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# MICROBIOLOGY

## A Very Short Introduction

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# Chapter 1
# Microbial diversity

Earth is dominated by microorganisms. It can be difficult to appreciate this fact for the obvious reason that these forms of life are invisible to the unaided eye. We see plants and animals and interact with them in a deliberate fashion, and for most of human history we had no proof that anything smaller than insects existed. The Roman philosopher Lucretius edged close to the truth with his conjecture that 'certain minute creatures... enter the body through the mouth and nose and cause serious diseases'. His musings began to make sense after the invention of the microscope in the 1600s. The numbers of microbes are staggering. Tens of millions of bacteria live in a pinch of soil; a drop of seawater contains 500,000 bacteria and tens of millions of viruses; the air is filled with microscopic fungal spores, and a hundred trillion bacteria swarm inside the human gut. Every macroscopic organism and every inanimate surface is coated with microbes; microbes grow around volcanoes and hydrothermal vents; they live in blocks of sea ice, in the deepest oceans, and thrive in ancient sediment on the seafloor. Microbiology is the scientific study of these smallest forms of life. It concerns the biology of the bacteria, archaea, fungi, and an astonishing variety of unicellular organisms called protists. Microbiologists also study viruses, whose structure is far simpler than any kind of cell.

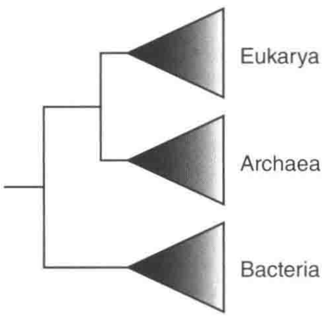
The majority of macroscopic organisms rely upon the energy harvested from the sun by photosynthesis: they are plants, they eat plants, or they consume animals that eat plants. Microorganisms show a greater range of metabolic lifestyle. The most familiar species act as decomposers, recycling the substance of dead plants and animals. Other microbes are photosynthetic. These include the cyanobacteria and a variety of protists that we call algae. In addition to decomposers and photosynthetic microbes, diverse bacteria and archaea are powered by metabolic processes fuelled by hydrogen gas, sulfur, and simple molecules including ammonia and methane. These chemical pathways enable microorganisms to support entire ecosystems in locations of perpetual darkness. The biochemical virtuosity of bacteria and archaea on Earth has encouraged astrobiologists to speculate about microbial life in a subsurface ocean on Jupiter's moon Europa and in a methane-fuelled ecosystem on Saturn's moon Titan.



Experiments in optics at the beginning of the 17th century allowed European scientists, including Galileo, to develop the first microscopes shortly after the invention of the telescope. The earliest microscopic observations were made on insects and the first illustrations of microorganisms were published in 1665 by Robert Hooke, who described the spore-producing structures of fungi. Hooke's contemporary Anton van Leeuwenhoek went much further in his investigations on the microbial world, being the first to describe bacteria, including large, crescent-shaped cells scraped from his teeth, a variety of protists, and yeast from beer. Despite important microscopic investigations by a handful of clever scientists in the 1700s, little progress was made in microbiology until the next century when Louis Pasteur demonstrated that sterilized broth remained sterile as long as it was isolated from airborne microbes. This rigorous experimental work in the 1860s disproved classical ideas about the spontaneous generation of organisms. Later, Pasteur developed vaccines against anthrax (caused by the bacterium *Bacillus anthracis*) and rabies (caused by a virus). Using mice in his experiments, Robert Koch identified the anthrax bacterium in the 1870s and designed a systematic method for identifying the cause of any infectious disease. This method, called Koch's Postulates, requires the investigator to identify the disease-causing organism in a diseased animal, grow this 'germ' in a pure culture, use this culture to infect a healthy animal, and isolate the same organism from the experimental host.

Even after the invention of the microscope, however, the classical Aristotelian division of life into animals and plants was unchallenged until Ernst Haeckel created a third category for unicellular species, named 'protista', in the 1860s. Today, we recognize three primary groups of organisms: Bacteria, Archaea, and Eukarya (Figure 1). Informal names of these groups—bacteria, archaea, and eukaryotes—are used throughout the rest of this book. Microbiologists study microscopic organisms belonging to



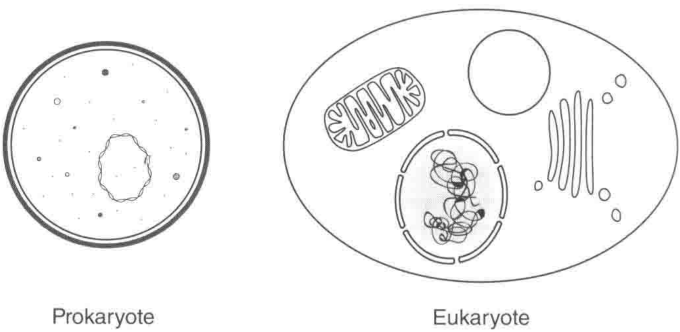



**1. Three primary groupings, or domains, of organisms displayed in the form of an evolutionary 'tree' in which genetic modification over the course of billions of years is indicated from left to right. The diagram shows that Bacteria, Archaea, and Eukarya evolved from a common ancestor, and that the Archaea and Eukarya are more closely related to one another than either group is connected to the Bacteria. Genetic comparisons suggest that the Eukarya may have evolved from a group of Archaea. According to this research it makes sense to condense all organisms into a pair of domains, the Bacteria and Archaea**

all three groups. All bacteria and archaea are microscopic; soil amoebae, diatoms, dinoflagellates, and single-celled green algae are examples of microscopic eukaryotes.

