase proteins, such as C-reactive protein (CRP), are also produced as part of the inflammatory process and participate in complement fixation with the antibodies on the immune complexes. The formation of immune complexes and reaction to the immune factors cause an inflammatory process in joints, particularly in the hands, feet, and legs. Diagnosis of RA is based on elevated levels of RF, anti-

CCP, quantitative CRP, and the erythrocyte sedimentation rate (ESR) (modified Westergren). In addition, radiographs, ultrasound, or magnetic resonance imaging scans can identify joint damage, such as erosions, a loss of bone within the joint, and narrowing of joint space.



Figure **25.2.5**: The radiograph (left) and photograph (right) show damage to the hands typical of rheumatoid arthritis. (credit right: modification of work by “handarmdoc”/Flickr)

Systemic Lupus Erythematosus

The damage and pathology of **systemic lupus erythematosus (SLE)** is caused by type III hypersensitivity reactions. Autoantibodies produced in SLE are directed against nuclear and cytoplasmic proteins. Anti-nuclear antibodies (ANAs) are present in more than 95% of patients with SLE,[4] with additional autoantibodies including anti-double–stranded DNA (ds-DNA) and anti-Sm antibodies (antibodies to small nuclear ribonucleoprotein). Anti-ds-DNA and anti-Sm antibodies are unique to patients with SLE; thus, their presence is included in the classification criteria of SLE. Cellular interaction with autoantibodies leads to nuclear and cellular destruction, with components released after cell death leading to the formation of immune complexes.

Because autoantibodies in SLE can target a wide variety of cells, symptoms of SLE can occur in many body locations. However, the most common symptoms include fatigue, fever with no other cause, hair loss, and a sunlight-sensitive "butterfly" or wolf-mask (lupus) rash that is found in about 50% of people with SLE (Figure **25.2.6**). The rash is most often seen over the cheeks and bridge of the nose, but can be widespread. Other symptoms may appear depending on affected areas. The joints may be affected, leading to arthritis of the fingers, hands, wrists, and knees. Effects on the brain and nervous system can lead to headaches, numbness, tingling, seizures, vision problems, and personality changes. There may also be abdominal pain, nausea, vomiting, arrhythmias, shortness of breath, and blood in the sputum. Effects on the skin can lead to additional areas of skin lesions, and vasoconstriction can cause color changes in the fingers when they are cold (Raynaud phenomenon). Effects on the kidneys can lead to edema in the legs and weight gain. A diagnosis of SLE depends on identification of four of 11 of the most common symptoms and confirmed production of an array of autoantibodies unique to SLE. A positive test for ANAs alone is not diagnostic.

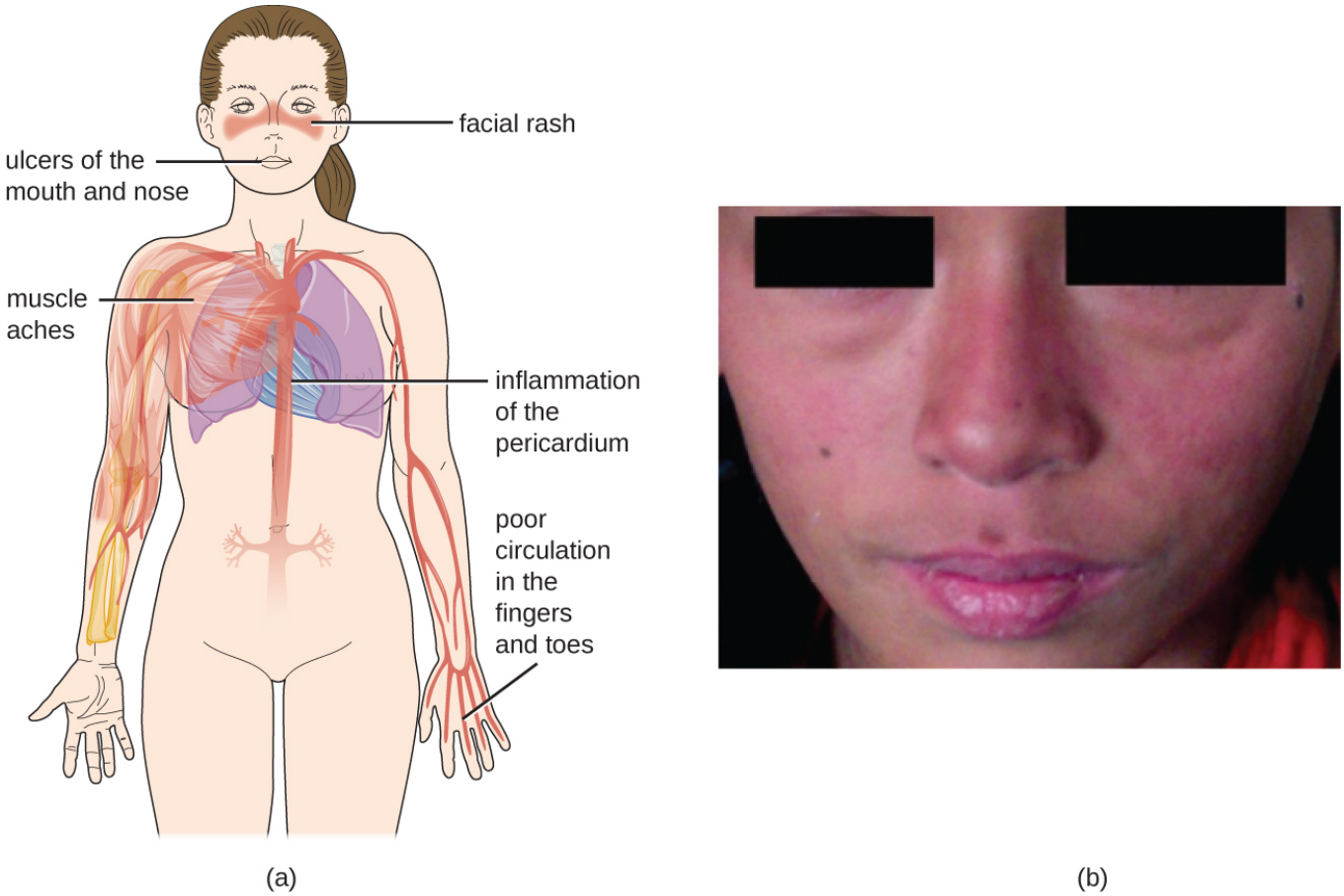


Figure 25.2.6: (a) Systemic lupus erythematosus is characterized by autoimmunity to the individual's own DNA and/or proteins. (b) This patient is presenting with a butterfly rash, one of the characteristic signs of lupus. (credit a: modification of work by Mikael Häggström; credit b: modification of work by Shrestha D, Dhakal AK, Shiva RK, Shakya A, Shah SC, Shakya H)

Table 25.2.1 summarizes the causes, signs, and symptoms of select autoimmune diseases.

Table 25.2.1: Select Autoimmune Diseases

Disease	Cause	Signs and Symptoms
Addison disease	Destruction of adrenal gland cells by cytotoxic T cells	Weakness, nausea, hypotension, fatigue; adrenal crisis with severe pain in abdomen, lower back, and legs; circulatory system collapse, kidney failure
Celiac disease	Antibodies to gluten become autoantibodies that target cells of the small intestine	Severe diarrhea, abdominal pain, anemia, malnutrition
Diabetes mellitus (type I)	Cytotoxic T-cell destruction of the insulin-producing β cells of the pancreas	Hyperglycemia, extreme increase in thirst and urination, weight loss, extreme fatigue
Graves disease	Autoantibodies target thyroid-stimulating hormone receptors, resulting in overstimulation of the thyroid	Hyperthyroidism with rapid and irregular heartbeat, heat intolerance, weight loss, goiter, exophthalmia
Hashimoto thyroiditis	Thyroid gland is attacked by cytotoxic T cells, lymphocytes, macrophages, and autoantibodies	Thyroiditis with goiter, cold intolerance, muscle weakness, painful and stiff joints, depression, memory loss
Multiple sclerosis (MS)	Cytotoxic T-cell destruction of the myelin sheath surrounding nerve axons in the central nervous system	Visual disturbances, muscle weakness, impaired coordination and balance, numbness, prickling or “pins and needles” sensations, impaired cognitive function and memory
Myasthenia gravis	Autoantibodies directed against acetylcholine receptors within the neuromuscular junction	Extreme muscle weakness eventually leading to fatal respiratory arrest
Psoriasis	Cytokine activation of keratinocytes causes rapid and excessive epidermal cell turnover	Itchy or sore patches of thick, red skin with silvery scales; commonly affects elbows, knees, scalp, back, face, palms, feet
Rheumatoid arthritis	Autoantibodies, immune complexes, complement activation, phagocytes, and T cells damage membranes and bone in joints	Joint inflammation, pain and disfigurement, chronic systemic inflammation
Systemic lupus erythematosus (SLE)	Autoantibodies directed against nuclear and cytoplasmic molecules form immune complexes that deposit in tissues. Phagocytic cells and complement activation cause tissue damage and inflammation	Fatigue, fever, joint pain and swelling, hair loss, anemia, clotting, a sunlight-sensitive "butterfly" rash, skin lesions, photosensitivity, decreased kidney function, memory loss, confusion, depression

Key Concepts and Summary

- Systemic autoimmune diseases include **multiple sclerosis**, **myasthenia gravis**, **psoriasis**, **rheumatoid arthritis**, and **systemic lupus erythematosus**.
- Treatments for autoimmune diseases generally involve anti-inflammatory and immunosuppressive drugs.

Footnotes

1. 4 C.C. Mok, C.S. Lau. “Pathogenesis of Systemic Lupus Erythematosus.” *Journal of Clinical Pathology* 56 no. 7 (2003):481—490.

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25.3: Organ Transplantation and Rejection

Learning Objectives

- Explain why human leukocyte antigens (HLAs) are important in tissue transplantation
- Explain the types of grafts possible and their potential for interaction with the immune system
- Describe the role of immunosuppression in transplantation
- Describe what occurs during graft-versus-host disease (GVHD)

A **graft** is the transplantation of an organ or tissue to a different location, with the goal of replacing a missing or damaged organ or tissue. Grafts are typically moved without their attachments to the circulatory system and must reestablish these, in addition to the other connections and interactions with their new surrounding tissues. There are different types of grafts depending on the source of the new tissue or organ. Tissues that are transplanted from one genetically distinct individual to another within the same species are called **allografts**. An interesting variant of the allograft is an **isograft**, in which tissue from one twin is transplanted to another. As long as the twins are monozygotic(therefore, essentially genetically identical), the transplanted tissue is virtually never rejected. If tissues are transplanted from one area on an individual to another area on the same individual (e.g., a skin graft on a burn patient), it is known as an **autograft**. If tissues from an animal are transplanted into a human, this is called a **xenograft**.

Transplant Rejection

The different types of grafts described above have varying risks for rejection (Table **25.3.1**). Rejection occurs when the recipient's immune system recognizes the donor tissue as foreign (non-self), triggering an immune response. The major histocompatibility complex markers MHC I and MHC II, more specifically identified as **human leukocyte antigens (HLAs)**, play a role in transplant rejection. The HLAs expressed in tissue transplanted from a genetically different individual or species may be recognized as non-self molecules by the host's dendritic cells. If this occurs, the dendritic cells will process and present the foreign HLAs to the host's helper T cells and cytotoxic T cells, thereby activating them. Cytotoxic T cells then target and kill the grafted cells through the same mechanism they use to kill virus-infected cells; helper T cells may also release cytokines that activate macrophages to kill graft cells.

Table **25.3.1**: Types of Tissue and Organ Grafts and Their Complications

Graft	Procedure	Complications
Autograft	From self to self	No rejection concerns
Isograft	From identical twin to twin	Little concern of rejection
Allograft	From relative or nonrelative to individual	Rejection possible
Xenograft	From animal to human	Rejection possible

With the three highly polymorphic MHC I genes in humans (*HLA-A*, *HLA-B*, and *HLA-C*) determining compatibility, each with many alleles segregating in a population, odds are extremely low that a randomly chosen donor will match a recipient's six-allele genotype (the two alleles at each locus are expressed codominantly). This is why a parent or a sibling may be the best donor in many situations—a genetic match between the MHC genes is much more likely and the organ is much less likely to be rejected.

Although matching all of the MHC genes can lower the risk for rejection, there are a number of additional gene products that also play a role in stimulating responses against grafted tissue. Because of this, no non-self grafted tissue is likely to completely avoid rejection. However, the more similar the MHC gene match, the more likely the graft is to be tolerated for a longer time. Most transplant recipients, even those with tissues well matched to their MHC genes, require treatment with immunosuppressant drugs for the rest of their lives. This can make them more vulnerable than the general population to complications from infectious diseases. It can also result in transplant-related malignancies because the body's normal defenses against cancer cells are being suppressed.

Graft-versus-Host Disease

A form of rejection called graft-versus-host disease (GVHD) primarily occurs in recipients of bone marrow transplants and peripheral blood stem cells. GHVD presents a unique situation because the transplanted tissue is capable of producing immune cells; APCs in the donated bone marrow may recognize the host cells as non-self, leading to activation of the donor cytotoxic T cells. Once activated, the donor's T cells attack the recipient cells, causing acute GVHD.

Acute GVHD typically develops within weeks after a bone marrow transplant, causing tissue damage affecting the skin, gastrointestinal tract, liver, and eyes. In addition, acute GVHD may also lead to a cytokine storm, an unregulated secretion of cytokines that may be fatal. In addition to acute GVHD, there is also the risk for chronic GVHD developing months after the bone marrow transplant. The mechanisms responsible for chronic GVHD are not well understood.

To minimize the risk of GVHD, it is critically important to match the HLAs of the host and donor as closely as possible in bone marrow transplants. In addition, the donated bone marrow is processed before grafting to remove as many donor APCs and T cells as possible, leaving mostly *hematopoietic* stem cells.

The Future of Transplantation

Historically speaking, the practice of transplanting tissues—and the complications that can accompany such procedures—is a relatively recent development. It was not until 1954 that the first successful organ transplantation between two humans was achieved. Yet the field of organ transplantation has progressed rapidly since that time.

Nonetheless, the practice of transplanting non-self tissues may soon become obsolete. Scientists are now attempting to develop methods by which new organs may be grown *in vitro* from an individual's own harvested cells to replace damaged or abnormal ones. Because organs produced in this way would contain the individual's own cells, they could be transplanted into the individual without risk for rejection.

An alternative approach that is gaining renewed research interest is genetic modification of donor animals, such as pigs, to provide transplantable organs that do not elicit an immune response in the recipient. The approach involves excising the genes in the pig (in the embryo) that are most responsible for the rejection reaction after transplantation. Finding these genes and effectively removing them is a challenge, however. So too is identifying and neutralizing risks from viral sequences that might be embedded in the pig genome, posing a risk for infection in the human recipient.

Key Concepts and Summary

- Grafts and transplants can be classified as **autografts**, **isografts, allografts**, or **xenografts** based on the genetic differences between the donor's and recipient's tissues.
- Genetic differences, especially among the MHC (HLA) genes, will dictate the likelihood that **rejection** of the transplanted tissue will occur.
- Transplant recipients usually require immunosuppressive therapy to avoid rejection, even with good genetic matching. This can create additional problems when immune responses are needed to fight off infectious agents and prevent cancer.
- **Graft-versus-host disease** can occur in bone marrow transplants, as the mature T cells in the transplant itself recognize the recipient's tissues as foreign.
- Transplantation methods and technology have improved greatly in recent decades and may move into new areas with the use of stem cell technology to avoid the need for genetic matching of MHC molecules.

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25.4: Immunodeficiency

Learning Objectives

- Compare the causes of primary and secondary immunodeficiencies
- Explain why patients with chronic granulomatous disease are especially susceptible to bacterial infections
- Explain why individuals with IgA deficiency are susceptible to respiratory and GI infections
- Describe treatments for primary and secondary immunodeficiencies
- Explain why secondary immunodeficiencies can sometimes be reversed

Immunodeficiencies are inherited (primary) or acquired (secondary) disorders in which elements of host immune defenses are either absent or functionally defective. In developed countries, most immunodeficiencies are inherited, and they are usually first seen in the clinic as recurrent or overwhelming infections in infants. However, on a global scale, malnutrition is the most common cause of immunodeficiency and would be categorized as an acquired immunodeficiency. Acquired immunodeficiencies are more likely to develop later in life, and the pathogenic mechanisms of many remain obscure.

Primary Immunodeficiency

Primary immunodeficiencies, which number more than 250, are caused by inherited defects of either nonspecific innate or specific adaptive immune defenses. In general, patients born with primary immunodeficiency (PI) commonly have an increased susceptibility to infection. This susceptibility can become apparent shortly after birth or in early childhood for some individuals, whereas other patients develop symptoms later in life. Some primary immunodeficiencies are due to a defect of a single cellular or humoral component of the immune system; others may result from defects of more than one component. Examples of primary immunodeficiencies include chronic granulomatous disease, X-linked agammaglobulinemia, selective IgA deficiency, and severe combined immunodeficiency disease.

Chronic Granulomatous Disease

The causes of **chronic granulomatous disease (CGD)** are defects in the NADPH oxidase system of phagocytic cells, including neutrophils and macrophages, that prevent the production of superoxide radicals in phagolysosomes. The inability to produce superoxide radicals impairs the antibacterial activity of phagocytes. As a result, infections in patients with CGD persist longer, leading to a chronic local inflammation called a granuloma. Microorganisms that are the most common causes of infections in patients with CGD include *Aspergillus* spp., *Staphylococcus aureus*, *Chromobacterium violaceum*, *Serratia marcescens*, and *Salmonella typhimurium*.

X-Linked Agammaglobulinemia

Deficiencies in B cells due to defective differentiation lead to a lack of specific antibody production known as **X-linked agammaglobulinemia**. In 1952, Ogden C. Bruton (1908–2003) described the first immunodeficiency in a boy whose immune system failed to produce antibodies. This defect is inherited on the X chromosome and is characterized by the absence of immunoglobulin in the serum; it is called Bruton X-linked agammaglobulinemia (XLA). The defective gene, *BTK*, in XLA is now known to encode a tyrosine kinase called Bruton tyrosine kinase (Btk). In patients whose B cells are unable to produce sufficient amounts of Btk, the B-cell maturation and differentiation halts at the pre-B-cell stage of growth. B-cell maturation and differentiation beyond the pre-B-cell stage of growth is required for immunoglobulin production. Patients who lack antibody production suffer from recurrent infections almost exclusively due to extracellular pathogens that cause pyogenic infections: *Haemophilus influenzae*, *Streptococcus pneumoniae*, *S. pyogenes*, and *S. aureus*. Because cell-mediated immunity is not impaired, these patients are not particularly vulnerable to infections caused by viruses or intracellular pathogens.

