

Microbes in Perspective: Of Collectors, Classifiers, and Microscopists



Looking Ahead

Despite their incredibly small size, microbes occupy extensive, well-established, and integral places in the living world. Furthermore, their names, chemical makeups, and other characteristics conform to the principles that apply to all life forms, as we shall note in these pages.

On completing this chapter, you should be capable of:

- Explaining how microbes are named.
- Illustrating how organisms are cataloged in the tree of life.
- Distinguishing between the different forms of microscopy.
- Constructing and labeling a typical eukaryotic and prokaryotic microbial cell.

Carolus Linnaeus (FIGURE 2.1) was in trouble. The ship from Africa was pulling into port, and soon the Swedish botanist would be confronted with new plants, new animals, and new problems. "What shall we call this one?" he would be asked. "Or this one?" "Or that one?" The museums were full of newly discovered organisms (such as penguins, manatees, kangaroos, tobacco plants, bananas, and potatoes) arriving from distant corners of the globe and people were

The diversity of life is enormous, coming in many forms and variations. Here, you see part of the Spectrum of Life exhibit in the Hall of Biodiversity at the American Museum of Natural History in New York City. The exhibit displays more than 1,500 specimens, from microorganisms to terrestrial and aquatic plants and animals—but this only represents a small fraction of all known living organisms on this planet.

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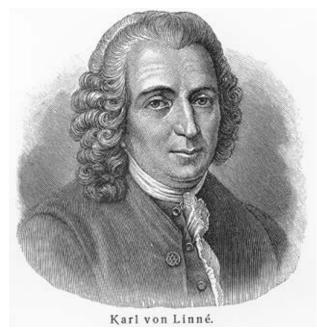


FIGURE 2.1 Carolus Linnaeus. Karl von Linne, the Swedish botanist known in scientific history as Carolus Linnaeus, took on the daunting task of classifying the known plants and animals of the biological world and giving them scientific names.

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bringing new specimens to him almost daily (he soon learned to dread the sight of arriving ships). Linnaeus even added to this frenzy: he inspired an unprecedented worldwide program of specimen hunting by sending his students around the globe in search of new and unknown plants and animals. This need to apply a scientific name to all organisms, called "nomenclature," had gotten thoroughly out of hand. To be sure, the biological world needed some order, but who had appointed Linnaeus king of nomenclature? It was exasperating, to say the least!

Linnaeus is just one of the collectors and classifiers we will encounter in this chapter as we fit microbes into the same scheme to which all living organisms belong and see how they are related to members of the visible world. Linnaeus could hardly anticipate what the future would hold.

2.1 Naming Microbes: What's in a Name?

Although at times it was exasperating, Linnaeus performed a valuable service. In a 1753 book on plants, he supplied scientific names for some 6,000 different plants known at that time. In his tenth edition of *Systema Naturae* (1759), Linnaeus extended the nomenclature scheme to thousands of animals. In the end, his scientific names were widely adopted by European scientists and were introduced around the world.

■ Escherichia coli esh-e'r-ē kē-ä kō lī (or kō lē)

Binomial and Common Names

Because all Linnaeus' assigned scientific names were derived from Latin, he had a monumental task of devising new Latin names and word endings for the thousands of new scientific names. So what is this naming system Linnaeus devised and which

is still used today?

Linnaeus' system is called binomial nomenclature; that is, each organism is assigned a two-word name, the binomial, derived from Latin. The first word is called the **genus** and it is followed by a second word, called the species modifier or the specific epithet. The binomial indicates the **species** of organism. For instance, a bacterial species normally found in the human colon is called *Escherichia coli* (FIGURE 2.2). The first part of the binomial, *Escherichia*, is the genus name to which the organism belongs. It is derived from the name of the scientist, Theodor Escherich, who first identified the microbe in 1888. The second word of the binomial, *coli*, is the specific epithet derived from the word "colon," which is where the bacterium was first found by Escherich. [The same rules apply to humans, who are scientifically known as *Homo sapiens* (*Homo* = "man"; *sapiens* = "wise")].

In Linnaeus' time and even today, when a new species is discovered the binomial name might reflect the discoverer's name, the organism's manner of growth, a location where the organism was first found, or even everyday names. A Closer Look 2.1 examines several other microbial species names and their origins.

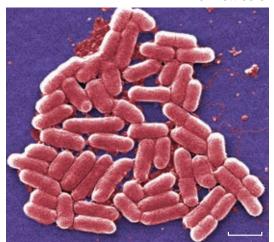


FIGURE 2.2 *Escherichia coli.* A false-color electron microscope image of *E. coli* cells. Most strains of the species are harmless and live in the intestines of healthy humans and animals. However, some strains such as this one (0157:H7) produce a powerful toxin that can cause severe illness. (Bar = $2 \mu m$.)

Courtesy of Janice Haney Carr/CDC

The correct way to write the species name for a microbe (or for any organism) is to capitalize the first letter of the genus name and to write the remainder of the genus name and the specific epithet in lowercase letters. The binomial should always be italicized (if this is not possible, it should be underlined). After the full species name has been introduced in a piece of writing, the name can be abbreviated by using the first letter of the genus name and the full specific epithet. Thus, *Escherichia coli* is abbreviated *E. coli*. Unfortunately, in today's newsprint and Internet sites, an organism's binomial often is written in normal text; for example, you might see Escherichia coli.

A microbial species may have various strains or subspecies whose identifiers are added to the binomial. An example is *Escherichia coli* O157:H7, a strain that does not occur normally in the human gut. The combination of letters and

A CLOSER LOOK 2.1

"What's in a Name?"

