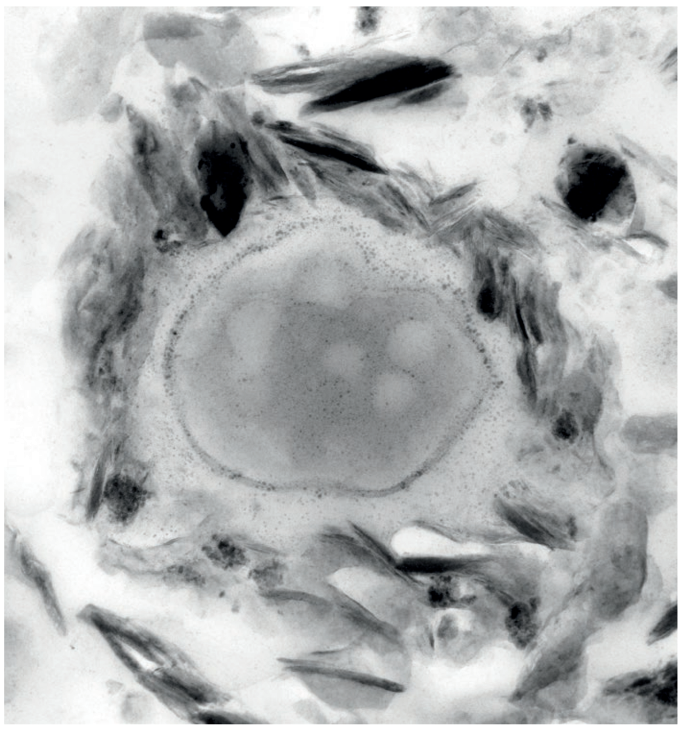
Prokaryota – Bacteria

Morphology

Bacteria are one of the two domains, along with Archaea, that include prokaryotic organisms [26]. The domain Bacteria comprises microscopic organisms, single-celled or with the cells forming simple associations. Most bacteria are 0.2 micrometres (µm) in diameter and 2-8 µm in length. Bacteria have a variety of shapes: round or spherical (commonly known as cocci), rod shaped (bacilli) and spiral (spirilla). However, many bacteria can assume several shapes (pleomorphic). Depending on how the newly formed cells adhere to each other, bacterial arrangements include singles, pairs, chains and clusters. When bacteria are motile (capable of moving) they have a specific structure (flagellum) for locomotion. The flagellum is a whip-like structure that can occur at one end, both ends, or all over the bacterial cell. Bacteria can live without oxygen (anaerobes) or depend on it to grow (aerobes). They can also be adapted to live either in the presence or absence of oxygen (facultative anaerobes). Some species of bacteria contain endospores or exospores (see box next page). If you break down the term endospore, 'endo-' means 'inside' and '-spore' refers to the 'dormant structure', so the endospore is a structure of resistance formed inside the cell. By contrast, the exospores develop externally. Spores are a bacterial cell's way of protecting itself against harsh changes in the environment or nutrient depletion. A spore protects the bacterial genetic material so that, when optimal conditions return, the bacterial cell can reform (germinate) and thrive again.

Taxonomy

Currently, there are 30 known and recognised phyla of bacteria. Highly diverse and abundant phyla in soil are Proteobacteria, Firmicutes, Actinobacteria and Cyanobacteria (see pages 34-35). However, some other phyla, such as Acidobacteria, can also be found in soil.



Transmission electron micrograph showing a soil bacterium (circle in the middle) surrounded by clay particles and compounds (polysaccharides) released by the bacterium itself. (CC)

Bacterial phyla

- In contrast to eukaryotic nomenclature, there is no official classification of prokaryotes because taxonomy remains a matter of scientific judgment and general agreement.
- The List of Prokaryotic names with Standing in Nomenclature (LPSN) is an online database that maintains and provides accurate names (nomenclature) and related information of prokaryotes according to the International Code of Nomenclature of Bacteria.
- The 30 phyla currently accepted by the LPSN are: Acidobacteria; Actinobacteria; Aquificae; Armatimonadetes; Bacteroidetes; Caldiserica; Chlamydiae; Chlorobi; Chloroflexi; Chrysiogenetes; Cyanobacteria; Deferribacteres; Deinococcus-Thermus; Dictyoglomi; Elusimicrobia; Fibrobacteres; Firmicutes; Fusobacteria; Gemmatimonadetes; Lentisphaerae; Nitrospira; Planctomycetes; Proteobacteria; Spirochaetes; Synergistetes; Tenericutes; Thermodesulfobacteria; Thermomicrobia; Thermotogae.
- Other existing phyla of bacteria, which cannot currently be cultured in the laboratory (see pages 64-65), are called candidate phyla. If these are included, the total number of phyla is 52.