Selective IgA Deficiency

The most common inherited form of immunoglobulin deficiency is selective IgA deficiency, affecting about one in 800 people. Individuals with **selective IgA deficiency** produce normal levels of IgG and IgM, but are not able to produce secretory IgA. IgA deficiency predisposes these individuals to lung and gastrointestinal infections for which secretory IgA is normally an important

defense mechanism. Infections in the lungs and gastrointestinal tract can involve a variety of pathogens, including *H. influenzae*, *S. pneumoniae*, *Moraxella catarrhalis*, *S. aureus*, *Giardia lamblia*, or pathogenic strains of *Escherichia coli*.

Severe Combined Immunodeficiency

Patients who suffer from **severe combined immunodeficiency (SCID)** have B-cell and T-cell defects that impair T-cell dependent antibody responses as well as cell-mediated immune responses. Patients with SCID also cannot develop immunological memory, so vaccines provide them no protection, and live attenuated vaccines (e.g., for varicella-zoster, measles virus, rotavirus, poliovirus) can actually cause the infection they are intended to prevent. The most common form is X-linked SCID, which accounts for nearly 50% of all cases and occurs primarily in males. Patients with SCID are typically diagnosed within the first few months of life after developing severe, often life-threatening, opportunistic infection by *Candida* spp., *Pneumocystis jirovecii*, or pathogenic strains of *E. coli*.

Without treatment, babies with SCID do not typically survive infancy. In some cases, a bone marrow transplant may successfully correct the defects in lymphocyte development that lead to the SCID phenotype, by replacing the defective component. However, this treatment approach is not without risks, as demonstrated by the famous case of David Vetter (1971–1984), better known as "Bubble Boy" (Figure 25.4.1). Vetter, a patient with SCID who lived in a protective plastic bubble to prevent exposure to opportunistic microbes, received a bone marrow transplant from his sister. Because of a latent Epstein-Barr virus infection in her bone marrow, however, he developed mononucleosis and died of Burkitt lymphoma at the age of 12 years.



Figure 25.4.1: David Vetter, popularly known as "The Bubble Boy," was born with SCID and lived most of his life isolated inside a plastic bubble. Here he is shown outside the bubble in a suit specially built for him by NASA. (credit: NASA Johnson Space Center)

Secondary Immunodeficiency

A secondary immunodeficiency occurs as a result an acquired impairment of function of B cells, T cells, or both. Secondary immunodeficiencies can be caused by:

- Systemic disorders such as diabetes mellitus, malnutrition, hepatitis, or HIV infection
- Immunosuppressive treatments such as cytotoxic chemotherapy, bone marrow ablation before transplantation, or radiation therapy
- Prolonged critical illness due to infection, surgery, or trauma in the very young, elderly, or hospitalized patients

Unlike primary immunodeficiencies, which have a genetic basis, secondary immunodeficiencies are often reversible if the underlying cause is resolved. Patients with secondary immunodeficiencies develop an increased susceptibility to an otherwise benign infection by opportunistic pathogens such as *Candida* spp., *P. jirovecii*, and *Cryptosporidium*.

HIV infection and the associated acquired immunodeficiency syndrome (AIDS) are the best-known secondary immunodeficiencies. AIDS is characterized by profound CD4 T-cell lymphopenia (decrease in lymphocytes). The decrease in CD4 T cells is the result of various mechanisms, including HIV-induced pyroptosis (a type of apoptosis that stimulates an inflammatory response), viral cytopathic effect, and cytotoxicity to HIV-infected cells.

The most common cause of secondary immunodeficiency worldwide is severe malnutrition, which affects both innate and adaptive immunity. More research and information are needed for the more common causes of secondary immunodeficiency; however, the number of new discoveries in AIDS research far exceeds that of any other single cause of secondary immunodeficiency. AIDS research has paid off extremely well in terms of discoveries and treatments; increased research into the most common cause of immunodeficiency, malnutrition, would likely be as beneficial.

An Immunocompromised Host

Benjamin, a 50-year-old male patient who has been receiving chemotherapy to treat his chronic myelogenous leukemia (CML), a disease characterized by massive overproduction of nonfunctional, malignant myelocytic leukocytes that crowd out other, healthy leukocytes, is seen in the emergency department. He is complaining of a productive, wet cough, dyspnea, and fatigue. On examination, his pulse is 120 beats per minute (bpm) (normal range is 60–100 bpm) and weak, and his blood pressure is 90/60 mm Hg (normal is 120/80 mm Hg). During auscultation, a distinct crackling can be heard in his lungs as he breathes, and his pulse-oximeter level (a measurement of blood-oxygen saturation) is 80% (normal is 95%–100%). He has a fever; his temperature is 38.9 °C (102 °F). Sputum cultures and blood samples are obtained and sent to the lab, but Benjamin goes into respiratory distress and dies before the results can be obtained.

Benjamin's death was a result of a combination of his immune system being compromised by his leukemia and his chemotherapy treatment further weakening his ability to mount an immune response. CML (and leukemia in general) and corresponding chemotherapy cause a decrease in the number of leukocytes capable of normal function, leading to secondary immunodeficiency. This increases the risk for opportunistic bacterial, viral, protozoal, and fungal infections that could include *Staphylococcus*, enteroviruses, *Pneumocystis*, *Giardia*, or *Candida*. Benjamin's symptoms were suggestive of bacterial pneumonia, but his leukemia and chemotherapy likely complicated and contributed to the severity of the pneumonia, resulting in his death. Because his leukemia was overproducing certain white blood cells, and those overproduced white blood cells were largely nonfunctional or abnormal in their function, he did not have the proper immune system blood cells to help him fight off the infection.

Table **25.4.1** summarizes primary and secondary immunodeficiencies, their effects on immune function, and typical outcomes.

Table **25.4.1**: Primary and Secondary Immunodeficiencies

Disease		Effect on Immune Function	Outcomes
Primary immunodeficiencies	Chronic granulomatous disease	Impaired killing of bacteria within the phagolysosome of neutrophils and macrophages	Chronic infections and granulomas
	Selective IgA deficiency	Inability to produce secretory IgA	Predisposition to lung and gastrointestinal infections
	Severe combined immunodeficiency disease (SCID)	Deficient humoral and cell-mediated immune responses	Early development of severe and life-threatening opportunistic infections
	X-linked agammaglobulinemia	Flawed differentiation of B cells and absence of specific antibodies	Recurrent infections almost exclusively due to pathogens that cause pyogenic infections
Secondary immunodeficiencies	Immunosuppressive therapies (e.g., chemotherapy, radiotherapy)	Impaired humoral and/or cell-mediated immune responses	Opportunistic infections, rare cancers
	Malnutrition	Impaired humoral and/or cell-mediated immune responses	Opportunistic infections, rare cancers
	Viral infection (e.g., HIV)	Impaired cell-mediated immune responses due to CD4 T-cell lymphopenia	Opportunistic infections, rare cancers

Key Concepts and Summary

- **Primary immunodeficiencies** are caused by genetic abnormalities; **secondary immunodeficiencies** are acquired through disease, diet, or environmental exposures.
- Primary immunodeficiencies may result from flaws in phagocyte killing of innate immunity, or impairment of T cells and B cells.
- Primary immunodeficiencies include chronic granulomatous disease, X-linked agammaglobulinemia, selective IgA deficiency, and severe combined immunodeficiency disease.
- Secondary immunodeficiencies result from environmentally induced defects in B cells and/or T cells.
- Causes for secondary immunodeficiencies include malnutrition, viral infection, diabetes, prolonged infections, and chemical or radiation exposure.

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25.E: Diseases of the Immune System (Exercises)

25.1: Organ Transplantation and Rejection

Grafts and transplants can be classified as autografts, isografts, allografts, or xenografts based on the genetic differences between the donor's and recipient's tissues. Genetic differences, especially among the MHC (HLA) genes, will dictate the likelihood that rejection of the transplanted tissue will occur. Transplant recipients usually require immunosuppressive therapy to avoid rejection, even with good genetic matching.

Matching

Match the graft with its description.

___autograft	A. donor is a different species than the recipient
___allograft	B. donor and recipient are the same individual
___xenograft	C. donor is an identical twin of the recipient
___isograft	D. donor is the same species as the recipient, but genetically different

Answer

B, D, A, C

Fill in the Blank

For a transplant to have the best chances of avoiding rejection, the genes coding for the _____ molecules should be closely matched between donor and recipient.

Answer

MHC

Because it is a "transplant" that can include APCs and T cells from the donor, a bone marrow transplant may induce a very specific type of rejection known as _____ disease.

Answer

graft-versus-host

Short Answer

Why is a parent usually a better match for transplanted tissue to a donor than a random individual of the same species?

25.2: Immunodeficiency

Primary immunodeficiencies are caused by genetic abnormalities; secondary immunodeficiencies are acquired through disease, diet, or environmental exposures. Primary immunodeficiencies may result from flaws in phagocyte killing of innate immunity, or impairment of T cells and B cells. Primary immunodeficiencies include chronic granulomatous disease, X-linked agammaglobulinemia, selective IgA deficiency, and severe combined immunodeficiency disease.

Multiple Choice

Which of the following is a genetic disease that results in lack of production of antibodies?

A. agammaglobulinemia
B. myasthenia gravis
C. HIV/AIDS
D. chronic granulomatous disease

Answer

A

Which of the following is a genetic disease that results in almost no adaptive immunity due to lack of B and/ or T cells?

A. agammaglobulinemia
B. severe combined immunodeficiency
C. HIV/AIDS
D. chronic granulomatous disease

Answer

B

All but which one of the following are examples of secondary immunodeficiencies?

A. HIV/AIDS
B. malnutrition
C. chronic granulomatous disease
D. immunosuppression due to measles infection

Answer

C

Fill in the Blank

Diseases due to _____ abnormalities are termed primary immunodeficiencies.

Answer

genetic

A secondary immunodeficiency is _____, rather than genetic.

Answer

acquired

Short Answer

Compare the treatments for primary and secondary immunodeficiencies.

25.3: Cancer Immunobiology and Immunotherapy

When control of the cell cycle is lost, the affected cells rapidly divide and often lose the ability to differentiate into the cell type appropriate for their location in the body. In addition, they lose contact inhibition and can start to grow on top of each other. This can result in formation of a tumor. It is important to make a distinction here: The term “cancer” is used to describe the diseases resulting from loss of cell-cycle regulation and subsequent cell proliferation.

Multiple Choice

Cancer results when a mutation leads to which of the following?

A. cell death
B. apoptosis
C. loss of cell-cycle control
D. shutdown of the cell cycle

Answer

C

Tumor antigens are _____ that are inappropriately expressed and found on abnormal cells.

A. self antigens
B. foreign antigens
C. antibodies
D. T-cell receptors

Answer

A

Fill in the Blank

A _____ cancer vaccine is one that stops the disease from occurring in the first place.

Answer

preventive

A _____ cancer vaccine is one that will help to treat the disease after it has occurred.

Answer

therapeutic

Short Answer

How can tumor antigens be effectively targeted without inducing an autoimmune (anti-self) response?

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CHAPTER OVERVIEW

26: Laboratory Analysis of the Immune Response

Many laboratory tests are designed to confirm a presumptive diagnosis by detecting antibodies specific to a suspected pathogen. Unfortunately, many such tests are time-consuming and expensive. That is now changing, however, with the development of new, miniaturized technologies that are fast and inexpensive. For example, researchers at Columbia University are developing a "lab-on-a-chip" technology that will test a single drop of blood for 15 different infectious diseases, including HIV and syphilis, in a matter of minutes.[1] The blood is pulled through tiny capillaries into reaction chambers where the patient's antibodies mix with reagents. A chip reader that attaches to a cell phone analyzes the results and sends them to the patient's healthcare provider. Currently the device is being field tested in Rwanda to check pregnant women for chronic diseases. Researchers estimate that the chip readers will sell for about $100 and individual chips for $1.[2]

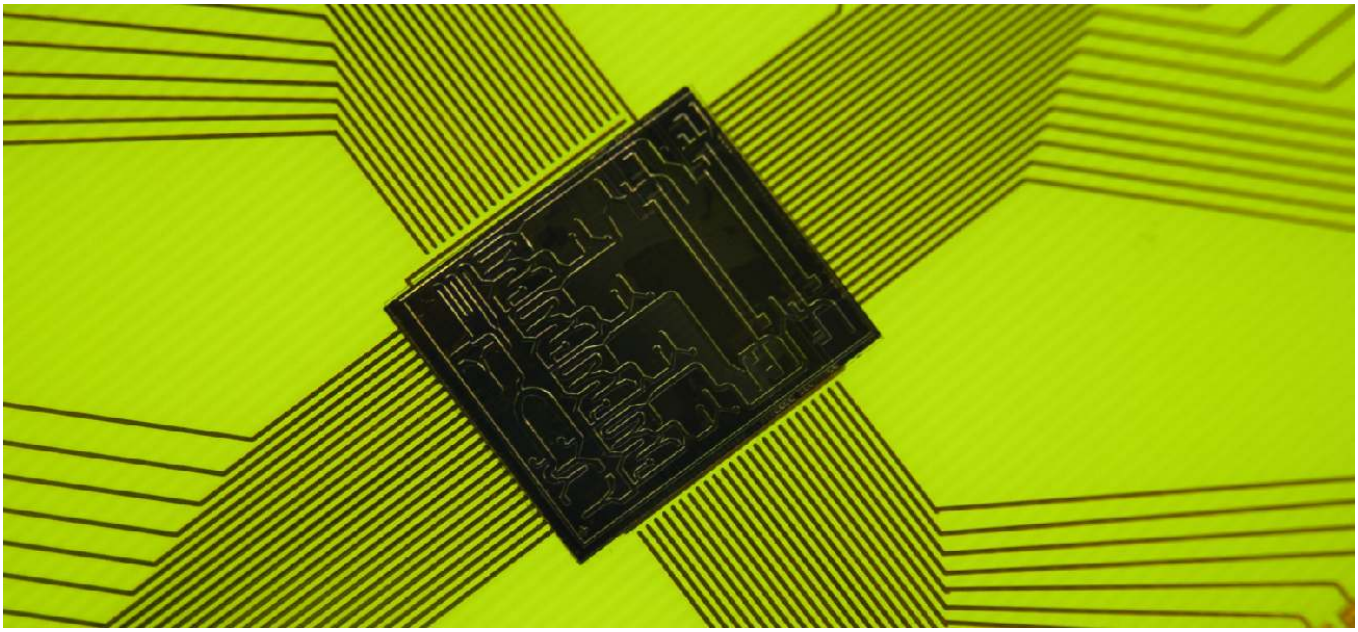


Figure **26.1**: Lab-on-a-chip technology allows immunological assays to be miniaturized so tests can be done rapidly with minimum quantities of expensive reagents. The chips contain tiny flow tubes to allow movement of fluids by capillary action, reactions sites with embedded reagents, and data output through electronic sensors. (credit: modification of work by Maggie Bartlett, NHGRI)

26.1: Practical Applications of Monoclonal and Polyclonal Antibodies

26.2: Detecting Antigen-Antibody Complexes in vitro

26.3: Agglutination Assays

26.4: Enzyme Immunoassays (EIA) and Enzyme-Linked Immunosorbent Assays (ELISA)

26.5: Fluorescent Auto-Antibody Techniques

26.E: Laboratory Analysis of the Immune Response (Exercises)

Footnotes

1. 1 Chin, Curtis D. et al., "Mobile Device for Disease Diagnosis and Data Tracking in Resource-Limited Settings," *Clinical Chemistry* 59, no. 4 (2013): 629-40.
2. 2 Evarts, H., "Fast, Low-Cost Device Uses the Cloud to Speed Up Testing for HIV and More," January 24, 2013. Accessed July 14, 2016. http://engineering.columbia.edu/fast...g-hiv-and-more.

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26.1: Practical Applications of Monoclonal and Polyclonal Antibodies

Learning Objectives

- Compare the method of development, use, and characteristics of monoclonal and polyclonal antibodies
- Explain the nature of antibody cross-reactivity and why it occurs
- Compare and contrast false-positive vs. false-negative test results

In addition to being crucial for our normal immune response, antibodies provide powerful tools for research and diagnostic purposes. The high specificity of antibodies makes them an excellent tool for detecting and quantifying a broad array of targets, from drugs to serum proteins to microorganisms. With *in vitro* assays, antibodies can be used to precipitate soluble antigens, agglutinate (clump) cells, opsonize and kill bacteria with the assistance of complement, and neutralize drugs, toxins, and viruses.

An antibody's **specificity** results from the antigen-binding site formed within the variable regions—regions of the antibody that have unique patterns of amino acids that can only bind to target antigens with a molecular sequence that provides complementary charges and noncovalent bonds. There are limitations to antibody specificity, however. Some antigens are so chemically similar that cross-reactivity occurs; in other words, antibodies raised against one antigen bind to a chemically similar but different antigen. Consider an antigen that consists of a single protein with multiple epitopes (Figure 26.1.1). This single protein may stimulate the production of many different antibodies, some of which may bind to chemically identical epitopes on other proteins.

Cross-reactivity is more likely to occur between antibodies and antigens that have low **affinity** or **avidity**. Affinity, which can be determined experimentally, is a measure of the binding strength between an antibody's binding site and an epitope, whereas avidity is the total strength of all the interactions in an antibody-antigen complex (which may have more than one bonding site). Avidity is influenced by affinity as well as the structural arrangements of the epitope and the variable regions of the antibody. If an antibody has a high affinity/avidity for a specific antigen, it is less likely to cross-react with an antigen for which it has a lower affinity/avidity.

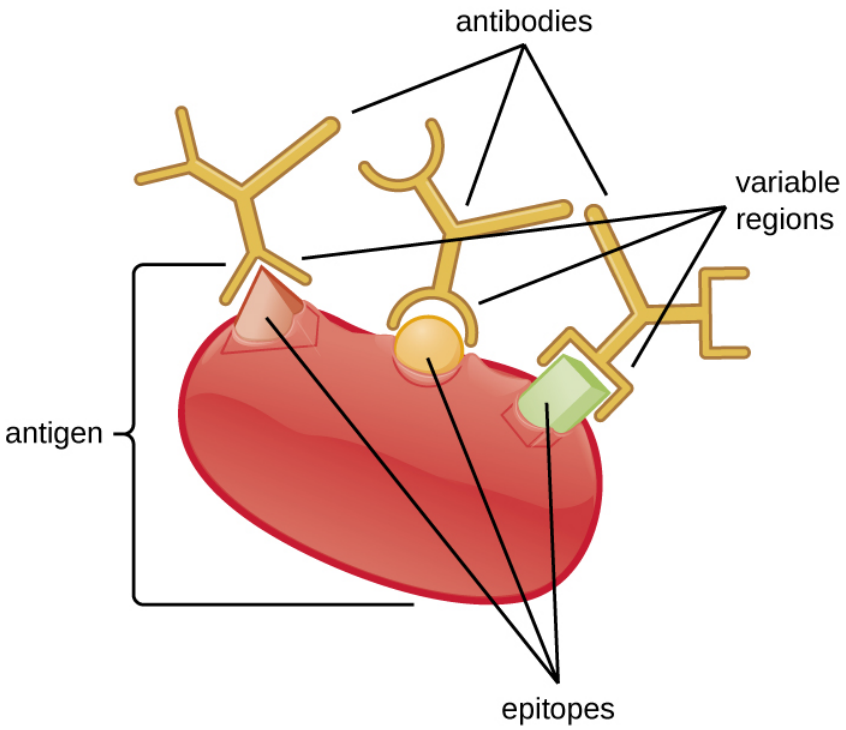


Figure 26.1.1: An antibody binds to a specific region on an antigen called an epitope. A single antigen can have multiple epitopes for different, specific antibodies.