In Shakespeare's *Romeo* and *Juliet*, Juliet tells Romeo a name is an artificial and meaningless convention. Perhaps from her perspective in convincing Romeo that she is in love with the man called Montegue and not the family Montegue, there is a love-based reason for "What's in a name." As you read this book, you have and will come

across many scientific names for microbes. Not only are many of these names tongue twisting to pronounce (many are listed with their pronunciation in the text side margins and inside the front and back covers), but how in the world did the organisms get those names? Most are derived from Greek or Roman word roots. Here are a few examples.

Species	Meaning of Name			
Genera Named After Individuals	5			
Bordetella pertussis (bor-de-tel'lä pe ⁻ r-tus'sis)	Named after the Belgian Jules Bordet who in 1906 identified the small bacterium (ella means "small") responsible for pertussis (whooping cough).			
Neisseria gonorrhoeae (nī-se'rē-ä go-nôr-rē' ī)	Named after Albert Neisser who discovered the bacterial organism in 1879. As the specific epithet points out, the disease it causes is gonorrhea.			
Genera Named for a Microbe's Shape				
<i>Vibrio cholerae</i> (vib'rē-ō kol'e ⁻ r-ī)	Vibrio means "comma-shaped," which describes the shape of the bacterial cells causing cholera.			
Staphylococcus epidermidis (staf-i-lō-kok kus e-pi-der/mi-dis)	The stem <i>staphylo</i> means "cluster" and <i>coccus</i> means "spheres." So, these bacterial cells form clusters of spheres found on the skin surface (epidermis).			
Genera Named After an Attribut	e of the Microbe			
Saccharomyces cerevisiae (sak-ä-rō-mī'sēs se-ri-vis'ē-ī)	In 1837, Theodor Schwann observed yeast cells and called them <i>Saccharomyces</i> (<i>saccharo</i> = "sugar"; <i>myce</i> = "fungus") because the yeast converted grape juice (sugar) into alcohol; <i>cerevisiae</i> (<i>Ceres</i> was the Roman goddess of agriculture) refers to the use of yeast since ancient times to make beer.			
Myxococcus xanthus (micks-ō-kok'kus zan'thus)	The stem <i>myxo</i> means "slime," so these are slime-producing spheres that appear as a yellow (<i>xantho</i> = "yellow") growth in culture.			

Lastly, in keeping with Shakespeare's poetic style, there is the organism *Thiomargarita namibiensis*. This bacterial species was first isolated in 1997 from sediment samples in the Atlantic Ocean off the coast of Namibia, a country in southwestern Africa (*ensis* = "belonging to"). These spherical-shaped bacterial cells accumulate sulfur (*thio* = "sulfur") so when they are observed with the microscope the cells appear white and look like a microscopic string of pearls (*margarit* = "pearl"; see A Closer Look 2.3). Thus, we have *Thiomargarita namibiensis* (thī'ō-mär-gä-rē-tä na'mi-bē-en-sis)—the "Sulfur Pearl of Namibia." Juliet would be impressed!

- Streptococcus pneumoniae strep-tō-kok kus nü-mō nē-ī
- Neisseria meningitidis nī-se rē-ä me-nin ji'ti-dis
- meningitis: An inflammation of the membranes surrounding and protecting the brain and spinal cord, often resulting from a bacterial or viral infection.

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numbers for the strain refers to specific markers found on the cell's surface, which distinguish it from other types of *E. coli*. In this case, the O157:H7 strain produces a powerful toxin, which if ingested in contaminated food, can cause intestinal hemorrhaging.

Finally, besides the binomial name, many microbes have a common name often used in conversation. For instance, one of the bacterial species causing bacterial pneumonia, *Streptococcus pneumoniae*, is commonly referred to as the pneumococcus; and a bacterial species causing bacterial meningitis, *Neisseria meningitidis*, is often called the meningococcus.

2.2 Taxonomy: Cataloging Life

In addition to providing the names to many organisms, Linnaeus also established the ground rules of **taxonomy**, which involves the classification of organisms into hierarchical groups that indicate natural relationships. Thus, the taxonomic rules bring order to the living world by placing all organisms into related categories that can be more easily studied and related to other microscopic and macroscopic forms of life.

The Hierarchical Groups

As outlined in TABLE 2.1, the least inclusive, most fundamental group is the species (pl. species). For most animals and plants, a species is usually defined as a group of organisms that interbreed sexually with one another in nature, produce offspring similar to the parents, and are fertile. This definition also holds true for the eukary-otic microbes, most of which also reproduce sexually. However, problems exist in classifying prokaryotic organisms because these organisms do not interbreed sexually. Therefore, one cannot define a prokaryotic species based on sexual reproductive patterns and a valid definition remains controversial. For our purposes, a group of bacteria belong to the same species if the members share many stable physiological, biochemical, and genetic properties that are absent from other prokaryotic groups.

In this Linnaean classification, a group of species closely related are gathered together to form a genus (pl. genera). For example, familiar to us are lions, tigers, and leopards. They all belong to separate species, but they are classified together in the same genus

2.1	Taxonomic Classification of Humans, Brewer's Yeast, and a Common Bacterium				
	Humans	Brewer's Yeast	Escherichia coli		
Domain	Eukarya	Eukarya	Bacteria		
Kingdom	Animalia	Fungi			
Phylum	Chordata	Ascomycota	Proteobacteria		
Class	Mammalia	Saccharomycotina	Gammaproteobacteria		
Order	Primates	Saccharomycetales	Enterobacteriales		
Family	Hominidae	Saccharomycetaceae	Enterobacteriaceae		
Genus	Ното	Saccharomyces	Escherichia		
Species	H. sapiens	S. cerevisiae	E. coli		

Taxonomy: Cataloging Life

Panthera because they have similar big cat-like features. Likewise, a group of similar genera comprise a **family**. For instance, lions, tigers, and leopards (genus *Panthera*) and domestic housecats (genus *Felis*) are categorized in the same family Felidae.