There is a crucial distinction between bacteria and archaea, which are prokaryotes, and the eukaryotic microbes (Figure 2). Genes of prokaryotes are organized in the form of a single circular chromosome situated in the fluid interior of the cell. This chromosome constitutes the genome of prokaryotes. The genome is defined as the entire collection of hereditary information in the cell. Genomes, microbial and otherwise, comprise genes that encode proteins, intervening sequences that regulate gene expression, and non-coding sequences that do not specify proteins and used to be called 'junk DNA'. (We know now that a great deal of the non-coding DNA performs important biological functions.) Eukaryotes tend to carry more genetic information than prokaryotes. Most of the genome of a eukaryote cell is encoded in multiple chromosomes surrounded by an envelope of membranes that defines the nucleus. DNA is also found in the form of circular chromosomes inside the mitochondria and






**2. Diagram showing the relatively simple cell structure of prokaryotes (bacteria and archaea) contrasted with the more complex makeup of eukaryotes. The single chromosome of bacteria and archaea is situated within the cytoplasm. The multiple chromosomes of eukaryotes are housed within the nucleus**

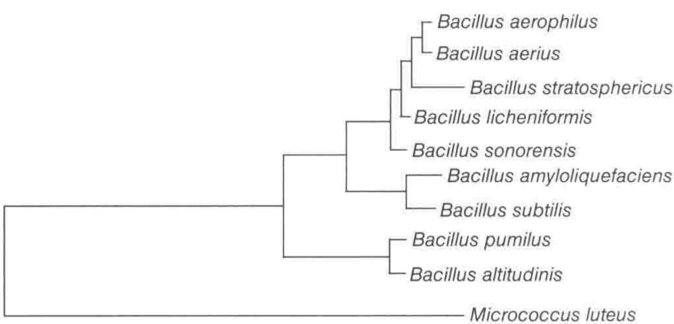
chloroplasts of eukaryotic cells. Mitochondria are organelles that supply eukaryote cells with energy. Chloroplasts are organelles that perform photosynthesis in plants and algae. Both types of organelle evolved from bacterial cells that were absorbed by the ancestors of today's eukaryotes. This process of incorporation is called endosymbiosis.

Structural and functional characteristics like cell shape and metabolic activity have been used to identify some of the groupings of bacteria since Pasteur's time. Cells of the syphilis bacterium (*Treponema pallidum*), for example, are coiled like a corkscrew. Their unusual shape is evident in samples of infected tissue from syphilis patients and is characteristic of related bacteria that we classify as Spirochetes. These structural features are a less reliable guide to relatedness in other cases and most rod-shaped bacteria and archaea look the same under the microscope. The Gram-staining technique, developed in the 19th century, is another guide to identification. This is described as a differential stain because it colours bacteria with thick cell walls purple (positive) and thin-walled bacteria pink (negative). Staining reactions are a useful diagnostic tool, allowing a medical technician to narrow the list of possible bacteria collected from a throat swab. But the Gram stain is a poor guide to the relatedness of species. For this reason, microscopic methods have been largely superseded by genetic techniques for developing modern classification schemes that reflect evolutionary kinship.

Research on evolutionary relatedness, known as molecular phylogenetic analysis, relies upon comparisons between the DNA sequences of different species. For bacteria, a gene that encodes part of a cell structure called the ribosome is crucial for the identification of species. Ribosomes are molecular machines that produce proteins. In general, if the sequence of this 16S ribosomal RNA (rRNA) gene of different bacterial isolates, or strains, differs by 3 per cent or less, these microorganisms are regarded as members of the same species. This is not a perfect method, and probably underestimates the number of species, but it is very useful for identifying bacteria from environmental samples. Comparisons between 16S rRNA genes are used to construct phylogenetic trees that reveal evolutionary relationships between different species and links between groups of bacteria (Figure 3). Analysis of other genes is essential for discriminating between strains within a single species. Comparisons of whole genomes have also been used with spectacular success to examine the details of bacterial evolution.

Specialists who study bacterial taxonomy have catalogued more than 11,000 species of bacteria. This list is biased toward bacteria of medical significance and those that can be grown in culture easily. Experiments in which bacterial genes have been sequenced without growing the cells in the laboratory show that a teaspoon of soil can contain thousands of unidentified species. Our bodies are home to an incredible range of microorganisms: molecular analysis has revealed 2,368 'species' of bacteria that live in the human navel! These results have encouraged researchers to suggest that there may be tens or even hundreds of millions of species of bacteria. Undercounting of bacteria, and other microorganisms,






**3. Phylogenetic tree showing relationships between species of *Bacillus* (bacteria) based upon comparisons between the sequences of their ribosomal RNA genes. The outgroup chosen as a reference for comparing the *Bacillus* species is the bacterium *Micrococcus luteus***