Producing Polyclonal Antibodies

Antibodies used for research and diagnostic purposes are often obtained by injecting a lab animal such as a rabbit or a goat with a specific antigen. Within a few weeks, the animal's immune system will produce high levels of antibodies specific for the antigen. These antibodies can be harvested in an **antiserum**, which is whole serum collected from an animal following exposure to an antigen. Because most antigens are complex structures with multiple epitopes, they result in the production of multiple antibodies in the lab animal. This so-called **polyclonal antibody** response is also typical of the response to infection by the human immune system. Antiserum drawn from an animal will thus contain antibodies from multiple clones of B cells, with each B cell responding to a specific epitope on the antigen (Figure 26.1.2).

Lab animals are usually injected at least twice with antigen when being used to produce antiserum. The second injection will activate memory cells that make class IgG antibodies against the antigen. The memory cells also undergo **affinity maturation**, resulting in a pool of antibodies with higher average affinity. Affinity maturation occurs because of mutations in the immunoglobulin gene variable regions, resulting in B cells with slightly altered antigen-binding sites. On re-exposure to the antigen, those B cells capable of producing antibody with higher affinity antigen-binding sites will be stimulated to proliferate and produce more antibody than their lower-affinity peers. An adjuvant, which is a chemical that provokes a generalized activation of the immune system that stimulates greater antibody production, is often mixed with the antigen prior to injection.

Antiserum obtained from animals will not only contain antibodies against the antigen artificially introduced in the laboratory, but it will also contain antibodies to any other antigens to which the animal has been exposed during its lifetime. For this reason, antisera must first be "purified" to remove other antibodies before using the antibodies for research or diagnostic assays.

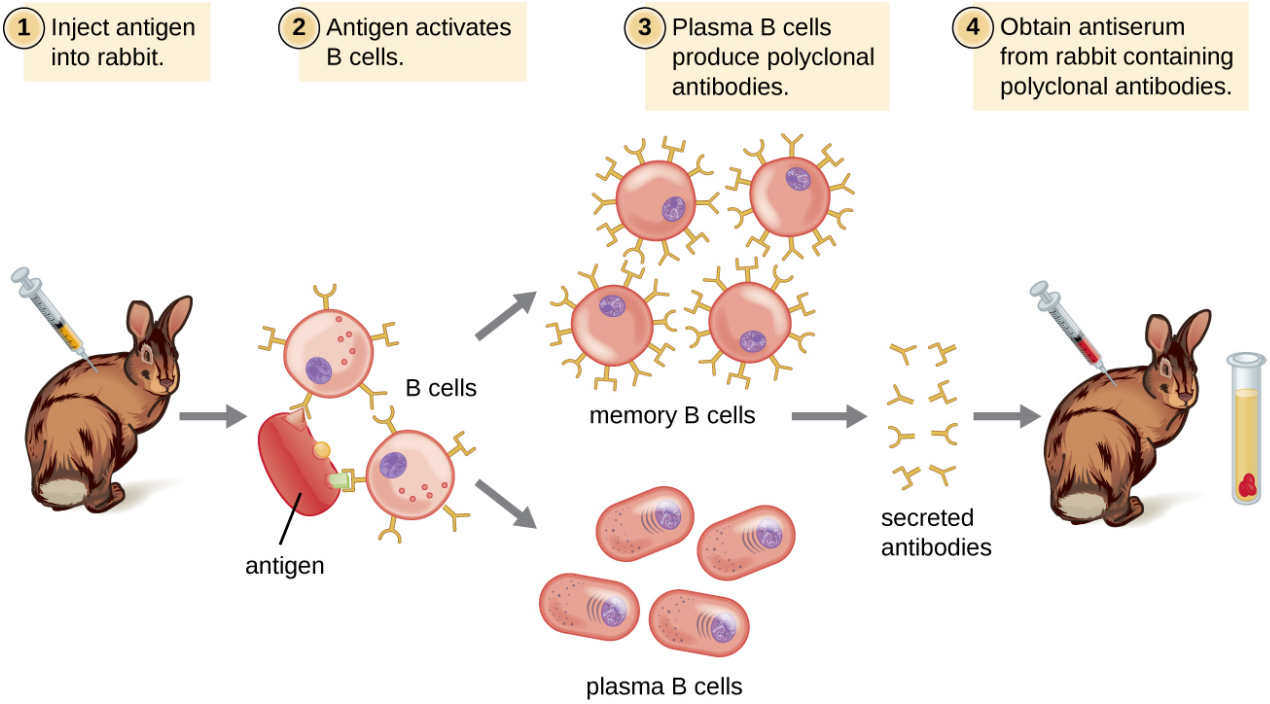


Figure 26.1.2: This diagram illustrates the process for harvesting polyclonal antibodies produced in response to an antigen.

Clinical Uses of Polyclonal Antisera

Polyclonal antisera are used in many clinical tests that are designed to determine whether a patient is producing antibodies in response to a particular pathogen. While these tests are certainly powerful diagnostic tools, they have their limitations, because they are an indirect means of determining whether a particular pathogen is present. Tests based on a polyclonal response can sometimes lead to a **false-positive** result—in other words, a test that confirms the presence of an antigen that is, in fact, not present. Antibody-based tests can also result in a **false-negative** result, which occurs when the test fails to detect an antibody that is, in fact, present.

The accuracy of antibody tests can be described in terms of **test sensitivity** and **test specificity**. Test sensitivity is the probability of getting a positive test result when the patient is indeed infected. If a test has high sensitivity, the probability of a false negative is

low. Test specificity, on the other hand, is the probability of getting a negative test result when the patient is not infected. If a test has high specificity, the probability of a false positive is low.

False positives often occur due to cross-reactivity, which can occur when epitopes from a different pathogen are similar to those found on the pathogen being tested for. For this reason, antibody-based tests are often used only as screening tests; if the results are positive, other confirmatory tests are used to make sure that the results were not a false positive.

For example, a blood sample from a patient suspected of having hepatitis C can be screened for the virus using antibodies that bind to antigens on hepatitis C virus. If the patient is indeed infected with hepatitis C virus, the antibodies will bind to the antigens, yielding a positive test result. If the patient is not infected with hepatitic C virus, the antibodies will generally not bind to anything and the test should be negative; however, a false positive may occur if the patient has been previously infected by any of a variety of pathogens that elicit antibodies that cross-react with the hepatitis C virus antigens. Antibody tests for hepatitis C have high sensitivity (a low probability of a false negative) but low specificity (a high probability of a false positive). Thus, patients who test positive must have a second, confirmatory test to rule out the possibility of a false positive. The confirmatory test is a more expensive and time-consuming test that directly tests for the presence of hepatitis C viral RNA in the blood. Only after the confirmatory test comes back positive can the patient be definitively diagnosed with a hepatitis C infection. Antibody-based tests can result in a false negative if, for any reason, the patient's immune system has not produced detectable levels of antibodies. For some diseases, it may take several weeks following infection before the immune system produces enough antibodies to cross the detection threshold of the assay. In immunocompromised patients, the immune system may not be capable of producing a detectable level of antibodies.

Another limitation of using antibody production as an indicator of disease is that antibodies in the blood will persist long after the infection has been cleared. Depending on the type of infection, antibodies will be present for many months; sometimes, they may be present for the remainder of the patient's life. Thus, a positive antibody-based test only means that the patient was infected at some point in time; it does not prove that the infection is active.

In addition to their role in diagnosis, polyclonal antisera can activate complement, detect the presence of bacteria in clinical and food industry settings, and perform a wide array of precipitation reactions that can detect and quantify serum proteins, viruses, or other antigens. However, with the many specificities of antibody present in a polyclonal antiserum, there is a significant likelihood that the antiserum will cross-react with antigens to which the individual was never exposed. Therefore, we must always account for the possibility of false-positive results when working with a polyclonal antiserum.

Key Concepts and Summary

- Antibodies bind with high **specificity** to antigens used to challenge the immune system, but they may also show **cross-reactivity** by binding to other antigens that share chemical properties with the original antigen.
- Injection of an antigen into an animal will result in a **polyclonal antibody** response in which different antibodies are produced that react with the various epitopes on the antigen.
- **Polyclonal antisera** are useful for some types of laboratory assays, but other assays require more specificity. Diagnostic tests that use polyclonal antisera are typically only used for screening because of the possibility of **false-positive** and **false-negative** results.

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26.2: Detecting Antigen-Antibody Complexes in vitro

Learning Objectives

- Describe various types of assays used to find antigen-antibody complexes
- Explain how antibodies in patient serum can be used to diagnose disease
- Describe the steps of Western blot
- Explain the function of the enzyme or the fluorophore in immuno assays

Laboratory tests to detect antibodies and antigens outside of the body (e.g., in a test tube) are called ***in vitro* assays**. When both antibodies and their corresponding antigens are present in a solution, we can often observe a precipitation reaction in which large complexes (lattices) form and settle out of solution. In the next several sections, we will discuss several common *in vitro* assays.

Neutralization Assay

To cause infection, viruses must bind to receptors on host cells. Antiviral antibodies can neutralize viral infections by coating the virions, blocking the binding (Figure 18.1.6). This activity neutralizes virions and can result in the formation of large antibody-virus complexes (which are readily removed by phagocytosis) or by antibody binding to the virus and blocking its binding to host cell receptors. This neutralization activity is the basis of **neutralization assays**, sensitive assays used for diagnoses of viral infections.

When viruses infect cells, they often cause damage (cytopathic effects) that may include lysis of the host cells. Cytopathic effects can be visualized by growing host cells in a petri dish, covering the cells with a thin layer of agar, and then adding virus (see Isolation, Culture, and Identification of Viruses). The virus will diffuse very slowly through the agar. A virus will enter a host cell, proliferate (causing cell damage), be released from the dead host cell, and then move to neighboring cells. As more and more cells die, plaques of dead cells will form (Figure 26.2.6).

During the course of a viral infection, the patient will mount an antibody response to the virus, and we can quantify those antibodies using a **plaque reduction assay**. To perform the assay, a serial dilution is carried out on a serum sample. Each dilution is then mixed with a standardized amount of the suspect virus. Any virus-specific antibodies in the serum will neutralize some of the virus. The suspensions are then added to host cells in culture to allow any nonneutralized virus to infect the cells and form plaques after several days. The titer is defined as the reciprocal of the highest dilution showing a 50% reduction in plaques. Titer is always expressed as a whole number. For example, if a 1/64 dilution was the highest dilution to show 50% plaque reduction, then the titer is 64.

The presence of antibodies in the patient's serum does not tell us whether the patient is currently infected or was infected in the past. Current infections can be identified by waiting two weeks and testing another serum sample. A four-fold increase in neutralizing titer in this second sample indicates a new infection.

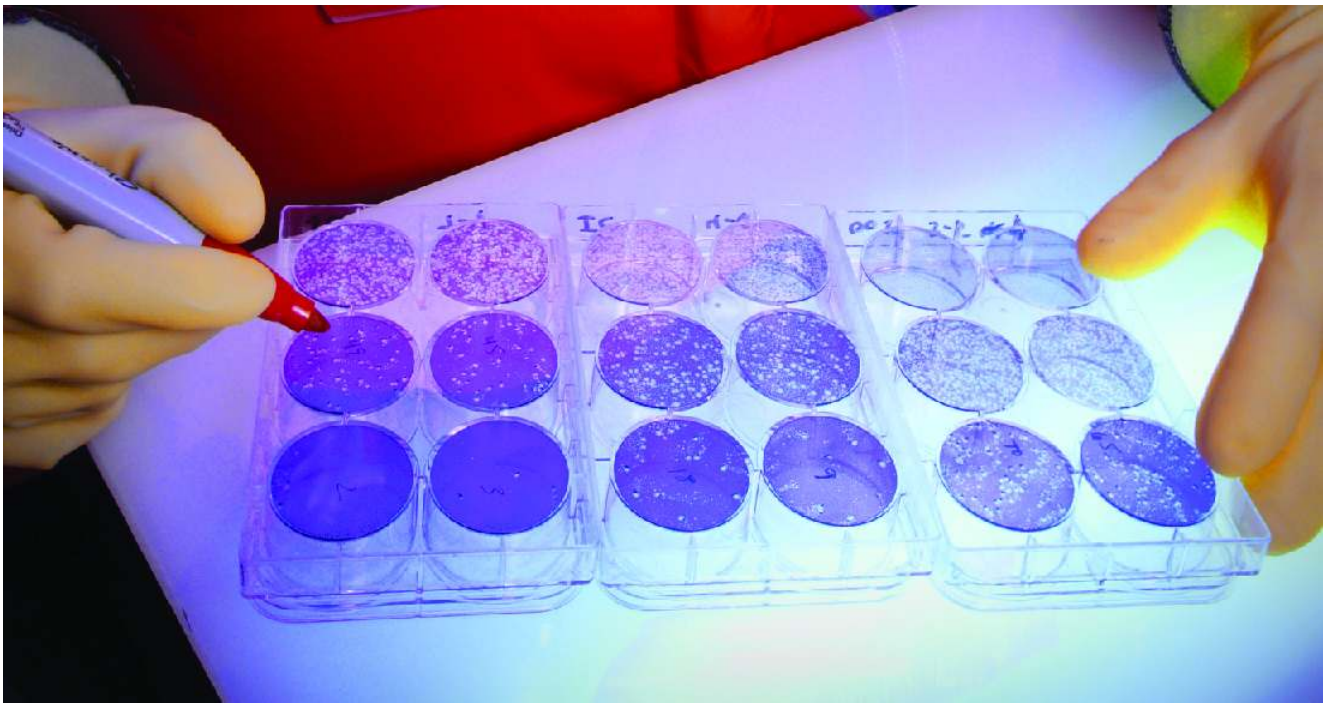


Figure 26.2.6: In a neutralization assay, antibodies in patient serum neutralize viruses added to the wells, preventing the formation of plaques. In the assay pictured, the wells with numerous plaques (white patches) contain a low concentration of antibodies. The wells with relatively few plaques have a high concentration of antibodies. (credit: modification of work by Centers for Disease Control and Prevention)

Immunoelectrophoresis

When a patient has elevated protein levels in the blood or is losing protein in the urine, a clinician will often order a **polyacrylamide gel electrophoresis (PAGE) assay** (see Visualizing and Characterizing DNA, RNA, and Protein). This assay compares the relative abundance of the various types of serum proteins. Abnormal protein electrophoresis patterns can be further studied using **immunoelectrophoresis (IEP)**. The IEP begins by running a PAGE. Antisera against selected serum proteins are added to troughs running parallel to the electrophoresis track, forming precipitin arcs similar to those seen in an Ouchterlony assay (Figure 26.2.7). This allows the identification of abnormal immunoglobulin proteins in the sample.

IEP is particularly useful in the diagnosis of multiple myeloma, a cancer of antibody-secreting cells. Patients with multiple myeloma cannot produce healthy antibodies; instead they produce abnormal antibodies that are monoclonal proteins (M proteins). Thus, patients with multiple myeloma will present with elevated serum protein levels that show a distinct band in the gamma globulin region of a protein electrophoresis gel and a sharp spike (in M protein) on the densitometer scan rather than the normal broad smear (Figure 26.2.7). When antibodies against the various types of antibody heavy and light chains are used to form precipitin arcs, the M protein will cause distinctly skewed arcs against one class of heavy chain and one class of light chain as seen in Figure 26.2.7.

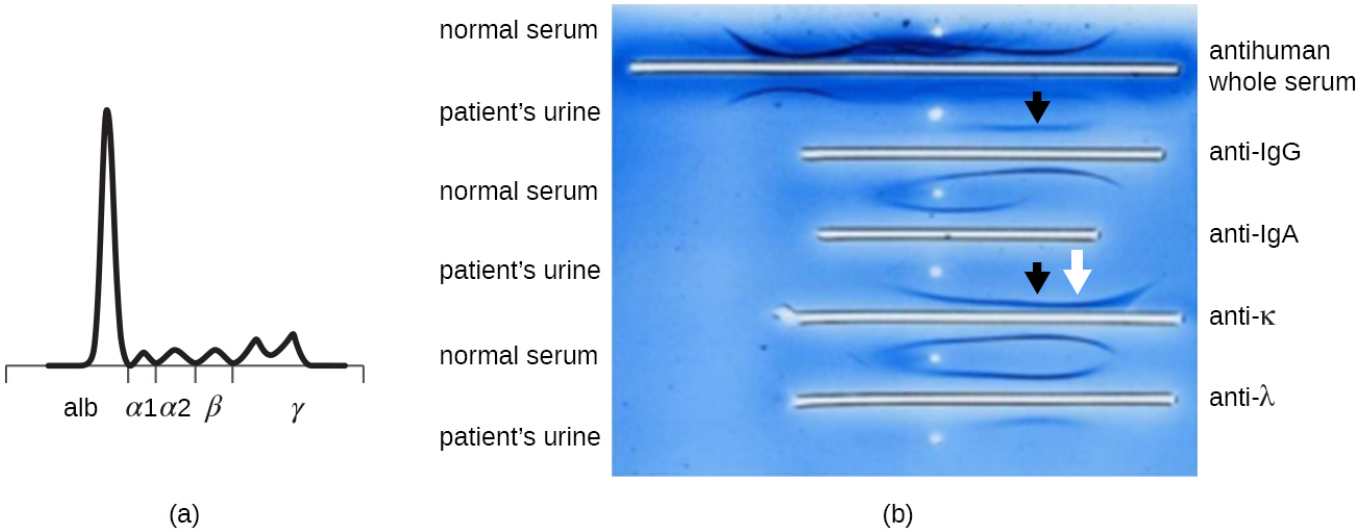


Figure **26.2.7**: (a) This graph shows normal measurements of serum proteins. (b) This photograph shows an immunoelectrophoresis of urine. After electrophoresis, antisera were added to the troughs and the precipitin arcs formed, illustrating the distribution of specific proteins. The skewed arcs (arrows) help to diagnose multiple myeloma. (credit a, b: modification of work by Izawa S, Akimoto T, Ikeuchi H, Kusano E, Nagata D)

Protein Electrophoresis and the Characterization of Immunoglobulin Structure

The advent of electrophoresis ultimately led to researching and understanding the structure of antibodies. When Swedish biochemist Arne Tiselius (1902–1971) published the first protein electrophoresis results in 1937,[2] he could identify the protein albumin (the smallest and most abundant serum protein) by the sharp band it produced in the gel. The other serum proteins could not be resolved in a simple protein electrophoresis, so he named the three broad bands, with many proteins in each band, alpha, beta, and gamma globulins. Two years later, American immunologist Elvin Kabat (1914–2000) traveled to Sweden to work with Tiselius using this new technique and showed that antibodies migrated as gamma globulins.[3] With this new understanding in hand, researchers soon learned that multiple myeloma, because it is a cancer of antibody-secreting cells, could be tentatively diagnosed by the presence of a large M spike in the gamma-globulin region by protein electrophoresis. Prior to this discovery, studies on immunoglobulin structure had been minimal, because of the difficulty of obtaining pure samples to study. Sera from multiple myeloma patients proved to be an excellent source of highly enriched monoclonal immunoglobulin, providing the raw material for studies over the next 20-plus years that resulted in the elucidation of the structure of immunoglobulin.