Taxonomists then use progressively more inclusive categories of classification. Related families are organized into an **order**, and orders are brought together in a **class**. Various classes comprise a **phylum** (pl. phyla) and all phyla are grouped together in a **kingdom**, except for the prokaryotes where there is no designed kingdom (see below). Notice in Table 2.1 that above the genus level, broader categories are not italicized but are capitalized.

The Domains of Life

Before the invention of the microscope, people believed the living world consisted of plants and animals. Therefore, they had little difficulty classifying life—after all, animals moved about, while plants were rigid and immobile. However, soon after Leeuwenhoek reported the existence of animalcules, it became clear that these organisms should be incorporated into the biological kingdoms. But scientists were unsure exactly where the microbes belonged. Even Linnaeus was unsure and therefore grouped them apart from plants and animals under the heading *Vermes* (as in "vermin") in a category he called *Chaos* (as in "confusion").

The classification of microbes remained somewhat chaotic until 1866, when the German naturalist Ernst H. Haeckel proposed a new kingdom called the Protista (the Greek word *protist* means "the very first"). This new third kingdom soon came to include virtually all microbes (bacteria, protozoa, algae, and fungi).

As the twentieth century progressed, advances in cell biology and microscopy identified structural differences between cells—the prokaryotes and eukaryotes. This and an interest in evolutionary biology led scientists to question the three-kingdom classification scheme. As a result, two more kingdoms were added to account for the increasing number of microorganisms being discovered and studied. All the fungi were placed in the kingdom Fungi and all the bacteria were placed in the kingdom Monera (FGURE 2.3). The kingdom Protista now included any living organism that was single-celled and not in the other kingdoms; namely, the unicellular algae and protozoa.

By the 1980s, better and more detailed research techniques were used to study living organisms. These techniques in biochemistry and molecular biology brought about another, and perhaps the most dramatic, change to the classification of life. First developed in the 1980s by Carl Woese and his coworkers at the University of Illinois, these scientists compared organisms based on specific genes in their DNA, the genetic information found in all organisms. Their comparisons from prokaryotes and eukaryotes showed all these genes could be lumped into three separate groups. Therefore, they suggested life should be cataloged into three groups called **domains**, which are the most inclusive and encompass all five original kingdoms. Today, this classification system is called the **three-domain system** (FIGURE 2.4). It is more commonly referred to as the "tree of life."

From the comparison of genes, molecular composition of the cell membrane, and other structural and biochemical traits, it was clear the prokaryotes in the kingdom Monera actually had two different evolutionary histories. Therefore, they represent two very different branches of prokaryotes and so were split into separate domains in the tree of life. The domain **Bacteria** includes most of the members with one set of similar genes, such as *E. coli*. The remaining prokaryotes with a different set of similar genes were assigned to the domain **Archaea**. Although many live at ordinary temperatures and conditions, such as floating in the open oceans and even living in the human mouth and gut, some members of the domain prefer more extreme environments, as **A Closer Look 2.2** highlights. The third domain, which includes all the eukaryotic organisms (protists, fungi, plants, animals), had a third set of similar genes and appropriately is called the domain **Eukarya**.

Archaea ar-kē' -a

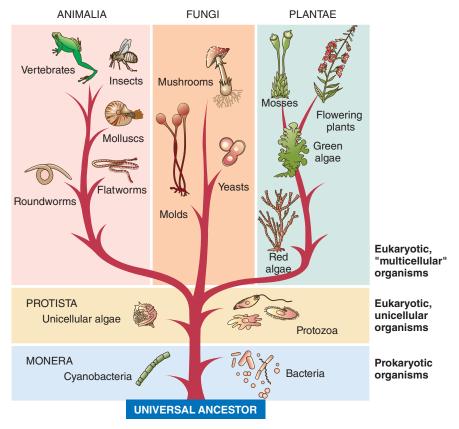


FIGURE 2.3 The Five-Kingdom System. This system of classification implies an evolutionary lineage, beginning with the Monera and extending to the Protista. Certain of the Protista were believed to be ancestors of the Plantae, Fungi, and Animalia. Divergence at each level was based on the mode of nutrition: photosynthesis, absorption, or ingestion. Unicellular or multicellular organization was also a key feature in the system.

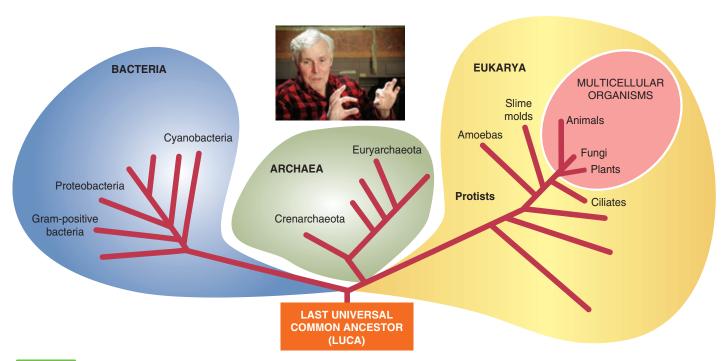


FIGURE 2.4 The Tree of Life. Carl Woese (inset) developed the tree of life and the three domain system to illustrate the relatedness of all living organisms. All organisms in the yellow portion of the domain Eukarya represent protists and all organisms in the three domains are single-celled, except for the plants, animals, and some fungi.

Inset © University of Illinois

A CLOSER LOOK 2.2

Prepare for Landing

"Ladies and gentlemen, as we start our descent to the San Francisco international airport, please make sure your seat backs and tray tables are in their full upright position. Make sure your seat belt is securely fastened and all carry-on luggage is stowed underneath the seat in front of you or in the overhead bins. Please turn off all electronic devices until we are safely parked at the gate. Thank you."—and, oh yes, look out your cabin window and marvel at the red-colored salt evaporation ponds below (see photo).