Typical structures, the nodules, occur on the roots of plants that associate with symbiotic nitrogen-fixing bacteria. Nodules in the roots of (**a**) *Mimosa foliolosa*, a plant native to Brazil, formed by the nitrogen-fixing bacteria *Burkholderia* sp., (**b**) cowpea (*Vigna unguiculata*) formed by *Bradyrhizobium* sp. and (**c**) *Medicago italica* formed by *Sinorhizobium meliloti*.(FC, FMSM, NI)

Diversity, abundance and biomass

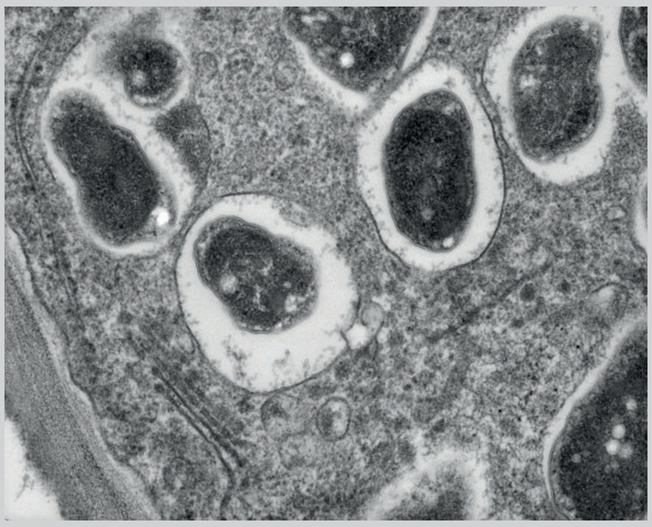
Most microbial species (more than 90 % according to the current estimates), including bacteria, still remain unculturable (i.e. they cannot be grown in any culture medium in the laboratory). This means that we do not yet know what they look like or what functions they carry out. Advances in molecular techniques (see pages 64-65) in the past 30 years have enabled us to understand more about these species by sequencing parts of their DNA. These advances have also allowed for the identification of new culturable species. Today there are approximately 2800 genera comprising approximately 15000 species of known bacteria. Soil microbial biomass is made up of bacteria, fungi and other microorganisms. This biomass represents 1 to 4 % of total soil carbon (up to three tonnes of carbon per hectare). The ratio of the size of bacterial to fungal biomass depends on soil properties and other environmental factors (e.g. soil pH, temperature and nutrient availability); for example, a 30-fold decrease in bacterial biomass was found when comparing high to low pH soils.