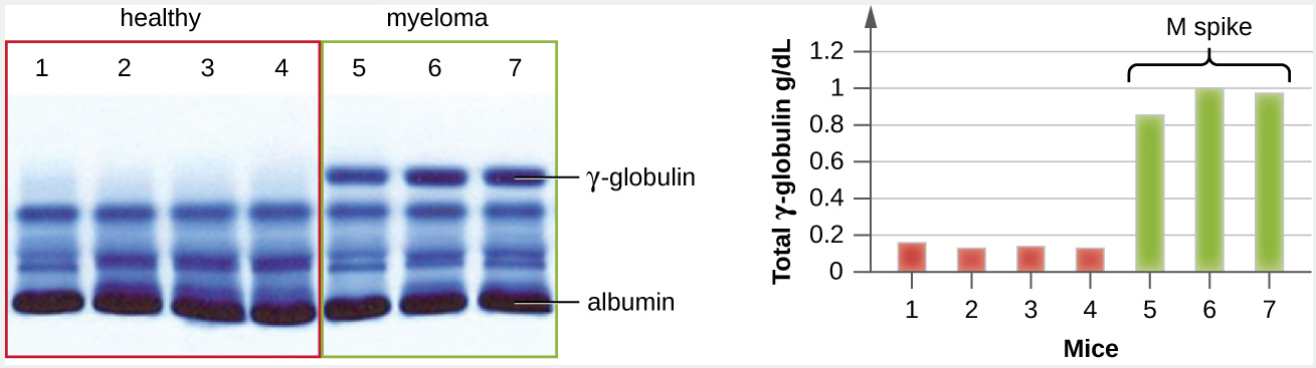


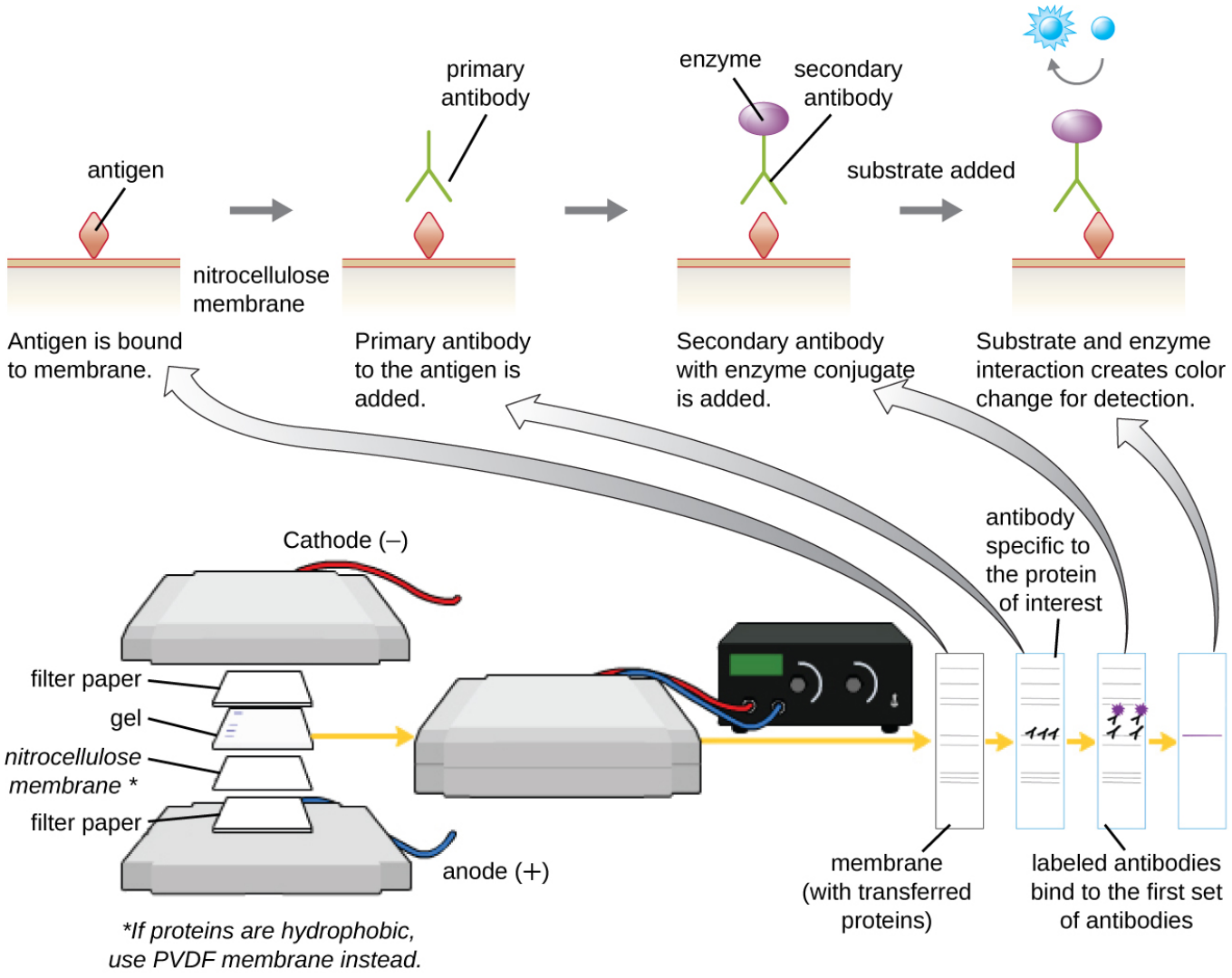
Figure **26.2.8**: Electrophoresis patterns of myeloma (right) and normal sera (left). The proteins have been stained; when the density of each band is quantified by densitometry, the data produce the bar graph on the right. Both gels show the expected dense band of albumin at the bottom and an abnormal spike in the gamma-globulin region. (credit: modification of work by Soodgupta D, Hurchla MA, Jiang M, Zheleznyak A, Weilbaecher KN, Anderson CJ, Tomasson MH, Shokeen M)

Immunoblot Assay: The Western Blot

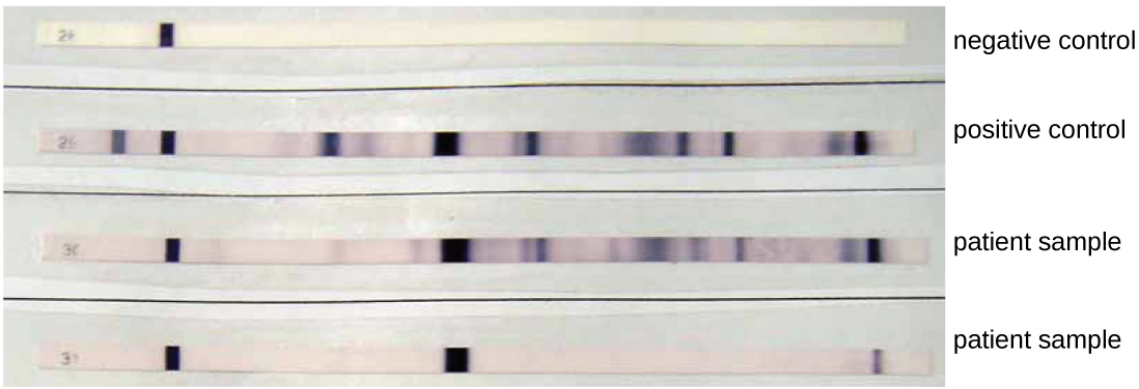
After performing protein gel electrophoresis, specific proteins can be identified in the gel using antibodies. This technique is known as the western blot. Following separation of proteins by PAGE, the protein antigens in the gel are transferred to and immobilized on a nitrocellulose membrane. This membrane can then be exposed to a primary antibody produced to specifically bind to the protein of interest. A second antibody equipped with a molecular beacon will then bind to the first. These secondary antibodies are coupled to another molecule such as an enzyme or a fluorophore (a molecule that fluoresces when excited by light). When using antibodies coupled to enzymes, a chromogenic substrate for the enzyme is added. This substrate is usually colorless but will develop color in the presence of the antibody. The fluorescence or substrate coloring identifies the location of the specific protein in the membrane to which the antibodies are bound (Figure **26.2.9**).

Typically, polyclonal antibodies are used for western blot assays. They are more sensitive than mAbs because of their ability to bind to various epitopes of the primary antigen, and the signal from polyclonal antibodies is typically stronger than that from mAbs. Monoclonal antibodies can also be used; however, they are much more expensive to produce and are less sensitive, since they are only able to recognize one specific epitope.

Several variations of the western blot are useful in research. In a southwestern blot, proteins are separated by SDS-PAGE, blotted onto a nitrocellulose membrane, allowed to renature, and then probed with a fluorescently or radioactively labeled DNA probe; the purpose of the southwestern is to identify specific DNA-protein interactions. Far-western blotsare carried out to determine protein-protein interactions between immobilized proteins (separated by SDS-PAGE, blotted onto a nitrocellulose membrane, and allowed to renature) and non-antibody protein probes. The bound non-antibody proteins that interact with the immobilized proteins in a far-western blot may be detected by radiolabeling, fluorescence, or the use of an antibody with an enzymatic molecular beacon.



(a)



(b)

Figure 26.2.9: (a) This diagram summarizes the process of western blotting. Antibodies are used to identify specific bands on the protein gel. (b) A western blot test for antibodies against HIV. The top strip is the negative control; the next strip is the positive control. The bottom two strips are patient serum samples containing antibodies. (credit a: modification of work by "Bensaccount"/Wikimedia Commons)

Key Concepts and Summary

- Viral infections can be detected by quantifying virus-neutralizing antibodies in a patient's serum.
- Different antibody classes in plasma or serum are identified by using **immunoelectrophoresis**.
- The presence of specific antigens (e.g., bacterial or viral proteins) in serum can be demonstrated by **western blot** assays, in which the proteins are transferred to a nitrocellulose membrane and identified using labeled antibodies.

Footnotes

1. 1 Ouchterlony, Örjan, "In Vitro Method for Testing the Toxin-Producing Capacity of Diphtheria Bacteria," *Acta Pathologica Microbiologica Scandinavica* 26, no. 4 (1949): 516-24.
2. 2 Tiselius, Arne, "Electrophoresis of Serum Globulin: Electrophoretic Analysis of Normal and Immune Sera," *Biochemical Journal* 31, no. 9 (1937): 1464.
3. 3 Tiselius, Arne and Elvin A. Kabat. "An Electrophoretic Study of Immune Sera and Purified Antibody Preparations," *The Journal of Experimental Medicine* 69, no. 1 (1939): 119-31.

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26.3: Agglutination Assays

Learning Objectives

- Compare direct and indirect agglutination
- Identify various uses of hemagglutination in the diagnosis of disease
- Explain how blood types are determined
- Explain the steps used to cross-match blood to be used in a transfusion

In addition to causing precipitation of soluble molecules and flocculation of molecules in suspension, antibodies can also clump together cells or particles (e.g., antigen-coated latex beads) in a process called **agglutination** (Figure 18.1.8). Agglutination can be used as an indicator of the presence of antibodies against bacteria or red blood cells. Agglutination assays are usually quick and easy to perform on a glass slide or microtiter plate (Figure **26.3.1**). Microtiter plates have an array of wells to hold small volumes of reagents and to observe reactions (e.g., agglutination) either visually or using a specially designed spectrophotometer. The wells come in many different sizes for assays involving different volumes of reagents.

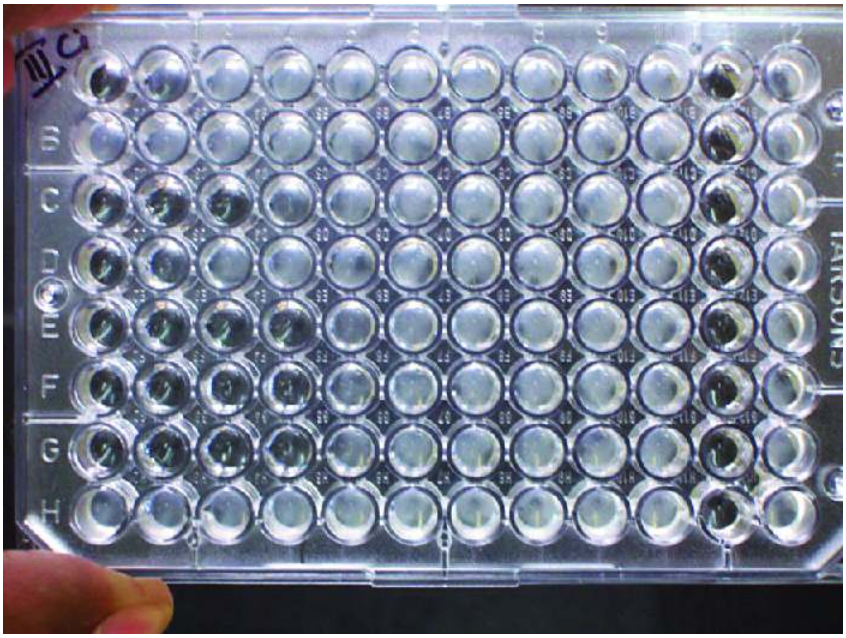


Figure **26.3.1**: Microtiter plates are used for conducting numerous reactions simultaneously in an array of wells. (credit: modification of work by "Microrao"/Wikimedia)

Agglutination of Bacteria and Viruses

The use of agglutination tests to identify streptococcal bacteria was developed in the 1920s by Rebecca Lancefieldworking with her colleagues A.R. Dochez and Oswald Avery.[1] She used antibodies to identify M protein, a virulence factor on streptococci that is necessary for the bacteria's ability to cause strep throat. Production of antibodies against M protein is crucial in mounting a protective response against the bacteria.

Lancefield used antisera to show that different strains of the same species of streptococci express different versions of M protein, which explains why children can come down with strep throat repeatedly. Lancefield classified beta-hemolytic streptococci into many groups based on antigenic differences in group-specific polysaccharides located in the bacterial cell wall. The strains are called **serovars** because they are differentiated using antisera. Identifying the serovars present in a disease outbreak is important because some serovars may cause more severe disease than others.

The method developed by Lancefield is a **direct agglutination assay**, since the bacterial cells themselves agglutinate. A similar strategy is more commonly used today when identifying serovars of bacteria and viruses; however, to improve visualization of the agglutination, the antibodies may be attached to inert latex beads. This technique is called an **indirect agglutination assay** (or latex

fixation assay), because the agglutination of the beads is a marker for antibody binding to some other antigen (Figure 26.3.2). Indirect assays can be used to detect the presence of either antibodies or specific antigens.

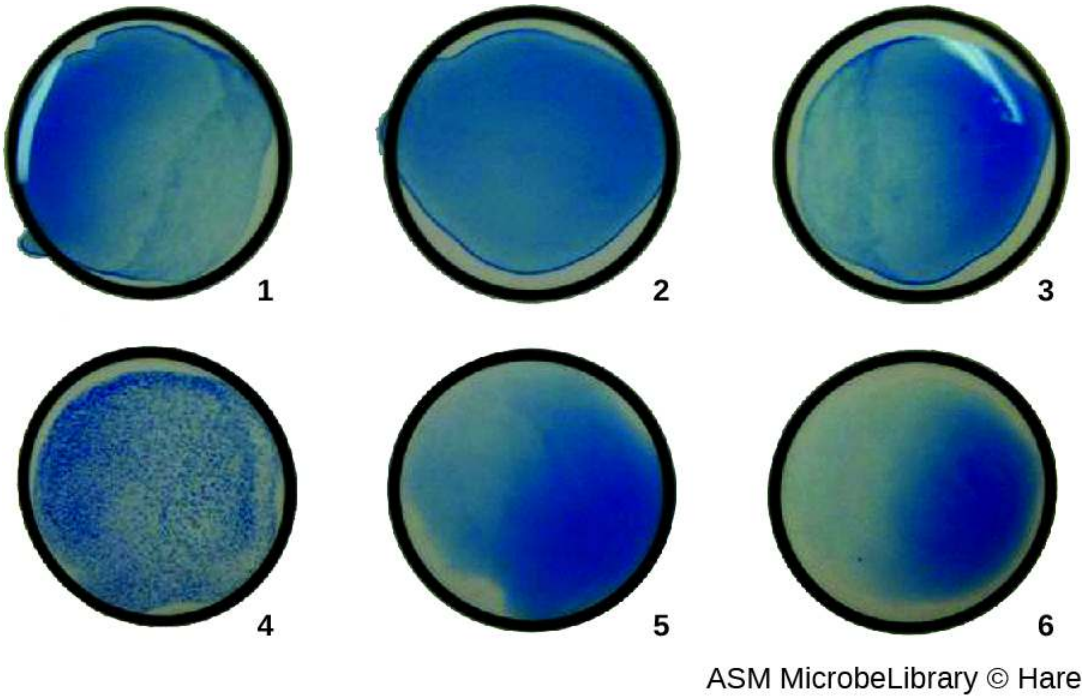


Figure 26.3.2: Antibodies against six different serovars of Group A strep were attached to latex beads. Each of the six antibody preparations was mixed with bacteria isolated from a patient. The tiny clumps seen in well 4 are indicative of agglutination, which is absent from all other wells. This indicates that the serovar associated with well 4 is present in the patient sample. (credit: modification of work by American Society for Microbiology)

To identify antibodies in a patient's serum, the antigen of interest is attached to latex beads. When mixed with patient serum, the antibodies will bind the antigen, cross-linking the latex beads and causing the beads to agglutinate indirectly; this indicates the presence of the antibody (Figure 26.3.3). This technique is most often used when looking for IgM antibodies, because their structure provides maximum cross-linking. One widely used example of this assay is a test for rheumatoid factor (RF) to confirm a diagnosis of rheumatoid arthritis. RF is, in fact, the presence of IgM antibodies that bind to the patient's own IgG. RF will agglutinate IgG-coated latex beads.

In the reverse test, soluble antigens can be detected in a patient's serum by attaching specific antibodies (commonly mAbs) to the latex beads and mixing this complex with the serum (Figure 26.3.3).

Agglutination tests are widely used in underdeveloped countries that may lack appropriate facilities for culturing bacteria. For example, the Widal test, used for the diagnosis of typhoid fever, looks for agglutination of *Salmonella enterica* subspecies *typhi* in patient sera. The Widal test is rapid, inexpensive, and useful for monitoring the extent of an outbreak; however, it is not as accurate as tests that involve culturing of the bacteria. The Widal test frequently produces false positives in patients with previous infections with other subspecies of *Salmonella*, as well as false negatives in patients with hyperproteinemia or immune deficiencies.