An aerial view of evaporation salt ponds.

© Aerial Archives/Alamy Images

Many members of the Archaea are truly startling in their ability to thrive in extremely hostile environments. Some of these so-called "extremophiles" can grow at extremely high temperatures, even above the boiling point of water. Others are quite happy growing in very acidic environments such as acid-laden streams around old mines. Yet other archaeal species prefer high concentrations of salt.

Salt evaporation ponds, also called salterns, are shallow, man-made ponds filled with seawater. As the water evaporates, the salt [primarily sodium chloride (NaCl) or

table salt] concentration rises. When the salt concentration goes above about 26 percent, NaCl starts to crystallize at the bottom of the ponds and it can be harvested.

Several microbial species found in these ponds are the halophiles (halo = "salt", phile = "loving"), which are microbial species that prefer growing in salty environments such as a saltern. Therefore, as the water in the salt ponds evaporates, a succession of microbes will develop as the salt concentration increases.

The salterns with the lowest concentration of salt appear green because they contain an abundance of halophilic green algae. Then, in ponds with somewhat higher salt concentrations, another green alga, *Dunaliella salina*, predominates. The green algae in such ponds not only provide pretty colors, they also speed up the rate of evaporation by absorbing sunlight (heat).

Then, as evaporation continues, ponds become too salty for the growth and survival of green algae and most microbes. Such high salt concentrations, however, become a perfect environment for the growth of the extreme halophiles—members of the Archaea. In high-salinity ponds, archaeal species, like *Halobacterium salinarum*, predominate and shift the color of the evaporation ponds from algal green to archaeal pink, orange, red, and even purple. Note: even though the genus *Halobacterium* sounds like it is in the domain Bacteria, it actually is a member of the Archaea.

Thus, the succession of microbes that develops in the various evaporation ponds not only provide vivid colors to the observer but also indicate the relative salinity of the pond. So, the next time you fly into San Francisco, look at the very colorful evaporation ponds filled with green algae or the extremely halophilic Archaea.

"On behalf of our airline and the entire crew, I'd like to thank you for joining us on this flight and we are looking forward to seeing you on board again in the near future. And weren't the microbes awesome?"

In the tree of life, the tips of the branches represent currently living organisms, be it a species, genus, phylum (e.g., Proteobacteria) or a common biological group (e.g., amoebas). They represent organisms that we actually know something about. Based on DNA sequence analysis, it is possible to trace evolutionary history (the red lines in Figure 2.4) back to hypothetical common ancestors of today's organisms, which are represented by the branch points called nodes (e.g., the node at the center right represents the ancestor that gave rise to the organisms in the domain Archaea and Eukarya). The root of the tree (e.g., the last common universal ancestor; LUCA) represents a hypothetical common ancestor that about 3.8 billion years ago gave rise to two lineages, one of which would evolve into the Bacteria. The other lineage in another 500 million years would split into two lines forming the beginnings of the Archaea and Eukarya.

Notice the viruses are not included in the tree of life. This omission is intentional because the viruses do not have the cellular organization characteristic of the prokaryotes and eukaryotes.

- Dunaliella salina dun-al-ē-el' -a sa-lī'-na
- Halobacterium salinarum hā'lo-bak-te·r' ē-um sal-inar'-um

2.3 Microscopy: Seeing the Unseen

As we have learned, the existence of microbes was largely a matter of speculation until the 1600s, for the simple reason that seeing them required a microscope and the magnifying lenses of the time lacked the quality needed to see tiny objects. Microscopes did not come into existence until the early 1600s, when a spectacle maker named Zacharius Janssen placed two lenses together to make a crude microscope. Galileo Galilei, the great astronomer, perfected the microscope in the 1620s, and Robert Hooke, the imaginative British microscopist, used the microscope to describe cells and other objects in the 1660s.

The stage now was set for discovering the microbial world. As mentioned in another chapter, Leeuwenhoek was the first to provide detailed descriptions of microbes, including protozoa, algae, and bacterial cells, because the lenses he made could magnify specimens over 200× and, by some accounts, up to about 400×. Hooke's microscope only magnified specimens about 20×.

Microbial Measurements and Cell Size

One of the defining features of microbes is their extremely small size. Therefore, they are not measured in inches, millimeters, or other well-known units, but in much smaller and less familiar units. The unit most often used for cellular measurements is the **micrometer** (sometimes referred to as the micron). A micrometer is a millionth of a meter. The abbreviation for a micrometer is expressed by using the Greek letter mu (written as μ) together with the letter m. Thus, the length of a typical bacterial cell would be expressed as 5 micrometers, or 5 μ m.

To conceptualize the extraordinarily small size of a micrometer, take a look at FIGURE 2.5. There are 1,000 μ m in one millimeter (mm). Put another way, the cells of the bacterial species Staphylococcus aureus (commonly called "staph") have a diameter of roughly 1 μ m. Therefore, 1 million staph cells lying side-by-side would occupy the space taken up by a single millimeter.

Most of the eukaryotic species of microbes are also measured in micrometers. For example, yeast cells are approximately 5 μ m in diameter and the cells of molds may be 25 μ m long or longer, with varying widths. Some protists may be as large as 100 μ m (0.1 mm), or about the size of the period at the end of this sentence.

At the opposite end of the microbial scale are the viruses, which are measured in **nanometers**. A nanometer (nm) is equivalent to one-thousandth of a micrometer (see Figure 2.5). Therefore, about 10 flu viruses (100 nm in diameter) would occupy the space of one micrometer.