Microhabitats

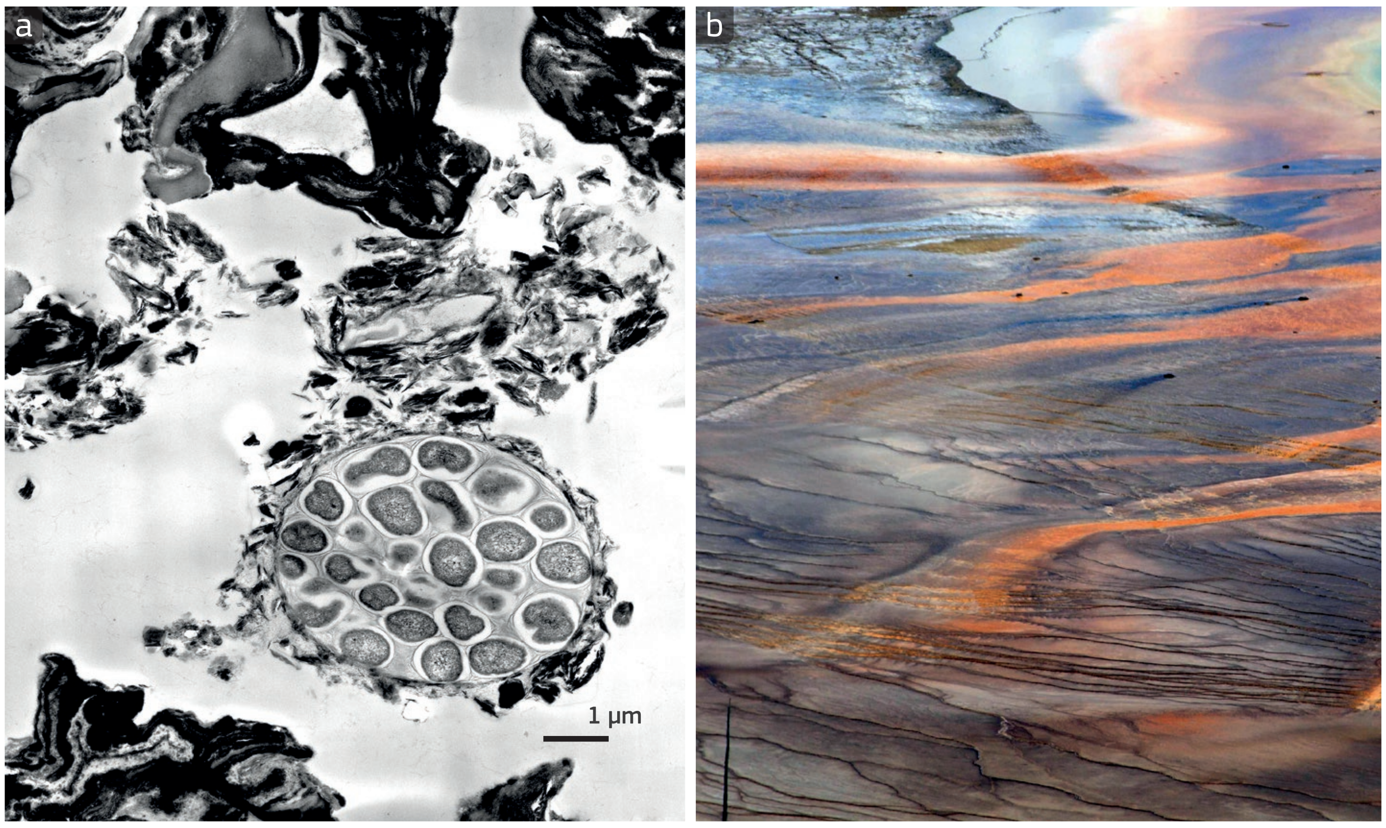
Unlike eukaryotes, bacteria can be found in a wide range of environmental, chemical and physical conditions including extremes of pH, temperature and salinity. Many soil bacteria are beneficial to human economic activities and are necessary for environmental sustainability. Bacteria are part of chemical cycles during which they release essential elements for recycling. They also decompose dead organic matter and are the only microbes capable of biological nitrogen N_2 fixation (see page 105). This is the ability to transform nitrogen (N_2) from the atmosphere (about 80 % of the atmosphere is N_2) into ammonium (NH_4^+) which is assimilated by eukaryotes, plants in particular. Bacteria can exist either as independent (free-living) organisms or as symbionts that depend on other organisms to live, subsisting either as mutualists, parasites or commensalists (see box below).

What is symbiosis?

- Symbiosis is a close and often long-term interaction between two different biological species.
- There are three main types of symbiosis:
  - mutualism is the way two organisms of different species exist in a relationship in which each individual benefits from the activity of the other;
  - commensalism is a class of relationship between two organisms where one organism benefits from the other without affecting it;
  - parasitism is a relationship between species, where one species, the parasite, benefits at the expense of the other, the host.
- Some symbiotic relationships are obligate, meaning that both symbionts entirely depend on each other for survival.
- Other relationships are facultative, meaning that they are not essential for the survival of either species. Individuals of each species engage in symbiosis when the other species is present.



The soil bacterium *Bradyrhizobium japonicum* colonises the roots of some plants and establishes a symbiosis. This high magnification image shows part of a plant cell with bacterial cells (dark circles) in it. (LH/DEMF)



(**a**) Soil bacteria living within the soil particles. They can also be found in extreme environments: (**b**) the reddish and brownish colours are mats of bacteria living around geothermal hot springs in Yellowstone Park in the USA. (FW, FKO)