In addition, agglutination tests are limited by the fact that patients generally do not produce detectable levels of antibody during the first week (or longer) of an infection. A patient is said to have undergone seroconversion when antibody levels reach the threshold for detection. Typically, seroconversion coincides with the onset of signs and symptoms of disease. However, in an HIV infection, for example, it generally takes 3 weeks for seroconversion to take place, and in some instances, it may take much longer.

Similar to techniques for the precipitin ring test and plaque assays, it is routine to prepare serial two-fold dilutions of the patient's serum and determine the titer of agglutinating antibody present. Since antibody levels change over time in both primary and secondary immune responses, by checking samples over time, changes in antibody titer can be detected. For example, a comparison of the titer during the acute phase of an infection versus the titer from the convalescent phase will distinguish whether an infection is current or has occurred in the past. It is also possible to monitor how well the patient's immune system is responding to the pathogen.

Blood Typing and Cross-Matching

In addition to antibodies against bacteria and viruses to which they have previously been exposed, most individuals also carry antibodies against blood types other than their own. There are presently 33 immunologically important blood-type systems, many of which are restricted within various ethnic groups or rarely result in the production of antibodies. The most important and perhaps best known are the ABO and Rh blood groups (see Figure 19.1.3).

When units of blood are being considered for transfusion, pretransfusion blood testing must be performed. For the blood unit, commercially prepared antibodies against the A, B, and Rh antigens are mixed with red blood cells from the units to initially confirm that the blood type on the unit is accurate. Once a unit of blood has been requested for transfusion, it is vitally important to make sure the donor (unit of blood) and recipient (patient) are compatible for these crucial antigens. In addition to confirming the blood type of the unit, the patient's blood type is also confirmed using the same commercially prepared antibodies to A, B, and Rh. For example, as shown in Figure **26.3.7**, if the donor blood is A-positive, it will agglutinate with the anti-A antiserum and with the anti-Rh antiserum. If no agglutination is observed with any of the sera, then the blood type would be O-negative.

Following determination of the blood type, immediately prior to releasing the blood for transfusion, a **cross-match** is performed in which a small aliquot of the donor red blood cells are mixed with serum from the patient awaiting transfusion. If the patient does have antibodies against the donor red blood cells, hemagglutination will occur. To confirm any negative test results and check for sensitized red blood cells, Coombs' reagent may be added to the mix to facilitate visualization of the antibody-red blood cell interaction.

Under some circumstances, a minor cross-match may be performed as well. In this assay, a small aliquot of donor serum is mixed with patient red blood cells. This allows the detection of agglutinizing antibodies in the donor serum. This test is rarely necessary because transfusions generally use packed red blood cells with most of the plasma removed by centrifugation.

Red blood cells have many other antigens in addition to ABO and Rh. While most people are unlikely to have antibodies against these antigens, women who have had multiple pregnancies or patients who have had multiple transfusions may have them because of repeated exposure. For this reason, an **antibody screen** test is used to determine if such antibodies are present. Patient serum is checked against commercially prepared, pooled, type O red blood cells that express these antigens. If agglutination occurs, the antigen to which the patient is responding must be identified and determined not to be present in the donor unit.

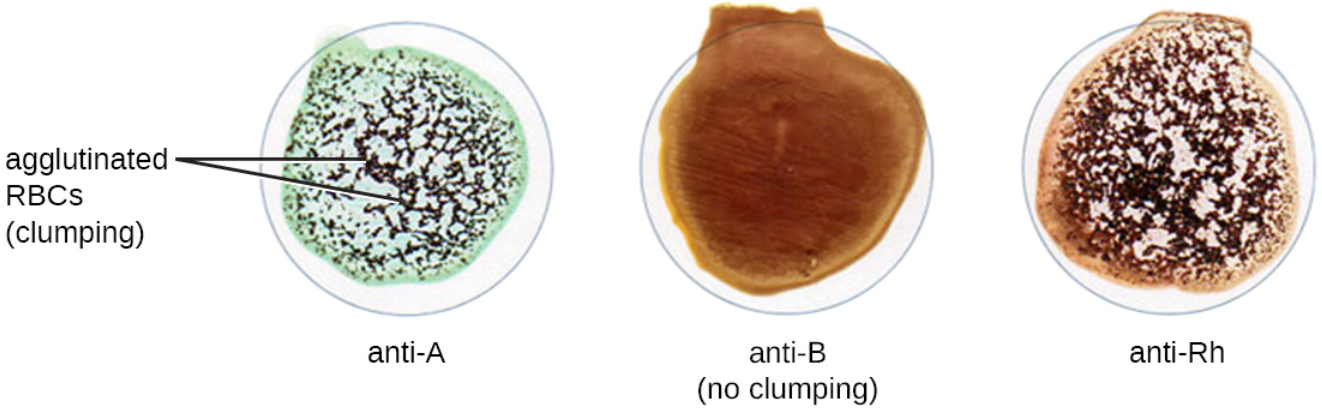


Figure **26.3.7**: This sample of a commercially produced "bedside" card enables quick typing of both a recipient's and donor's blood before transfusion. The card contains three reaction sites or wells. One is coated with an anti-A antibody, one with an anti-B antibody, and one with an anti-Rh antibody. Agglutination of red blood cells in a given site indicates a positive identification of the blood antigens: in this case, A and Rh antigens for blood type A-positive.

Table **26.3.1** summarizes the various kinds of agglutination assays discussed in this section.

Table **26.3.1**: Mechanisms of Select Antibody-Antigen Assays

Type of Assay	Mechanism	Example
Agglutination	Direct: Antibody is used to clump bacterial cells or other large structures	Serotyping bacteria
	Indirect: Latex beads are coupled with antigen or antibody to look for antibody or antigen, respectively, in patient serum	Confirming the presence of rheumatoid factor (IgM-binding Ig) in patient serum
Hemagglutination	Direct: Some bacteria and viruses cross-link red blood cells and clump them together	Diagnosing influenza, mumps, and measles
	Direct Coombs' test (DAT): Detects nonagglutinating antibodies or complement proteins on red blood cells *in vivo*	Checking for maternal antibodies binding to neonatal red blood cells
	Indirect Coombs' test (IAT): Screens an individual for antibodies against red blood cell antigens (other than the A and B antigens) that are unbound in a patient's serum *in vitro*	Performing pretransfusion blood testing
	Viral hemagglutination inhibition: Uses antibodies from a patient to inhibit viral agglutination	Diagnosing various viral diseases by the presence of patient antibodies against the virus
	Blood typing and cross-matching: Detects ABO, Rh, and minor antigens in the blood	Matches donor blood to recipient immune requirements

Key Concepts and Summary

- Antibodies can agglutinate cells or large particles into a visible matrix. **Agglutination** tests are often done on cards or in **microtiter plates** that allow multiple reactions to take place side by side using small volumes of reagents.
- Using antisera against certain proteins allows identification of **serovars** within species of bacteria.
- Detecting antibodies against a pathogen can be a powerful tool for diagnosing disease, but there is a period of time before patients go through **seroconversion** and the level of antibodies becomes detectable.
- Agglutination of latex beads in **indirect agglutination assays** can be used to detect the presence of specific antigens or specific antibodies in patient serum.
- **Neutralization assays** quantify the level of virus-specific antibody by measuring the decrease in hemagglutination observed after mixing patient serum with a standardized amount of virus.
- Hemagglutination assays are also used to screen and **cross-match** donor and recipient blood to ensure that the transfusion recipient does not have antibodies to antigens in the donated blood.

Footnotes

1. 1 Lancefield, Rebecca C., "The Antigenic Complex of *Streptococcus haemoliticus*. I. Demonstration of a Type-Specific Substance in Extracts of *Streptococcus haemolyticus*," *The Journal of Experimental Medicine* 47, no. 1 (1928): 91-103.

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26.4: Enzyme Immunoassays (EIA) and Enzyme-Linked Immunosorbent Assays (ELISA)

Learning Objectives

- Explain the differences and similarities between EIA, FEIA, and ELISA
- Describe the difference and similarities between immunohistochemistry and immunocytochemistry
- Describe the different purposes of direct and indirect ELISA

Similar to the western blot, **enzyme immunoassays (EIAs)** use antibodies to detect the presence of antigens. However, EIAs differ from western blots in that the assays are conducted in microtiter plates or *in vivo* rather than on an absorbent membrane. There are many different types of EIAs, but they all involve an antibody molecule whose constant region binds an enzyme, leaving the variable region free to bind its specific antigen. The addition of a substrate for the enzyme allows the antigen to be visualized or quantified (Figure **26.4.1**).

In EIAs, the substrate for the enzyme is most often a chromogen, a colorless molecule that is converted into a colored end product. The most widely used enzymes are **alkaline phosphatase** and **horseradish peroxidase** for which appropriate substrates are readily available. In some EIAs, the substrate is a **fluorogen**, a nonfluorescent molecule that the enzyme converts into a fluorescent form. EIAs that utilize a fluorogen are called **fluorescent enzyme immunoassays (FEIAs)**. Fluorescence can be detected by either a fluorescence microscope or a spectrophotometer.

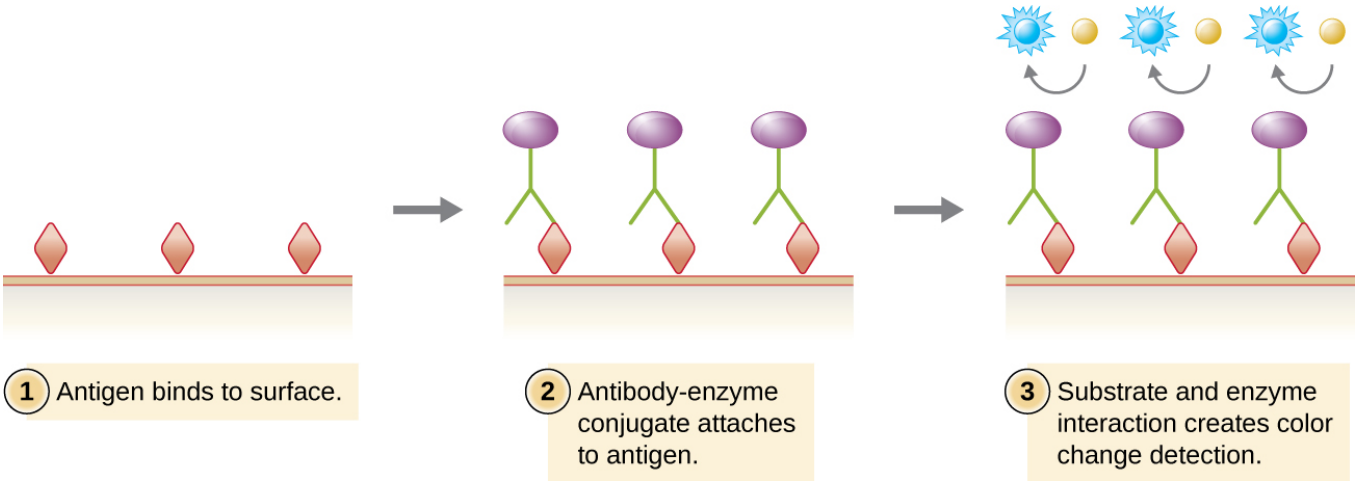


Figure **26.4.1**: Enzyme immunoassays, such as the direct ELISA shown here, use an enzyme-antibody conjugate to deliver a detectable substrate to the site of an antigen. The substrate may be a colorless molecule that is converted into a colored end product or an inactive fluorescent molecule that fluoresces after enzyme activation. (credit: modification of work by "Cavitri"/Wikimedia Commons)

The MMR Titer

The MMR vaccine is a combination vaccine that provides protection against measles, mumps, and rubella (German measles). Most people receive the MMR vaccine as children and thus have antibodies against these diseases. However, for various reasons, even vaccinated individuals may become susceptible to these diseases again later in life. For example, some children may receive only one round of the MMR vaccine instead of the recommended two. In addition, the titer of protective antibodies in an individual's body may begin to decline with age or as the result of some medical conditions.

To determine whether the titer of antibody in an individual's bloodstream is sufficient to provide protection, an MMR titer test can be performed. The test is a simple immunoassay that can be done quickly with a blood sample. The results of the test will indicate whether the individual still has immunity or needs another dose of the MMR vaccine.

Submitting to an MMR titer is often a pre-employment requirement for healthcare workers, especially those who will frequently be in contact with young children or immunocompromised patients. Were a healthcare worker to become infected

with measles, mumps, or rubella, the individual could easily pass these diseases on to susceptible patients, leading to an outbreak. Depending on the results of the MMR titer, healthcare workers might need to be revaccinated prior to beginning work.

Immunostaining

One powerful use of EIA is **immunostaining**, in which antibody-enzyme conjugates enhance microscopy. **Immunohistochemistry (IHC)** is used for examining whole tissues. As seen in Figure 26.4.2, a section of tissue can be stained to visualize the various cell types. In this example, a mAb against CD8 was used to stain CD8 cells in a section of tonsil tissue. It is now possible to count the number of CD8 cells, determine their relative numbers versus the other cell types present, and determine the location of these cells within this tissue. Such data would be useful for studying diseases such as AIDS, in which the normal function of CD8 cells is crucial for slowing disease progression.

Immunocytochemistry (ICC) is another valuable form of immunostaining. While similar to IHC, in ICC, extracellular matrix material is stripped away, and the cell membrane is etched with alcohol to make it permeable to antibodies. This allows antibodies to pass through the cell membrane and bind to specific targets inside the cell. Organelles, cytoskeletal components, and other intracellular structures can be visualized in this way. While some ICC techniques use EIA, the enzyme can be replaced with a fluorescent molecule, making it a fluorescent immunoassay.

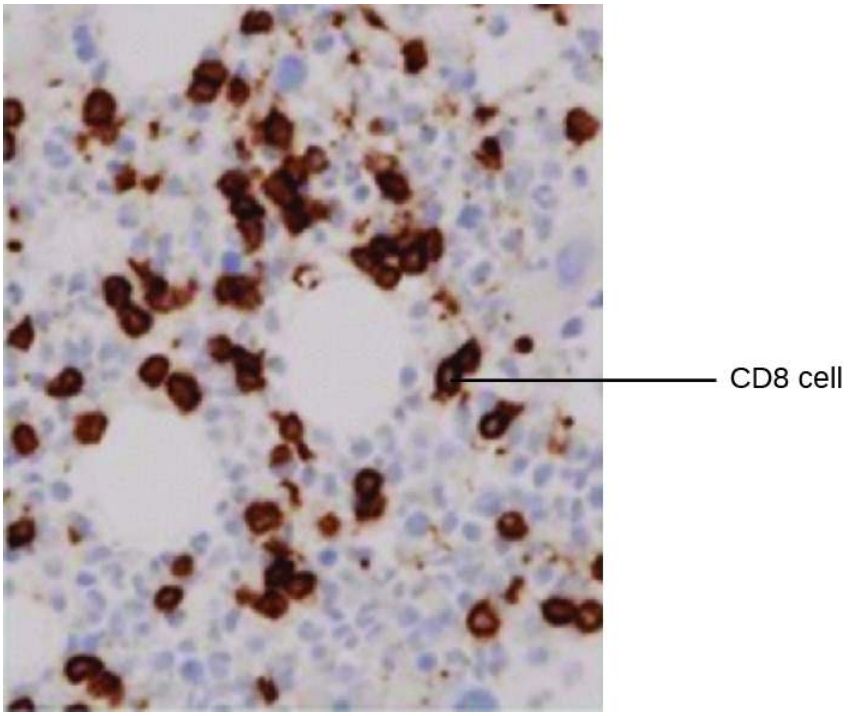


Figure 26.4.2: Enzyme-linked antibodies against CD8 were used to stain the CD8 cells in this preparation of bone marrow using a chromogen. (credit: modification of work by Yamashita M, Fujii Y, Ozaki K, Urano Y, Iwasa M, Nakamura S, Fujii S, Abe M, Sato Y, Yoshino T)

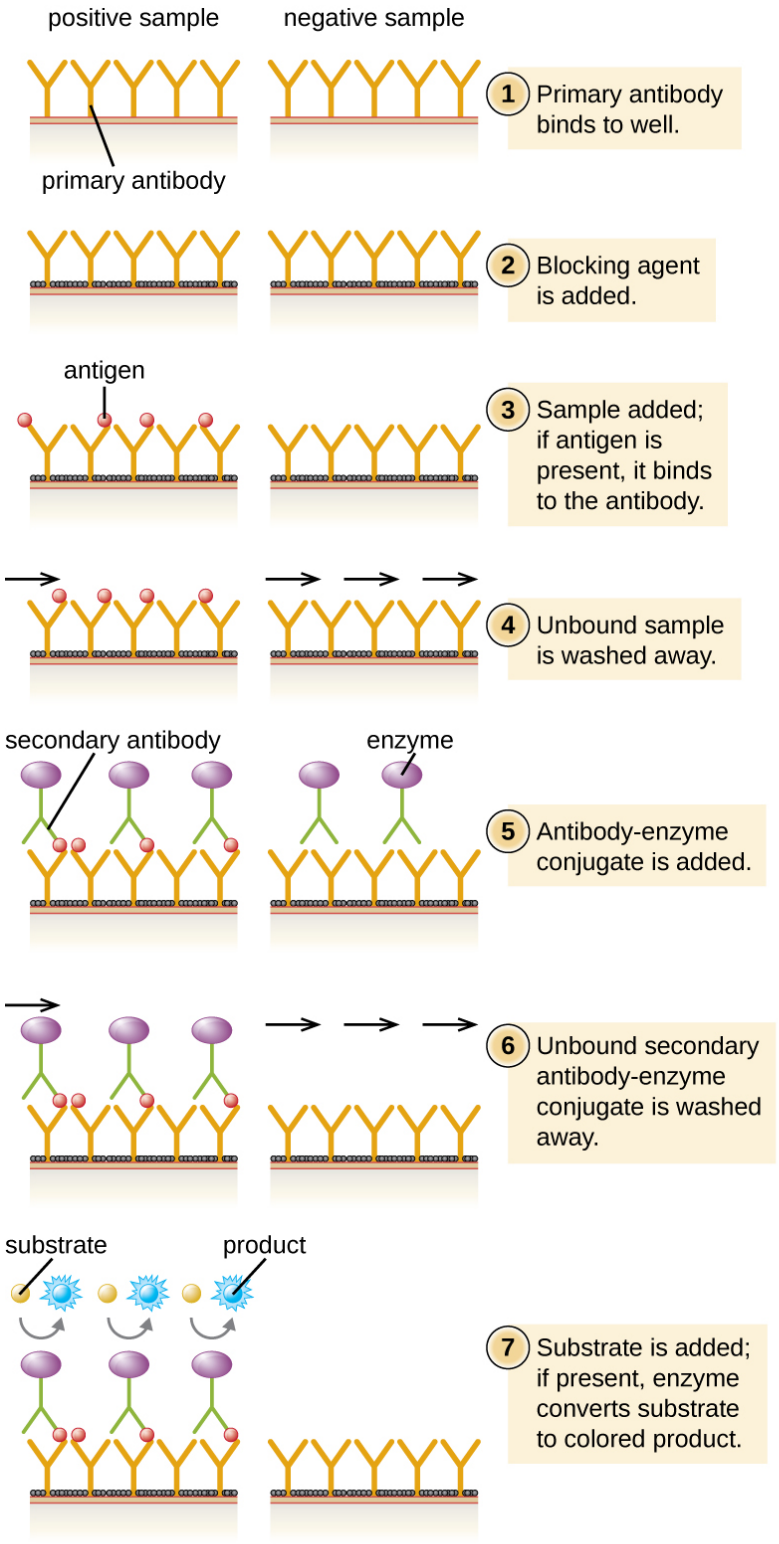
Enzyme-linked Immunosorbent Assays (ELISAs)

The **enzyme-linked immunosorbent assays (ELISAs)** are widely used EIAs. In the **direct ELISA**, antigens are immobilized in the well of a microtiter plate. An antibody that is specific for a particular antigen and is conjugated to an enzyme is added to each well. If the antigen is present, then the antibody will bind. After washing to remove any unbound antibodies, a colorless substrate (chromogen) is added. The presence of the enzyme converts the substrate into a colored end product (Figure 26.4.1). While this technique is faster because it only requires the use of one antibody, it has the disadvantage that the signal from a direct ELISA is lower (lower sensitivity).