Many cell structures are also measured in nanometers. For example, the bacterial ribosome, which manufactures proteins, is about 20 nm in diameter, so you could line up 50 ribosomes in the space of one micrometer.

FIGURE 2.6 illustrates the broad spectrum of microbial sizes, from the incredibly tiny viruses to the near visible protists. In fact, some filamentous and colonial algae and fungal molds are visible to the unaided eye. However, there are some notable size exceptions, as A Closer Look 2.3 describes.

Light Microscopy

Since Leeuwenhoek's time, great strides have been made in the construction of **light microscopes**, and today's instruments routinely achieve magnifications of 1,000× (**FIGURE 2.7a**). The component parts of the compound microscope are the ocular lens (or eyepieces), the objective lenses (closest to the object), and the substage condenser, which concentrates light coming from the light source on the object.

Most microscopes have a revolving nosepiece with three or more objective lenses: the low-power lens (10x), the high-power lens (40x), and the oil-immersion lens (100x).

■ Staphylococcus aureus staf-i-lō-kok' kus ô-rē-us

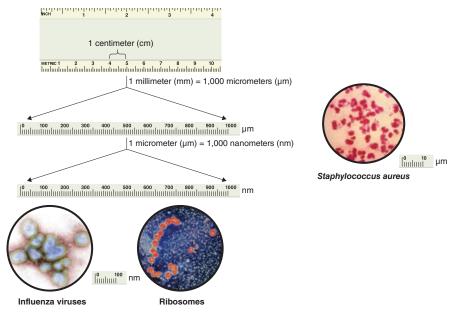


FIGURE 2.5 Measurement of Size. In the sciences, and in over two-thirds of the world's nations, the metric system is used to measure volumes, weights, and lengths (distance). The advantages are its accuracy and simplicity, as seen for the measurement of cell and virus size.

(a) Courtesy of F. A. Murphy/CDC (b) © Science Source (c) Courtesy of Dr. Norman Jacobs/CDC

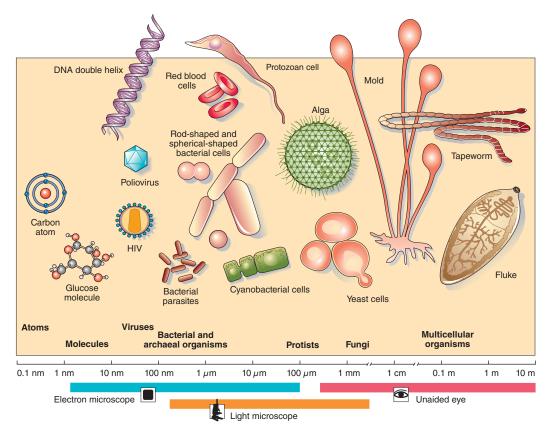


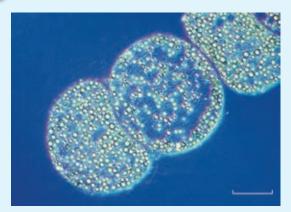
FIGURE 2.6 Size Comparisons among Atoms, Molecules, Viruses, and Microbes (not drawn to scale). Although tapeworms and flukes usually are macroscopic, they are often studied by microbiologists because of their disease potential.

A CLOSER LOOK 2.3

Exotic and Extreme

Copper mines can be acidic caldrons. Take, for example, the Richmond Mine at Iron Mountain near Redding, California. It is the source of the most acidic water naturally found on Earth. And believe it or not, there are acid-loving prokaryotes present that form a pink, floating film several millimeters thick on the surface of the hot, toxic water, which has a pH of 0.8. If that isn't amazing enough, in 2006 University of California scientists identified one of these microbes, a member of the domain Archaea, as the smallest living organism yet discovered. Called ARMAN (Archaeal Richmond Mine Acidophilic Nanoorganism), it is only 0.2 to 0.4 µm in diameter (about the size of a large virus). By comparison, an Escherichia coli cell is three times this diameter and has up to 100 times the cell volume. ARMAN cells are free living, they contain few ribosomes, and possess a relatively small number of genes.

At the other extreme, while on an expedition off the coast of Namibia (western coast of southern Africa) in 1997, scientists from the Max Planck Institute for Marine Microbiology in Bremen, Germany, found a bacterial monster in sediment samples from the sea floor. Named *Thiomargarita namibiensis* (see A Closer Look 2.1), these chains of spherical cells (see figure) were 100 µm to 300 µm in diameter—but some as large as 750 µm—about the diameter of the



Light microscope image of *Thiomargarita* namibiensis cells. (Bar = 100 μ m.)

Courtesy of Heide Schulz-Vogt, Max Planck Institute of Marine Microbiology, Germany

period in this sentence. Their volume is about 3 million times greater than that of *E. coli*. Another closely related strain was discovered in the Gulf of Mexico in 2005.

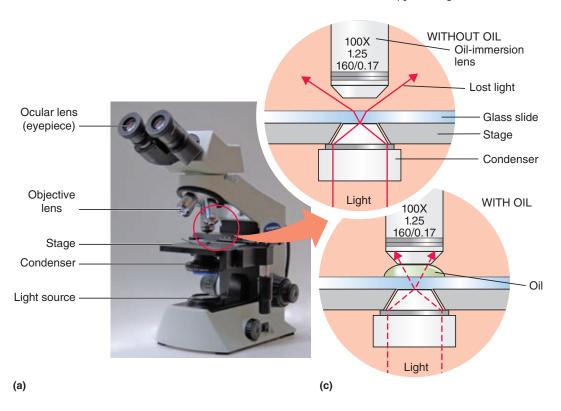
Yes, the vast majority of microorganisms are of typical microscopic size, but exceptions have been found in some extremely exotic places.