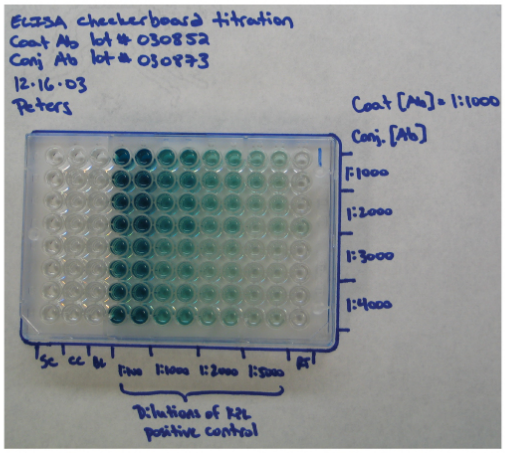
In a **sandwich ELISA**, the goal is to use antibodies to precisely quantify specific antigen present in a solution, such as antigen from a pathogen, a serum protein, or a hormone from the blood or urine to list just a few examples. The first step of a sandwich ELISA is to add the **primary antibody** to all the wells of a microtiter plate (Figure **26.4.3**). The antibody sticks to the plastic by hydrophobic interactions. After an appropriate incubation time, any unbound antibody is washed away. Comparable washes are used between each of the subsequent steps to ensure that only specifically bound molecules remain attached to the plate. A blocking protein is then added (e.g., albumin or the milk protein casein) to bind the remaining nonspecific protein-binding sites in the well. Some of the wells will receive known amounts of antigen to allow the construction of a standard curve, and unknown antigen solutions are added to the other wells. The primary antibody captures the antigen and, following a wash, the **secondary antibody** is added, which is a polyclonal antibody that is conjugated to an enzyme. After a final wash, a colorless substrate (chromogen) is added, and the enzyme converts it into a colored end product. The color intensity of the sample caused by the end product is measured with a spectrophotometer. The amount of color produced (measured as absorbance) is directly proportional to the amount of enzyme, which in turn is directly proportional to the captured antigen. ELISAs are extremely sensitive, allowing antigen to be quantified in the nanogram (10^{-9} g) per mL range.

In an **indirect ELISA**, we quantify antigen-specific antibody rather than antigen. We can use indirect ELISA to detect antibodies against many types of pathogens, including *Borrelia burgdorferi* (Lyme disease) and HIV. There are three important differences between indirect and direct ELISAs as shown in Figure **26.4.4**. Rather than using antibody to capture antigen, the indirect ELISA starts with attaching known antigen (e.g., peptides from HIV) to the bottom of the microtiter plate wells. After blocking the unbound sites on the plate, patient serum is added; if antibodies are present (primary antibody), they will bind the antigen. After washing away any unbound proteins, the secondary antibody with its conjugated enzyme is directed against the primary antibody (e.g., antihuman immunoglobulin). The secondary antibodyallows us to quantify how much antigen-specific antibody is present in the patient's serum by the intensity of the color produced from the conjugated enzyme-chromogen reaction.

As with several other tests for antibodies discussed in this chapter, there is always concern about cross-reactivity with antibodies directed against some other antigen, which can lead to false-positive results. Thus, we cannot definitively diagnose an HIV infection (or any other type of infection) based on a single indirect ELISA assay. We must confirm any suspected positive test, which is most often done using either an immunoblot that actually identifies the presence of specific peptides from the pathogen or a test to identify the nucleic acids associated with the pathogen, such as reverse transcriptase PCR (RT-PCR) or a nucleic acid antigen test.



(a)



(b)

Figure 26.4.3: (a) In a sandwich ELISA, a primary antibody is used to first capture an antigen with the primary antibody. A secondary antibody conjugated to an enzyme that also recognizes epitopes on the antigen is added. After the addition of the chromogen, a spectrophotometer measures the absorbance of end product, which is directly proportional to the amount of captured antigen. (b) An ELISA plate shows dilutions of antibodies (left) and antigens (bottom). Higher concentrations result in a darker final color. (credit b: modification of work by U.S. Fish and Wildlife Service Pacific Region)

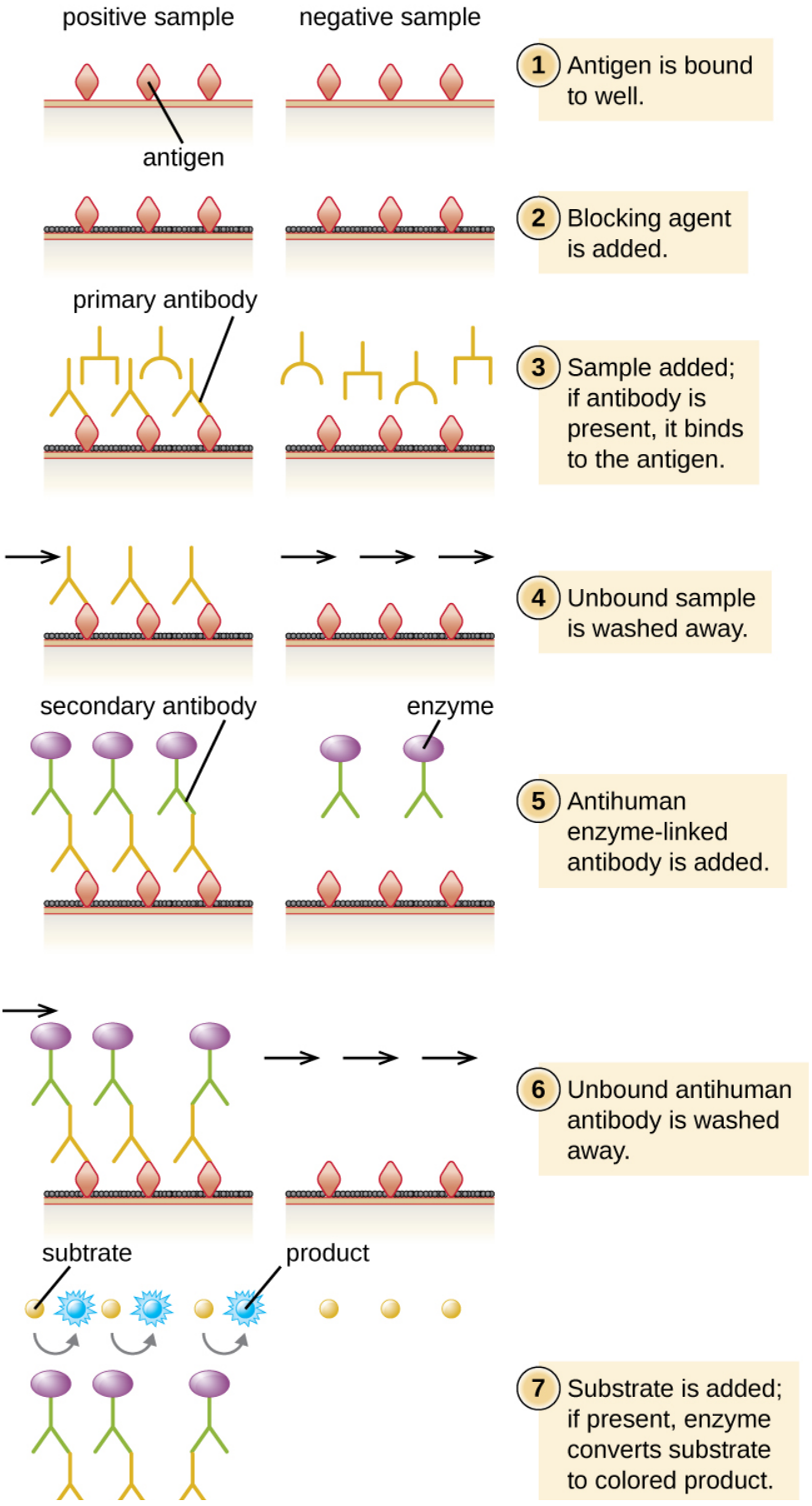

positive sample
negative sample
antigen
1 Antigen is bound to well.
2 Blocking agent is added.
primary antibody
3 Sample added; if antibody is present, it binds to the antigen.
4 Unbound sample is washed away.
secondary antibody
enzyme
5 Antihuman enzyme-linked antibody is added.
6 Unbound antihuman antibody is washed away.
subtrate
product
7 Substrate is added; if present, enzyme converts substrate to colored product.

Figure 26.4.4: The indirect ELISA is used to quantify antigen-specific antibodies in patient serum for disease diagnosis. Antigen from the suspected disease agent is attached to microtiter plates. The primary antibody comes from the patient's serum, which is subsequently bound by the enzyme-conjugated secondary antibody. Measuring the production of end product allows us to detect or quantify the amount of antigen-specific antibody present in the patient's serum.

Immunofiltration and Immunochromatographic Assays

For some situations, it may be necessary to detect or quantify antigens or antibodies that are present at very low concentration in solution. Immunofiltration techniques have been developed to make this possible. In **immunofiltration**, a large volume of fluid is passed through a porous membrane into an absorbent pad. An antigen attached to the porous membrane will capture antibody as it passes; alternatively, we can also attach an antibody to the membrane to capture antigen.

The method of immunofiltration has been adapted in the development of **immunochromatographic assays**, commonly known as **lateral flow tests** or strip tests. These tests are quick and easy to perform, making them popular for point-of-care use (i.e., in the doctor's office) or in-home use. One example is the TORCH test that allows doctors to screen pregnant women or newborns for infection by an array of viruses and other pathogens (*Toxoplasma*, other viruses, rubella, cytomegalovirus, herpes simplex). In-home pregnancy tests are another widely used example of a lateral flow test (Figure 26.4.5). Immunofiltration tests are also popular in developing countries, because they are inexpensive and do not require constant refrigeration of the dried reagents. However, the technology is also built into some sophisticated laboratory equipment.

In lateral flow tests (Figure 26.4.6), fluids such as urine are applied to an absorbent pad on the test strip. The fluid flows by capillary action and moves through a stripe of beads with antibodies attached to their surfaces. The fluid in the sample actually hydrates the reagents, which are present in a dried state in the stripe. Antibody-coated beads made of latex or tiny gold particles will bind antigens in the test fluid. The antibody-antigen complexes then flow over a second stripe that has immobilized antibody against the antigen; this stripe will retain the beads that have bound antigen. A third control stripe binds any beads. A red color (from gold particles) or blue (from latex beads) developing at the test line indicates a positive test. If the color only develops at the control line, the test is negative.

Like ELISA techniques, lateral flow tests take advantage of antibody sandwiches, providing sensitivity and specificity. While not as quantitative as ELISA, these tests have the advantage of being fast, inexpensive, and not dependent on special equipment. Thus, they can be performed anywhere by anyone. There are some concerns about putting such powerful diagnostic tests into the hands of people who may not understand the tests' limitations, such as the possibility of false-positive results. While home pregnancy tests have become widely accepted, at-home antibody-detection tests for diseases like HIV have raised some concerns in the medical community. Some have questioned whether self-administration of such tests should be allowed in the absence of medical personnel who can explain the test results and order appropriate confirmatory tests. However, with growing numbers of lateral flow tests becoming available, and the rapid development of lab-on-a-chip technology (Figure 20.1), home medical tests are likely to become even more commonplace in the future.

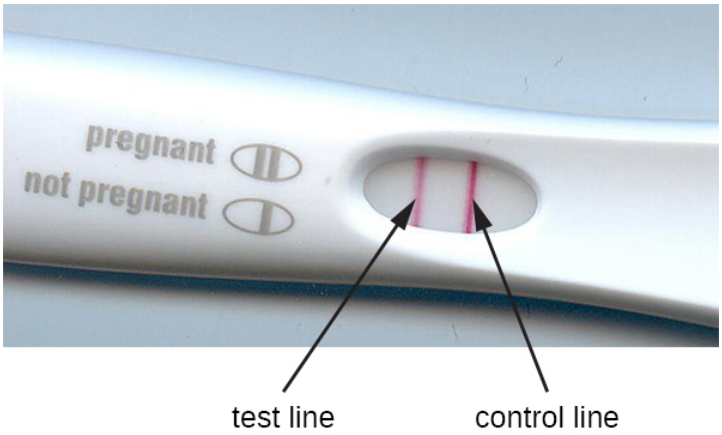


Figure 26.4.5: A lateral flow test detecting pregnancy-related hormones in urine. The control stripe verifies the validity of the test and the test line determines the presence of pregnancy-related hormones in the urine. (credit: modification of work by Klaus Hoffmeier)

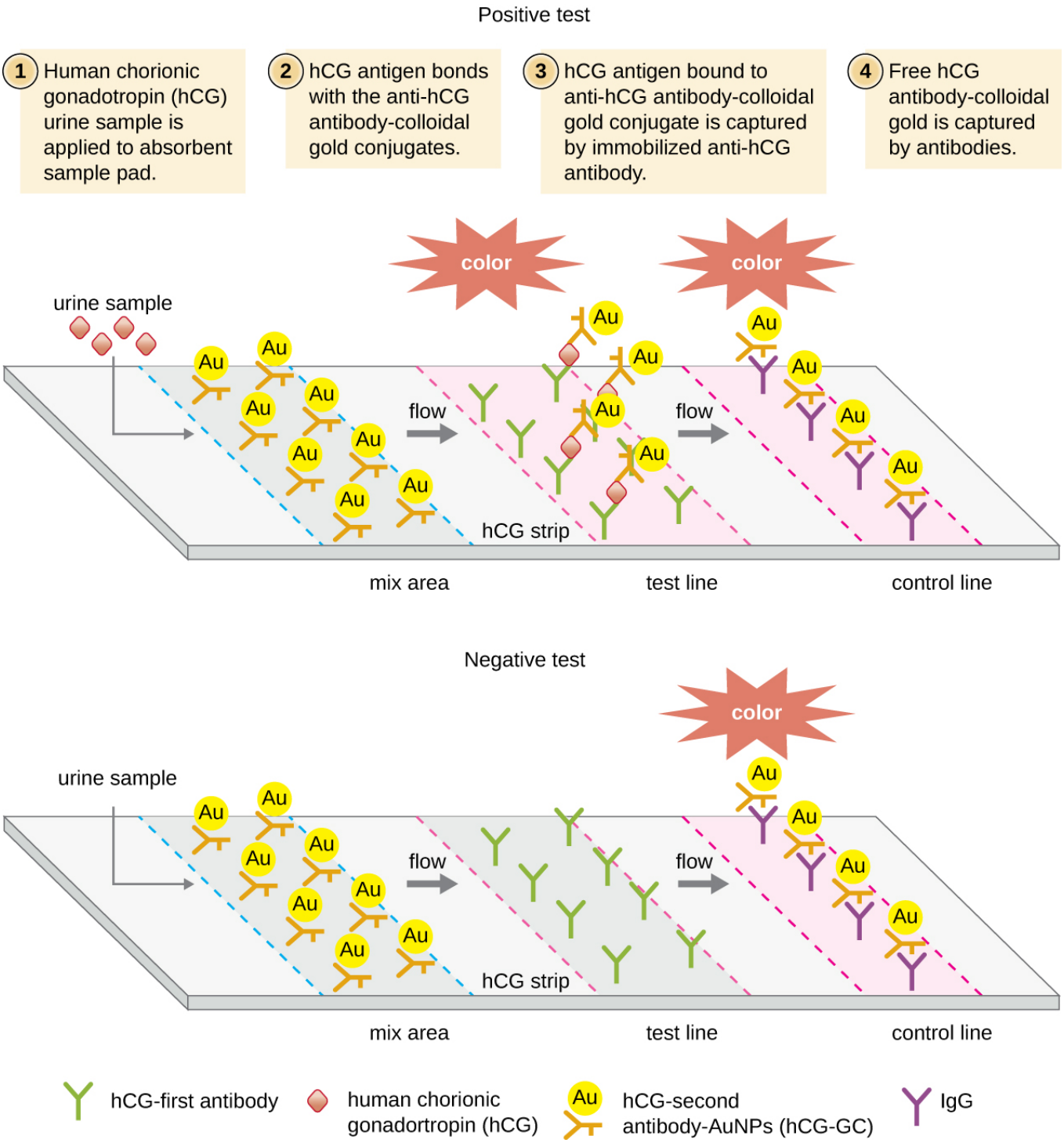


Figure **26.4.6**: Immunochromatographic assays, or lateral flow tests, allow the testing of antigen in a dilute solution. As the fluid flows through the test strip, it rehydrates the reagents. Antibodies conjugated to small particles bind the antigen in the first stripe and then flow onto the second stripe where they are bound by a second, fixed antibody. This produces a line of color, depending on the color of the beads. The third, control stripe binds beads as well to indicate that the test is working properly. (credit: modification of work by Yeh CH, Zhao ZQ, Shen PL, Lin YC)

Table **26.4.1** compares some of the key mechanisms and examples of some of the EIAs discussed in this section as well as immunoblots, which were discussed in Detecting Antigen-Antibody Complexes.