Each objective lens magnifies the object and creates an "intermediate image" in the tube of the microscope (FIGURE 2.7b). The eyepiece then uses this image as an object, magnifying it even more, and forms the final image seen by the observer. The total magnification achieved by the instrument is thus the magnification of the objective lens multiplied by the magnification of the eyepiece. For example, the low-power objective (10×) when used with a 10× eyepiece lens yields a total magnification of 100×.

Because the oil-immersion objective lens must be placed extremely close to the microscope slide, it is very difficult to obtain sufficient light for viewing because as light rays pass through the top surface of the microscope slide, they are bent by the glass and miss the exceptionally small opening in the oil-immersion objective (FIGURE 2.7c). The amount of light can be increased considerably by placing a drop of oil in the gap between the oil-immersion objective and the slide. The oil, known as immersion oil, has the same refractive index (or light-bending ability) as glass, so the light coming out of the glass slide does not bend away as it does in air. Rather, the light continues on a straight path into the 100× objective, and the object is illuminated sufficiently to be seen clearly.

Other Types of Light Microscopy

While the light microscope is considered the standard tool of microbiology, scientists use other types of optical configurations on the light microscope for viewing particular types of microbes or behaviors. One example is **dark-field microscopy** that produces a white image on a dark or black background. The light microscope uses a special condenser system to illuminate objects from the sides rather than from the bottom. The effect is somewhat like seeing dust particles illuminated when a beam of sunlight



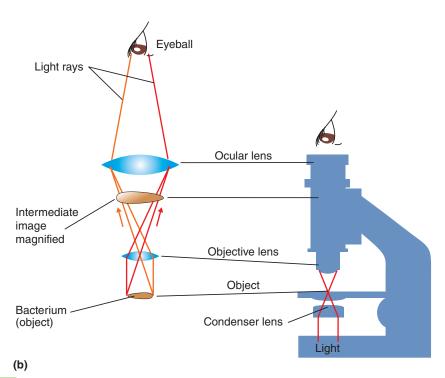


FIGURE 2.7 Light Microscopy. (a) This is a familiar light microscope used in many instructional and clinical laboratories. Note the important features of the microscope that contribute to the visualization of the object. (b) Image formation with the light microscope requires the light to pass through the objective lens, forming an intermediate image. (c) When using the oil-immersion (100×) lens, light rays enter the air and bend (solid arrows), missing the objective lens. However, they remain on a straighter line (dashed arrows) when oil is placed between lens and slide.

(a) Courtesy of Dr. Jeffrey Pommerville

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passes through a darkened room. Spiral-shaped bacterial cells can be seen clearly with this instrument (FIGURE 2.8a).

Another valuable configuration is fluorescence microscopy. For this set up, the light microscope contains an ultraviolet (UV) light source. After the slide specimen has been coated with a fluorescent dye, the UV light is directed at the specimen now on the microscope. When the UV light strikes the dye, the dye emits visible light and the object appears as a brightly glowing image whose color varies with the type of fluorescent dye used (FIGURE 2.8b).

Electron Microscopy

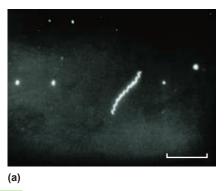
The light microscope with its optical configurations increases the lower limits of human vision and permits us to see most microbial cells. However, with the development of the electron microscope in the 1940s, a whole new world opened up to scientists because this instrument represented a quantum leap in magnification beyond the capabilities of the light microscope. Microbiologists could now see the viruses, an entire group of microbes previously invisible, and they could visualize the finer structures inside prokaryotic and especially eukaryotic microbial cells.

With an electron microscope, a beam of electrons passes through a vacuum tube. Then, magnets, rather than glass lenses, focus the beam on the object prepared for viewing. Acting similar to a beam of light, the electrons bounce off, are absorbed by, or are transmitted through the object and create a final image. The image can be viewed on a microscope screen or television monitor, or captured in digital format for a permanent record.

Two types of electron microscopes are in widespread use today: the transmission electron microscope (TEM) and the scanning electron microscope (SEM). The TEM (FIGURE 2.9) produces images of a specimen that previously had been cut into thin slices (100 nm thick), while the SEM uses whole cells and permits us to see the surfaces of cells in three dimensions. The final magnification possible with the TEM is approximately 200,000× and the microscope can see objects as small as 2 nm. The SEM produces a final magnification of about 20,000× and will resolve objects as small as about 7 nm.

FIGURE 2.10 shows images of the bacterium Pseudomonas taken with the light microscope, and with the TEM and SEM.

Pseudomonas sū-dō-mō näs



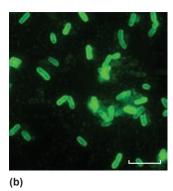


FIGURE 2.8 Observing Cells with Other Types of Light Microscopy. The two images show some common techniques ([a] dark field; [b] fluorescence) for contrasting bacterial cells. These methods require special optical configurations added onto the light microscope. (a and b, Bar = 10 μ m.)

(a) Courtesy of Schwartz/CDC. (b) Courtesy of Larry Stauffer, Oregon State Public Health Laboratory/CDC.

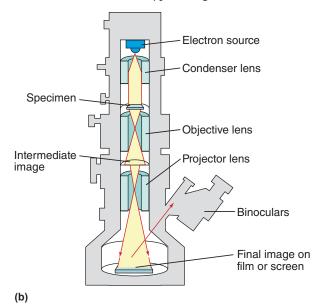




FIGURE 2.9 The Electron Microscope. (a) A transmission electron microscope (TEM). (b) A schematic of the vacuum tube. A beam of electrons is emitted from the electron source and electromagnets function as lenses to focus the beam on the specimen. The image is magnified by objective and projector lenses.