Table **26.4.1**: Immunoblots & Enzyme Immunoassays

Type of Assay	Mechanism	Specific Procedures	Examples
Immunoblots	Uses enzyme-antibody conjugates to identify specific proteins that have been transferred to an absorbent membrane	Western blot: Detects the presence of a particular protein	Detecting the presence of HIV peptides (or peptides from other infectious agents) in patient sera
Immunostaining	Uses enzyme-antibody conjugates to stain specific molecules on or in cells	Immunohistochemistry: Used to stain specific cells in a tissue	Stain for presence of CD8 cells in host tissue
Enzyme-linked immunosorbent assay (ELISA)	Uses enzyme-antibody conjugates to quantify target molecules	Direct ELISA: Uses a single antibody to detect the presence of an antigen	Detection of HIV antigen p24 up to one month after being infected
		Indirect ELISA: Measures the amount of antibody produced against an antigen	Detection of HIV antibodies in serum
Immunochromatographic (lateral flow) assays	Techniques use the capture of flowing, color-labeled antigen-antibody complexes by fixed antibody for disease diagnosis	Sandwich ELISA: Measures the amount of antigen bound by the antibody	Detection of antibodies for various pathogens in patient sera (e.g., rapid strep, malaria dipstick)
			Pregnancy test detecting human chorionic gonadotrophin in urine

Key Concepts and Summary

- **Enzyme immunoassays (EIA)** are used to visualize and quantify antigens. They use an antibody conjugated to an enzyme to bind the antigen, and the enzyme converts a substrate into an observable end product. The substrate may be either a chromogen or a fluorogen.
- **Immunostaining** is an EIA technique for visualizing cells in a tissue (**immunohistochemistry**) or examining intracellular structures (**immunocytochemistry**).
- **Direct ELISA** is used to quantify an antigen in solution. The primary antibody captures the antigen, and the secondary antibody delivers an enzyme. Production of end product from the chromogenic substrate is directly proportional to the amount of captured antigen.
- **Indirect ELISA** is used to detect antibodies in patient serum by attaching antigen to the well of a microtiter plate, allowing the patient (primary) antibody to bind the antigen and an enzyme-conjugated secondary antibody to detect the primary antibody.
- **Immunofiltration and immunochromatographic assays** are used in **lateral flow tests**, which can be used to diagnose pregnancy and various diseases by detecting color-labeled antigen-antibody complexes in urine or other fluid samples

Footnotes

1. 1 Thomas, Justin G., Victor Jaffe, Judith Shaffer, and Jose Abreu, “HIV Testing: US Recommendations 2014,” *Osteopathic Family Physician* 6, no. 6 (2014).

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26.5: Fluorescent Auto-Antibody Techniques

Learning Objectives

- Describe the benefits of immunofluorescent antibody assays in comparison to nonfluorescent assays
- Compare direct and indirect fluorescent antibody assays
- Explain how a flow cytometer can be used to quantify specific subsets of cells present in a complex mixture of cell types
- Explain how a fluorescence-activated cell sorter can be used to separate unique types of cells

Rapid visualization of bacteria from a clinical sample such as a throat swab or sputum can be achieved through **fluorescent antibody (FA) techniques** that attach a fluorescent marker (fluorogen) to the constant region of an antibody, resulting in a reporter molecule that is quick to use, easy to see or measure, and able to bind to target markers with high specificity. We can also label cells, allowing us to precisely quantify particular subsets of cells or even purify these subsets for further research.

As with the enzyme assays, FA methods may be direct, in which a labeled mAb binds an antigen, or indirect, in which secondary polyclonal antibodies bind patient antibodies that react to a prepared antigen. Applications of these two methods were demonstrated in Figure 2.3.8. FA methods are also used in automated cell counting and sorting systems to enumerate or segregate labeled subpopulations of cells in a sample.

Direct Fluorescent Antibody Techniques

Direct fluorescent antibody (DFA) tests use a fluorescently labeled mAb to bind and illuminate a target antigen. DFA tests are particularly useful for the rapid diagnosis of bacterial diseases. For example, fluorescence-labeled antibodies against *Streptococcus pyogenes* (group A strep) can be used to obtain a diagnosis of strep throat from a throat swab. The diagnosis is ready in a matter of minutes, and the patient can be started on antibiotics before even leaving the clinic. DFA techniques may also be used to diagnose pneumonia caused by *Mycoplasma pneumoniae* or *Legionella pneumophila* from sputum samples (Figure 26.5.1). The fluorescent antibodies bind to the bacteria on a microscope slide, allowing ready detection of the bacteria using a fluorescence microscope. Thus, the DFA technique is valuable for visualizing certain bacteria that are difficult to isolate or culture from patient samples.

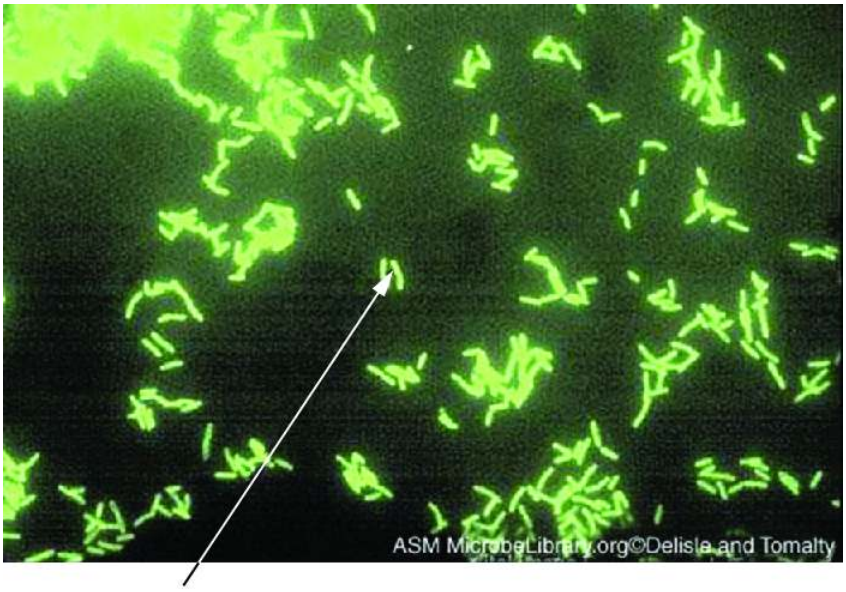


Figure 26.5.1: A green fluorescent mAb against *L. pneumophila* is used here to visualize and identify bacteria from a smear of a sample from the respiratory tract of a pneumonia patient. (credit: modification of work by American Society for Microbiology)

Indirect Fluorescent Antibody Techniques

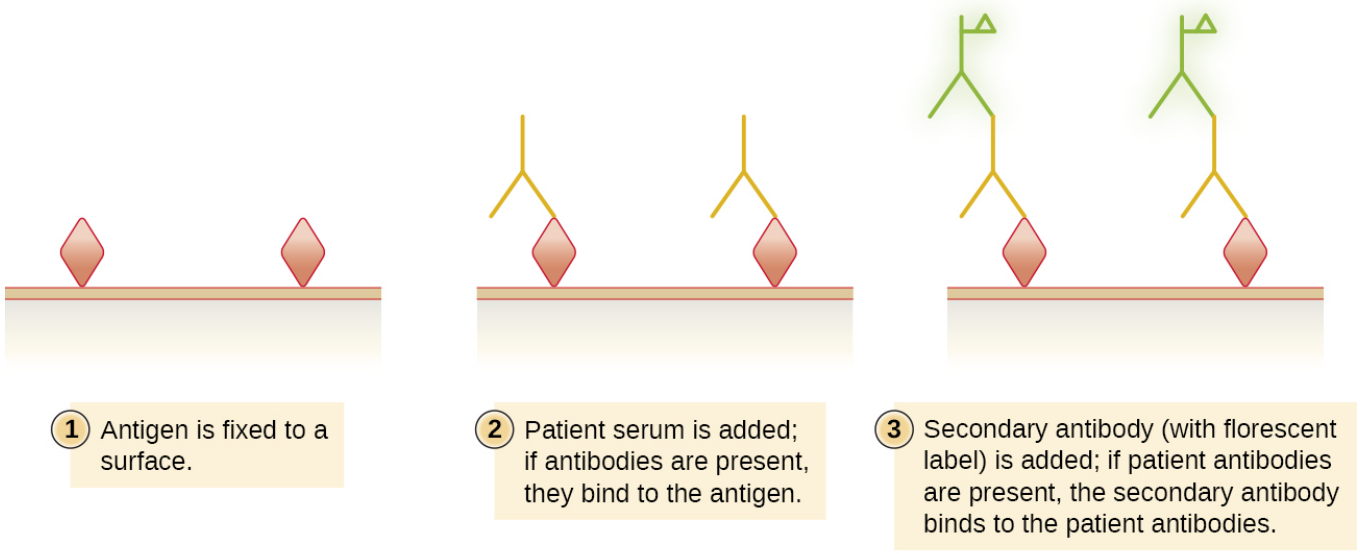
Indirect fluorescent antibody (IFA) tests (Figure 26.5.2) are used to look for antibodies in patient serum. For example, an IFA test for the diagnosis of syphilis uses *T. pallidum* cells isolated from a lab animal (the bacteria cannot be grown on lab media) and a

smear prepared on a glass slide. Patient serum is spread over the smear and anti-treponemal antibodies, if present, are allowed to bind. The serum is washed off and a secondary antibody added. The secondary antibody is an antihuman immunoglobulin conjugated to a fluorogen. On examination, the *T. pallidum* bacteria will only be visible if they have been bound by the antibodies from the patient's serum.

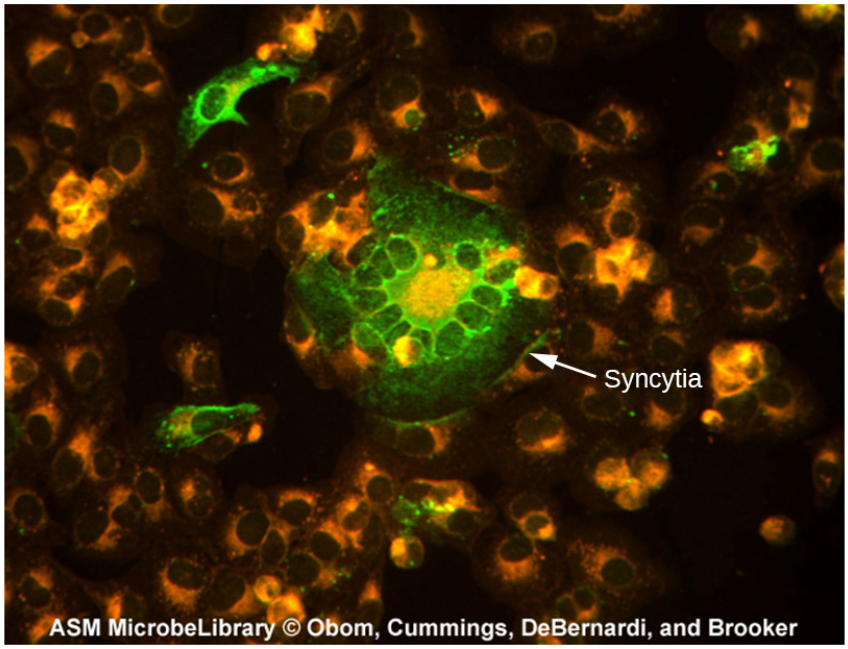
The IFA test for syphilis provides an important complement to the VDRL test discussed in Detecting Antigen-Antibody Complexes. The VDRL is more likely to generate false-positive reactions than the IFA test; however, the VDRL is a better test for determining whether an infection is currently active.

IFA tests are also useful for the diagnosis of autoimmune diseases. For example, systemic lupus erythematosus (SLE) (see Autoimmune Disorders) is characterized by elevated expression levels of antinuclear antibodies (ANA). These autoantibodies can be expressed against a variety of DNA-binding proteins and even against DNA itself. Because autoimmunity is often difficult to diagnose, especially early in disease progression, testing for ANA can be a valuable clue in making a diagnosis and starting appropriate treatment.

The IFA for ANA begins by fixing cells grown in culture to a glass slide and making them permeable to antibody. The slides are then incubated with serial dilutions of serum from the patient. After incubation, the slide is washed to remove unbound proteins, and the fluorescent antibody (antihuman IgG conjugated to a fluorogen) added. After an incubation and wash, the cells can be examined for fluorescence evident around the nucleus (Figure **26.5.3**). The titer of ANA in the serum is determined by the highest dilution showing fluorescence. Because many healthy people express ANA, the American College of Rheumatology recommends that the titer must be at least 1:40 in the presence of symptoms involving two or more organ systems to be considered indicative of SLE.[1]



(a)



(b)

Figure **26.5.2**: (a) The IFA test is used to detect antigen-specific antibodies by allowing them to bind to antigen fixed to a surface and then illuminating these complexes with a secondary antibody-fluorogen conjugate. (b) In this example of a micrograph of an indirect fluorescent antibody test, a patient's antibodies to the measles virus bind to viral antigens present on inactivated measles-infected cells affixed to a slide. Secondary antibodies bind the patient's antibodies and carry a fluorescent molecule. (credit b: modification of work by American Society for Microbiology)

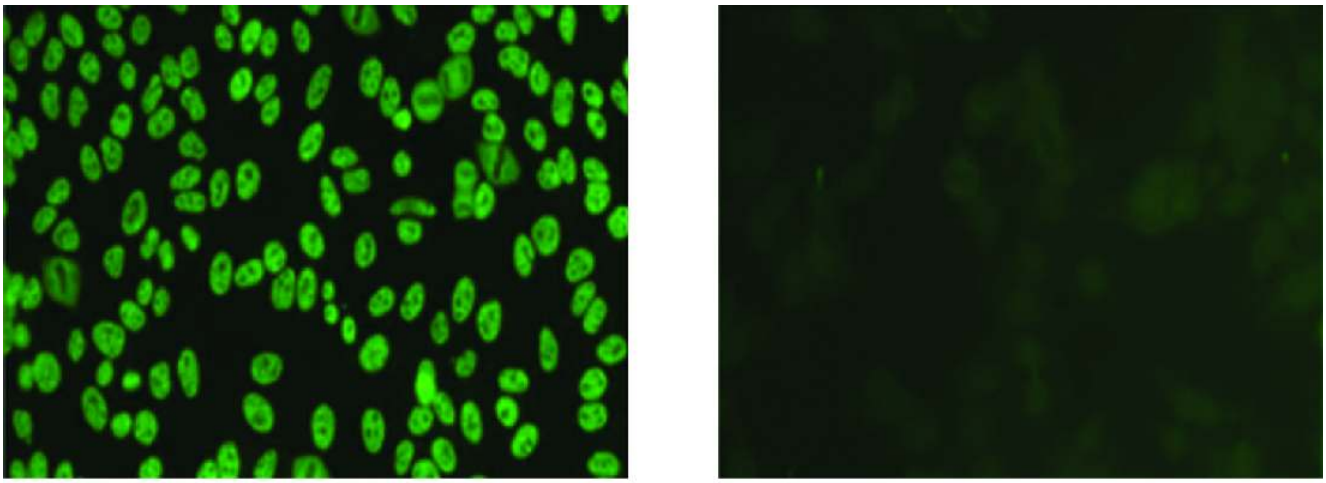


Figure **26.5.3**: In this test for antinuclear antibodies (ANA), cells are exposed to serum from a patient suspected of making ANA and then to a fluorescent mAb specific for human immunoglobulin. As a control, serum from a healthy patient is also used. Visible fluorescence around the nucleus demonstrates the presence of ANA in the patient's serum. In the healthy control where lower levels of ANA are produced, very faint green is detected. (credit left, right: modification of work by Al-Hussaini AA, Alzahrani MD, Alenizi AS, Suliman NM, Khan MA, Alharbi SA, Chentoufi AA)

Flow Cytometry

Fluorescently labeled antibodies can be used to quantify cells of a specific type in a complex mixture using **flow cytometry** (Figure **26.5.4**), an automated, cell-counting system that detects fluorescing cells as they pass through a narrow tube one cell at a time. For example, in HIV infections, it is important to know the level of CD4 T cells in the patient's blood; if the numbers fall below 500 per µL of blood, the patient becomes more likely to acquire opportunistic infections; below 200 per µL, the patient can no longer mount a useful adaptive immune response at all. The analysis begins by incubating a mixed-cell population (e.g., white blood cells from a donor) with a fluorescently labeled mAb specific for a subpopulation of cells (e.g., anti-CD4). Some experiments look at two cell markers simultaneously by adding a different fluorogen to the appropriate mAb. The cells are then introduced to the flow cytometer through a narrow capillary that forces the cells to pass in single file. A laser is used to activate the fluorogen. The fluorescent light radiates out in all directions, so the fluorescence detector can be positioned at an angle from the incident laser light.

Figure **26.5.4** shows the obscuration bar in front of the forward-scatter detector that prevents laser light from hitting the detector. As a cell passes through the laser bar, the forward-scatter detector detects light scattered around the obscuration bar. The scattered light is transformed into a voltage pulse, and the cytometer counts a cell. The fluorescence from a labeled cell is detected by the side-scatter detectors. The light passes through various dichroic mirrors such that the light emitted from the fluorophore is received by the correct detector.

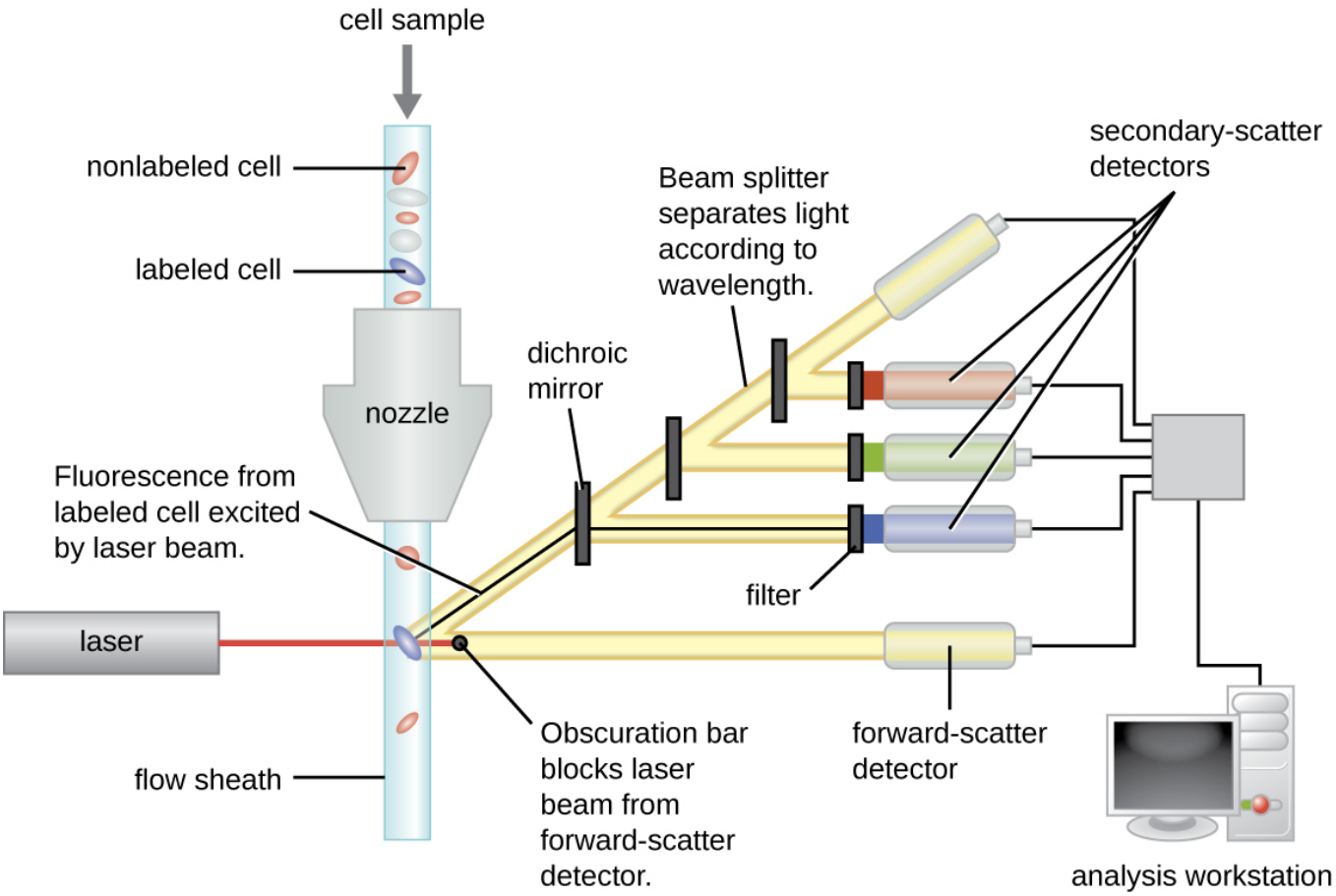


Figure 26.5.4: In flow cytometry, a mixture of fluorescently labeled and unlabeled cells passes through a narrow capillary. A laser excites the fluorogen, and the fluorescence intensity of each cell is measured by a detector. (credit: modification of work by "Kierano"/Wikimedia Commons)

Data are collected from both the forward- and side-scatter detectors. One way these data can be presented is in the form of a histogram. The forward scatter is placed on the *y*-axis (to represent the number of cells), and the side scatter is placed on the *x*-axis (to represent the fluoresence of each cell). The scaling for the *x*-axis is logarithmic, so fluorescence intensity increases by a factor of 10 with each unit increase along the axis. Figure **26.5.5** depicts an example in which a culture of cells is combined with an antibody attached to a fluorophore to detect CD8 cells and then analyzed by flow cytometry. The histogram has two peaks. The peak on the left has lower fluorescence readings, representing the subset of the cell population (approximately 30 cells) that does not fluoresce; hence, they are not bound by antibody and therefore do not express CD8. The peak on the right has higher fluorescence readings, representing the subset of the cell population (approximately 100 cells) that show fluorescence; hence, they are bound by the antibody and therefore do express CD8.