(a) Courtesy of Cynthia Goldsmith/James Gathany/CDC

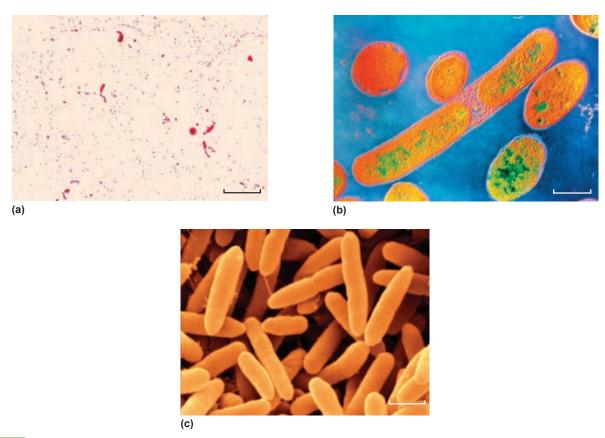


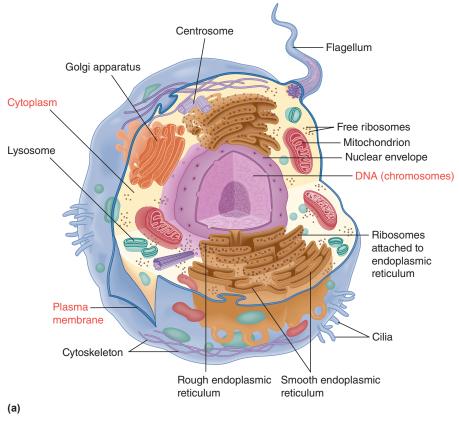
FIGURE 2.10 Light, Transmission, and Scanning Electron Microscopy Compared. Three false-color images of the bacterial genus *Pseudomonas* as seen with three types of microscopy. (a) A photograph of stained cells as seen with the light microscope. (Bar = 5.0 μm.) (b) A view of sectioned cells taken with a transmission electron microscope. (Bar = 1.0 μm.) (c) A view of whole cells taken with a scanning electron microscope. (Bar = 2.0 μm.) The difference in perspective between the three microscope images is clear.

(a) Courtesy of Dr. William A. Clark/CDC. (b) © CNRI/Science Source. (c) © SciMAT/Science Source.



2.4 Cell Ultrastructure: Comparing Eukaryotic and Prokaryotic Cells

With the use of the electron microscopes, especially the TEM, a much clearer and more detailed view inside cells is possible. Today, we have a fairly good idea of the finer structural details inside a cell, which is often referred to as the **ultrastructure** of the cell. To finish this chapter, let's see what's inside a typical eukaryotic and prokaryotic cell (FIGURE 2.11). Another chapter will discuss the bacterial structures in more detail.



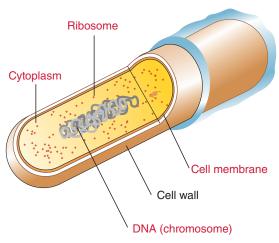


FIGURE 2.11 A Stylized Comparison of a Eukaryotic and Prokaryotic Cell. (a) A protistan cell represents a typical eukaryotic cell. Note the variety of the cellular features, some of which are noted in the text. (b) A prokaryotic cell, typical of *Escherichia coli*. Relatively few structures are seen with light microscopy, the presence of a cell nucleus is the primary feature distinguishing the eukaryotic cell from the prokaryotic cell. Universal structures common to all cells are indicated in red.

(b)

Cell Structure

All cells are surrounded by a selective barrier called the **cell** or **plasma membrane** that separates the environment from the semifluid **cytoplasm** inside the cell. Being a selective barrier means the membrane can control what enters and leaves the cell. The cytoplasm has the consistency of olive oil and contains many nutrients like sugars and salts. Suspended in the cytoplasm are various cellular components.

Notice in Figure 2.11 that both prokaryotic and eukaryotic cells contain **chromosomes**, which carry the genetic information (genes) in the form of DNA. One major difference between the two cell types is the way the DNA is organized. In eukaryotic cells, the chromosomes are surrounded by a membrane (nuclear) envelope forming the **cell nucleus**. In a prokaryotic cell, the DNA chromosome is not surrounded by any membranes and lies "bare" in the cytoplasm.

Also inside the eukaryotic cell are several complex, membrane-enclosed cellular compartments, called **organelles** having specific functions. Among the more prominent organelles are the cell nucleus, an "endomembrane system" and mitochondria (and chloroplasts in plants and algal cells). Bacterial and archaeal cells have no such organelles, but both prokaryotes and eukaryotes have **ribosomes**, the tiny bodies in the cytoplasm where proteins are constructed based on information received from the genes. In fact, it was one of the genes needed to produce ribosomes that Woese used to construct the three domains and the tree of life. **TABLE 2.2** lists many of the cell structures and organelles with their functions.

Throughout this discussion of cells and sizes, have you wondered why most microbial cells stay so small compared to their eukaryotic relatives? A Closer Look 2.4 provides the answer.

A CLOSER LOOK 2.4

Size Matters

Take a look back at A Closer Look 2.3. The *Thiomargarita* namibiensis cells shown in the photo are huge! Most bacterial cells are at least 100-times smaller; that is, not the 100-300 μ m but rather more like 2 μ m typical of an *Escherichia coli* cell. How can some bacterial species like *Thiomargarita*, though rare, survive as such large cells?

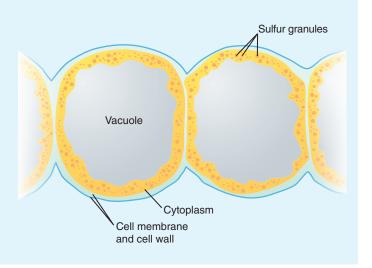
The basics of cell size can be simply stated as follows: size matters. If a cell gets too large, nutrients needed for metabolism cannot pass across the cell membrane fast enough to accommodate the metabolic demands of the cell. In most cases, such cells would die either from "starvation" or from the buildup of toxic waste products in the cytoplasm that cannot be eliminated fast enough.

So, for species like *E. coli*, when the cell reaches a critical size, it will divide into two smaller cells, making the transport of nutrients and waste products much easier and highly efficient.

But what about *T. namibiensis* and similar "bacterial giants?" The cells of *T. namibiensis* have solved the size problem by "pushing" all the cytoplasm out toward the edge of the cell (see figure). In other words, the whole central part of the cell is one big cavity – well it is not really a cavity but rather a space filled up with essential

nutrients to "feed" the thin layer (0.5-2 μ m thick) of cytoplasm. In fact, this thickness is identical to the cytoplasmic thickness of an *E. coli* cell.

By the way, the larger eukaryotic cells have specialized transport systems (endomembrane and cytoskeleton) to more rapidly transport essential materials throughout the cytoplasm. And the viruses have no independent metabolism or need for nutrients, so they can be exceedingly small – as most are.



2.2 A Comparison of Eukaryotic and Prokaryotic Cell Structures/Processes					
Cell Structure/ Process	Function	Prokaryotes	Eukaryotes		
Cell/plasma membrane	Semipermeable barrier separating environment from cell cytoplasm	Yes	Yes		
Cell nucleus	Houses the genetic information needed for growth and metabolism	No structure	Yes		
	Presence of DNA	Yes, as a single circular chromosome in cytoplasm	Yes, as multiple, linear chromosomes surrounded by a double membrane		
Cytoplasm	Fluid and contents that fill the cell and in which most metabolism occurs	Yes	Yes		
• Endomembrane system	Membranes that divide the cell into functional and structural compartments and regulates protein traffic	No	Yes		
• Cytoskeleton	Cytoplasmic cellular scaffolding for transport and cell division	Yes (organization unique from eukaryotes)	Yes		
 Ribosomes 	Site of protein manufacture	Yes	Yes		
• Mitochondria	Conversion of chemical energy to cellular energy (ATP)	No structure	Most		
	Make ATP	Yes (on cell membrane)	Yes		
 Chloroplasts 	Conversion of light energy to chemical energy	No structure	Algae and plants only		
	(photosynthesis)Carry out photosynthesis	Some (on cell membrane)	Algae and plants		
Exterior structures					
• Cell walls	Cell structure and water balance	Most	Algae, fungi, and plants		
• Flagella	Cell movement (motility)	Some (structurally unique from eukaryotes)	Some		
• Cilia	Cell movement (motility)	No	Some protists and animals only		

A Final Thought

The diversity of life forms has astounded scientists for centuries, and the challenge of characterizing and categorizing organisms (taxonomy) has been the focus of many scientists. The challenge of naming and categorizing this vast diversity is nowhere more complicated than it is with the microbial world. When it comes to microscopic life forms, which are often unicellular, the scientist is dependent on different forms of microscopy and various biochemical, genetic, and molecular means for detecting cellular activity. The fruits of this labor are shown in the three domain system, which visibly describes the various forms of microscopic life and which has challenged our thinking about the basic nature of life itself. The history of taxonomy, from Linnaeus to Woese, is a magnificent study of life forms, and we encourage you in this Chapter to gain an appreciation of the diversity of microorganisms, how they can be viewed, and the fundamental differences in cell architecture between domains.

Questions to Consider

- 1. A student is asked on an examination to write a description of the fungi. She blanks out. However, she remembers that fungi are eukaryotes, and she recalls the properties of eukaryotes. What information about eukaryotes from this chapter can she use to answer the question?
- 2. A local newspaper once contained an article about "the famous bacterial genus ecoli." How many errors can you find in this phrase? Rewrite the phrase with the mistakes corrected.
- 3. In 1987, in a respected scientific journal, an author wrote, "Linnaeus gave each life form two Latin names, the first denoting its genus and the second its species." A few lines later, the author wrote, "Man was given his own genus and species *Homo sapiens*." What is conceptually and technically wrong with both statements?
- **4.** Biologists tend to be collectors and compulsive classifiers. Why do you think this is so? Also, which classifier mentioned in this chapter do you think had the most impact on the science of his day?
- **5.** Microbes have been described as the most chemically diverse, the most adaptable, and the most ubiquitous organisms on Earth. From this chapter, what can you add to this list of "mosts"?
- **6.** Prokaryotic cells lack the extensive group of organelles found in most eukaryotic cells. Provide a reason for the structural difference. (Hint: remember size matters!)
- 7. A classmate who missed last week's first lab on microscopy observes a lab partner in this week's lab using oil immersion with the 100× objective lens. She asks her partner why he used it. "To increase the magnification of the microscope" was his answer. Do you agree or disagree? Why?
- 8. Every state has an official animal, flower, and/or tree, but one state has an official bacterial species named in its honor: *Methanohalophilus oregonense*. What's the state and decipher the meaning of the genus name. (Note: *ense* = "belonging to")

Key Terms

Informative facts are necessary for the expression of every concept, and the information for a concept is founded in a set of key terms. The following terms form the basis for the concepts of this chapter. On completing the chapter, you should be able to explain and/ or define each one.

ArchaeakingdomBacterialight microscopebinomial nomenclaturemicrometer (μm)cell (plasma) membranenanometer (nm)

cell nucleus order
chromosome organelles
class phylum (pl. phyla)
cytoplasm prokaryote
dark-field microscopy ribosome

domain scanning electron microscope (SEM)

Eukarya species eukaryote taxonomy

family three-domain system

fluorescence microscopy transmission electron microscope (TEM)

genus (pl. genera) ultrastructure