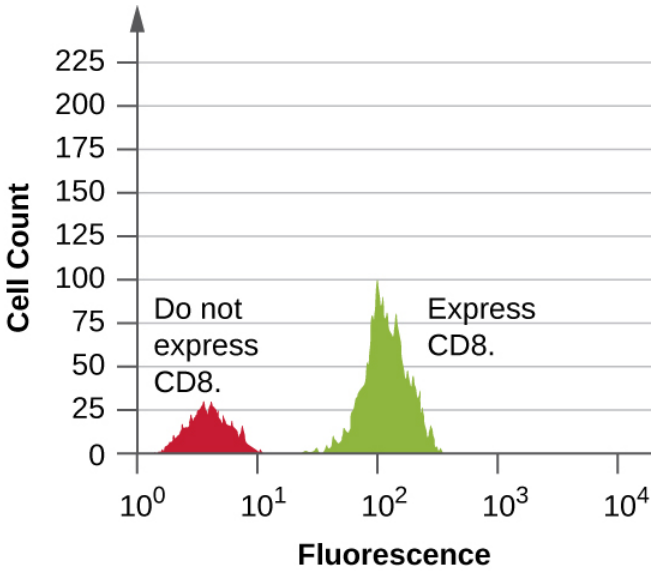


Figure **26.5.5**: Flow cytometry data are often compiled as a histogram. In the histogram, the area under each peak is proportional to the number of cells in each population. The *x*-axis is the relative fluorescence expressed by the cells (on a log scale), and the *y*-axis represents the number of cells at a particular level of fluorescence.

Cell Sorting Using Immunofluorescence

The flow cytometer and immunofluorescence can also be modified to sort cells from a single sample into purified subpopulations of cells for research purposes. This modification of the flow cytometer is called a **fluorescence-activated cell sorter (FACS)**. In a FACS, fluorescence by a cell induces the device to put a charge on a droplet of the transporting fluid containing that cell. The charge is specific to the wavelength of the fluorescent light, which allows for differential sorting by those different charges. The sorting is accomplished by an electrostatic deflector that moves the charged droplet containing the cell into one collecting vessel or another. The process results in highly purified subpopulations of cells.

One limitation of a FACS is that it only works on isolated cells. Thus, the method would work in sorting white blood cells, since they exist as isolated cells. But for cells in a tissue, flow cytometry can only be applied if we can excise the tissue and separate it into single cells (using proteases to cleave cell-cell adhesion molecules) without disrupting cell integrity. This method may be used on tumors, but more often, immunohistochemistry and immunocytochemistry are used to study cells in tissues.

Link to Learning

Watch videos to learn more about how flow cytometry and a FACS work.

Table **26.5.1** compares the mechanisms of the fluorescent antibody techniques discussed in this section.

Table **26.5.1**: Fluorescent Antibody Techniques

Type of Assay	Mechanism	Examples
Direct fluorescent antibody (DFA)	Uses fluorogen-antibody conjugates to label bacteria from patient samples	Visualizing *Legionella pneumophila* from a throat swab
Indirect fluorescent antibody (IFA)	Detects disease-specific antibodies in patent serum	Diagnosing syphilis; detecting antinuclear antibodies (ANA) for lupus and other autoimmune diseases
Flow cytometry	Labels cell membranes with fluorogen-antibody conjugate markers excited by a laser; machine counts the cell and records the relative fluorescence	Counting the number of fluorescently labeled CD4 or CD8 cells in a sample
Fluorescence activated cell sorter (FACS)	Form of flow cytometry that both counts cells and physically separates them into pools of high and low fluorescence cells	Sorting cancer cells

Key Concepts and Summary

- **Immunofluorescence** assays use antibody-fluorogen conjugates to illuminate antigens for easy, rapid detection.
- **Direct immunofluorescence** can be used to detect the presence of bacteria in clinical samples such as sputum.
- **Indirect immunofluorescence** detects the presence of antigen-specific antibodies in patient sera. The fluorescent antibody binds to the antigen-specific antibody rather than the antigen.
- The use of indirect immunofluorescence assays to detect **antinuclear antibodies** is an important tool in the diagnosis of several autoimmune diseases.
- **Flow cytometry** uses fluorescent mAbs against cell-membrane proteins to quantify specific subsets of cells in complex mixtures.
- **Fluorescence-activated cell sorters** are an extension of flow cytometry in which fluorescence intensity is used to physically separate cells into high and low fluorescence populations.

Footnotes

1. 1 Gill, James M., ANNA M. Quisel, PETER V. Rocca, and DENE T. Walters. “Diagnosis of systemic lupus erythematosus.” *American family physician* 68, no. 11 (2003): 2179-2186.

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26.E: Laboratory Analysis of the Immune Response (Exercises)

26.1: Practical Applications of Monoclonal and Polyclonal Antibodies

In addition to being crucial for our normal immune response, antibodies provide powerful tools for research and diagnostic purposes. The high specificity of antibodies makes them an excellent tool for detecting and quantifying a broad array of targets, from drugs to serum proteins to microorganisms. With in vitro assays, antibodies can be used to precipitate soluble antigens, agglutinate cells, and neutralize drugs, toxins, and viruses.

Multiple Choice

For many uses in the laboratory, polyclonal antibodies work well, but for some types of assays, they lack sufficient _____ because they cross-react with inappropriate antigens.

A. specificity
B. sensitivity
C. accuracy
D. reactivity

Answer

A

How are monoclonal antibodies produced?

A. Antibody-producing B cells from a mouse are fused with myeloma cells and then the cells are grown in tissue culture.
B. A mouse is injected with an antigen and then antibodies are harvested from its serum.
C. They are produced by the human immune system as a natural response to an infection.
D. They are produced by a mouse's immune system as a natural response to an infection.

Answer

A

Fill in the Blank

When we inject an animal with the same antigen a second time a few weeks after the first, _____ takes place, which means the antibodies produced after the second injection will on average bind the antigen more tightly.

Answer

affinity maturation

When using mAbs to treat disease in humans, the mAbs must first be _____ by replacing the mouse constant region DNA with human constant region DNA.

Answer

humanized

If we used normal mouse mAbs to treat human disease, multiple doses would cause the patient to respond with _____ against the mouse antibodies.

Answer

neutralizing antibodies

A polyclonal response to an infection occurs because most antigens have multiple _____,

Answer

epitopes

Short Answer

Describe two reasons why polyclonal antibodies are more likely to exhibit cross-reactivity than monoclonal antibodies.

Critical Thinking

Suppose you were screening produce in a grocery store for the presence of *E. coli* contamination. Would it be better to use a polyclonal anti-*E. coli* antiserum or a mAb against an *E. coli* membrane protein? Explain.

26.2: Detecting Antigen-Antibody Complexes in vitro

Laboratory tests to detect antibodies and antigens outside of the body (e.g., in a test tube) are called *in vitro* assays. When both antibodies and their corresponding antigens are present in a solution, we can often observe a precipitation reaction in which large complexes (lattices) form and settle out of solution.

Multiple Choice

The formation of _____ is a positive result in the VDRL test.

A. flocculant
B. precipitin
C. coagulation
D. a bright pink color

Answer

A

The titer of a virus neutralization test is the highest dilution of patient serum

A. in which there is no detectable viral DNA.
B. in which there is no detectable viral protein.
C. that completely blocks plaque formation.
D. that reduces plaque formation by at least 50%.

Answer

D

In the Ouchterlony assay, we see a sharp precipitin arc form between antigen and antiserum. Why does this arc remain visible for a long time?

A. The antibody molecules are too large to diffuse through the agar.
B. The precipitin lattice is too large to diffuse through the agar.
C. Methanol, added once the arc forms, denatures the protein and blocks diffusion.
D. The antigen molecules are chemically coupled to the gel matrix.

Answer

B

Fill in the Blank

When slowly adding antigen to an antiserum, the amount of precipitin would gradually increase until reaching the _____; addition of more antigen after this point would actually decrease the amount of precipitin.

Answer

equivalence zone or zone of equivalence

The radial immunodiffusion test quantifies antigen by mixing _____ into a gel and then allowing antigen to diffuse out from a well cut in the gel.

Answer

antiserum

Short Answer

Explain why hemolysis in the complement fixation test is a negative test for infection.

What is meant by the term “neutralizing antibodies,” and how can we quantify this effect using the viral neutralization assay?

Critical Thinking

Both IgM and IgG antibodies can be used in precipitation reactions. However, one of these immunoglobulin classes will form precipitates at much lower concentrations than the other. Which class is this, and why is it so much more efficient in this regard?

26.3: Agglutination Assays

In addition to causing precipitation of soluble molecules and flocculation of molecules in suspension, antibodies can also clump together cells or particles (e.g., antigen-coated latex beads) in a process called agglutination. Agglutination can be used as an indicator of the presence of antibodies against bacteria or red blood cells. Agglutination assays are usually quick and easy to perform on a glass slide or microtiter plate.

Multiple Choice

We use antisera to distinguish between various _____ within a species of bacteria.

A. isotypes
B. serovars
C. subspecies
D. lines

Answer

B

When using antisera to characterize bacteria, we will often link the antibodies to _____ to better visualize the agglutination.

A. latex beads
B. red blood cells
C. other bacteria
D. white blood cells

Answer

A

The antibody screening test that is done along with pretransfusion blood typing is used to ensure that the recipient

A. does not have a previously undetected bacterial or viral infection.
B. is not immunocompromised.
C. actually does have the blood type stated in the online chart.
D. is not making antibodies against antigens outside the ABO or Rh systems.

Answer

D

The direct Coombs’ test is designed to detect when people have a disease that causes them to

A. have an excessively high fever.

B. quit making antibodies.
C. make too many red blood cells.
D. produce antibodies that bind to their own red blood cells.

Answer

D

Viral hemagglutination assays only work with certain types of viruses because

A. the virus must be able to cross-link red blood cells directly.
B. the virus must be able to lyse red blood cells.
C. the virus must not be able to lyse red blood cells.
D. other viruses are too dangerous to work with in a clinical lab setting.

Answer

A

Fill in the Blank

In the major cross-match, we mix _____ with the donor red blood cells and look for agglutination.

Answer

patient serum

Coombs' reagent is an antiserum with antibodies that bind to human _____.

Answer

immunoglobulins/antibodies and/or complement

Short Answer

Explain why the titer of a direct hemagglutination assay is the highest dilution that still causes hemagglutination, whereas in the viral hemagglutination inhibition assay, the titer is the highest dilution at which hemagglutination is not observed.

Why would a doctor order a direct Coombs' test when a baby is born with jaundice?

Critical Thinking

When shortages of donated blood occur, O-negative blood may be given to patients, even if they have a different blood type. Why is this the case? If O-negative blood supplies were depleted, what would be the next-best choice for a patient with a different blood type in critical need of a transfusion? Explain your answers.

26.4: Enzyme Immunoassays (EIA) and Enzyme-Linked Immunosorbent Assays (ELISA)

Enzyme immunoassays (EIA) are used to visualize and quantify antigens. They use an antibody conjugated to an enzyme to bind the antigen, and the enzyme converts a substrate into an observable end product. The substrate may be either a chromogen or a fluorogen. Immunostaining is an EIA technique for visualizing cells in a tissue (immunohistochemistry) or examining intracellular structures (immunocytochemistry). Direct ELISA is used to quantify an antigen in solution.

Multiple Choice

In an enzyme immunoassay, the enzyme

A. is bound by the antibody's antigen-binding site.
B. is attached to the well of a microtiter plate.
C. is conjugated to the suspect antigen.
D. is bound to the constant region of the secondary antibody.

Answer

D

When using an EIA to study microtubules or other structures inside a cell, we first chemically fix the cell and then treat the cells with alcohol. What is the purpose of this alcohol treatment?

A. It makes holes in the cell membrane large enough for antibodies to pass.
B. It makes the membrane sticky so antibodies will bind and be taken up by receptor-mediated endocytosis.
C. It removes negative charges from the membrane, which would otherwise repulse the antibodies.
D. It prevents nonspecific binding of the antibodies to the cell membrane.

Answer

A

In a lateral-flow pregnancy test, you see a blue band form on the control line and no band form on the test line. This is probably a _____ test for pregnancy.

A. positive
B. false-positive
C. false-negative
D. negative

Answer

D

When performing an FEIA, the fluorogen replaces the _____ that is used in an EIA.

A. antigen
B. chromogenic substrate
C. enzyme
D. secondary antibody

Answer

B

Fill in the Blank

To detect antibodies against bacteria in the bloodstream using an EIA, we would run a(n) _____, which we would start by attaching antigen from the bacteria to the wells of a microtiter plate.

Answer

indirect ELISA

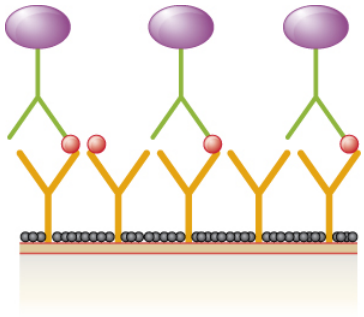
Short Answer

Why is it important in a sandwich ELISA that the antigen has multiple epitopes? And why might it be advantageous to use polyclonal antisera rather than mAb in this assay?

The pregnancy test strip detects the presence of human chorionic gonadotrophin in urine. This hormone is initially produced by the fetus and later by the placenta. Why is the test strip preferred for this test rather than using either a direct or indirect ELISA with their more quantifiable results?

Critical Thinking

Label the primary and secondary antibodies, and discuss why the production of end product will be proportional to the amount of antigen.

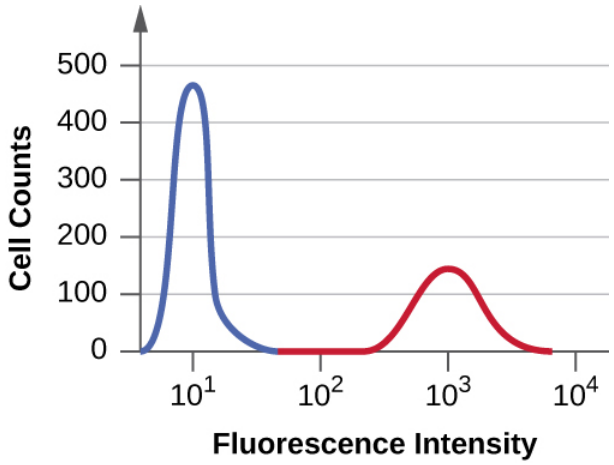


26.5: Fluorescent Auto-Antibody Techniques

Rapid visualization of bacteria from a clinical sample such as a throat swab or sputum can be achieved through fluorescent antibody (FA) techniques that attach a fluorescent marker (fluorogen) to the constant region of an antibody, resulting in a reporter molecule that is quick to use, easy to see or measure, and able to bind to target markers with high specificity. We can also label cells, allowing us to precisely quantify particular subsets of cells or even purify them for further research.

Multiple Choice

Suppose you need to quantify the level of CD8 T cells in the blood of a patient recovering from influenza. You treat a sample of the patient's white blood cells using a fluorescent mAb against CD8, pass the cells through a flow cytometer, and produce the histogram shown below. The area under the peak to the left (blue) is three times greater than the area of the peak on the right (red). What can you determine from these data?



A. There are no detectable CD8 cells.
B. There are three times as many CD4 cells than CD8 cells.
C. There are three times as many CD8 cells than CD4 cells.
D. CD8 cells make up about one-fourth of the total number of cells.

Answer

D

In the data described in the previous question, the average fluorescence intensity of cells in the second (red) peak is about _____ that in the first (blue) peak.

A. three times
B. 100 times
C. one-third
D. 1000 times

Answer

B

In a direct fluorescent antibody test, which of the following would we most likely be looking for using a fluorescently-labeled mAb?

A. bacteria in a patient sample
B. bacteria isolated from a patient and grown on agar plates
C. antiserum from a patient smeared onto a glass slide
D. antiserum from a patient that had bound to antigen-coated beads

Answer

A

Fill in the Blank

In flow cytometry, cell subsets are labeled using a fluorescent antibody to a membrane protein. The fluorogen is activated by a(n) _____ as the cells pass by the detectors.

Answer

laser

Fluorescence in a flow cytometer is measured by a detector set at an angle to the light source. There is also an in-line detector that can detect cell clumps or _____.

Answer

fragments

Critical Thinking

A patient suspected of having syphilis is tested using both the VDRL test and IFA. The IFA test comes back positive, but the VDRL test is negative. What is the most likely reason for these results?

A clinician suspects that a patient with pneumonia may be infected by *Legionella pneumophila*. Briefly describe two reasons why a DFA test might be better for detecting this pathogen than standard bacteriology techniques.

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