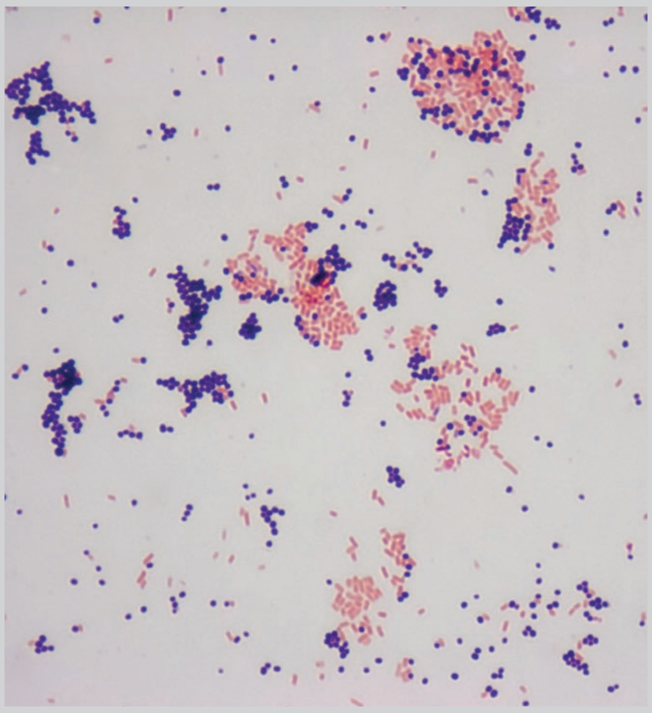
Prokaryota – Bacteria

Proteobacteria

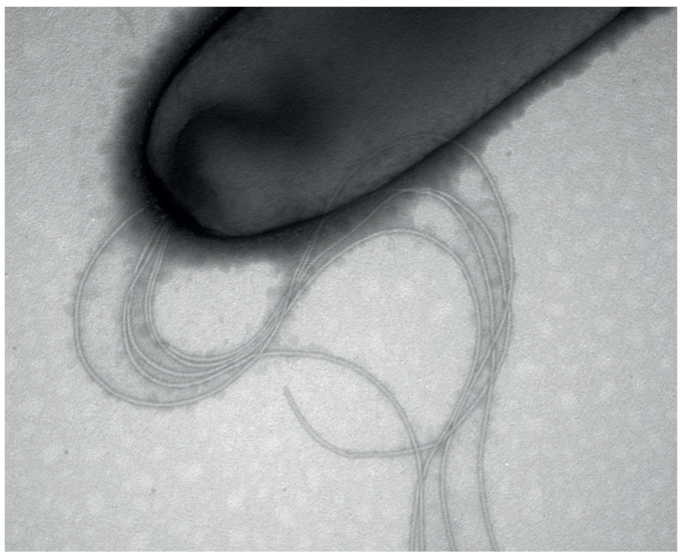
Proteobacteria is the largest and most diverse bacterial phylum [26]. It contains about 30 % of the total number of bacterial species. Proteobacteria comes from the name of the Greek god *Proteus*, which could take various forms, thus reflecting the enormous diversity of morphological and physiological characteristics observed in this bacterial phylum. Proteobacteria comprises the majority of Gram-negative (see box below) bacteria of medical (e.g. *Helicobacter*), veterinary (e.g. *Acinetobacter*), industrial (e.g. *Campylobacter*) and agricultural interest (e.g. *Bradyrhizobium*). It also comprises bacteria involved in carbon, sulphur and nitrogen cycles (including N_2 fixers – see pages 99, 105), phototrophic (i.e. organisms that obtain energy from light) and non-phototrophic, aerobic and anaerobic bacteria.

Gram-positive and Gram-negative

- Gram staining, also called Gram's method, is a method of differentiating bacterial species into two large groups: Gram-positive and Gram-negative, respectively. The name comes from the Danish bacteriologist Hans Christian Gram, who developed the technique. The technique is based on the use of a chemical compound, the crystal violet. The name refers to its colour, similar to that of the petals of a gentian flower. The Gram stain is almost always the first step in the identification of bacterial organisms.
- Gram-positive bacteria are bacteria that give a positive result in the Gram stain test. Gram-positive bacteria take up the crystal violet stain used in the test, and then appear to be purple-coloured when seen through a microscope. This is because in the cell wall of the Gram-positive bacteria there is a layer that retains the stain after it is washed away from the rest of the sample, in the decolourisation stage of the test.
- Gram-negative bacteria are a group of bacteria that do not retain the crystal violet stain. After staining with crystal violet, the excess is washed off with alcohol, which decolourises the bacteria since the layer in their cell wall is too thin to retain the stain. A counterstain is then added, which colours the bacteria red or pink.



Two species of bacteria, one of which is a Gram-positive coccus (*Staphylococcus aureus*, stained dark purple) and the other a Gram-negative bacillus (*Escherichia coli*, stained pink). (MP)



Transmission electron microscope image of *Pseudomonas aeruginosa*. *Pseudomonas* is a genus of Gram-negative Proteobacteria that can be found in many different environments, including soil. (LH/DEMF)

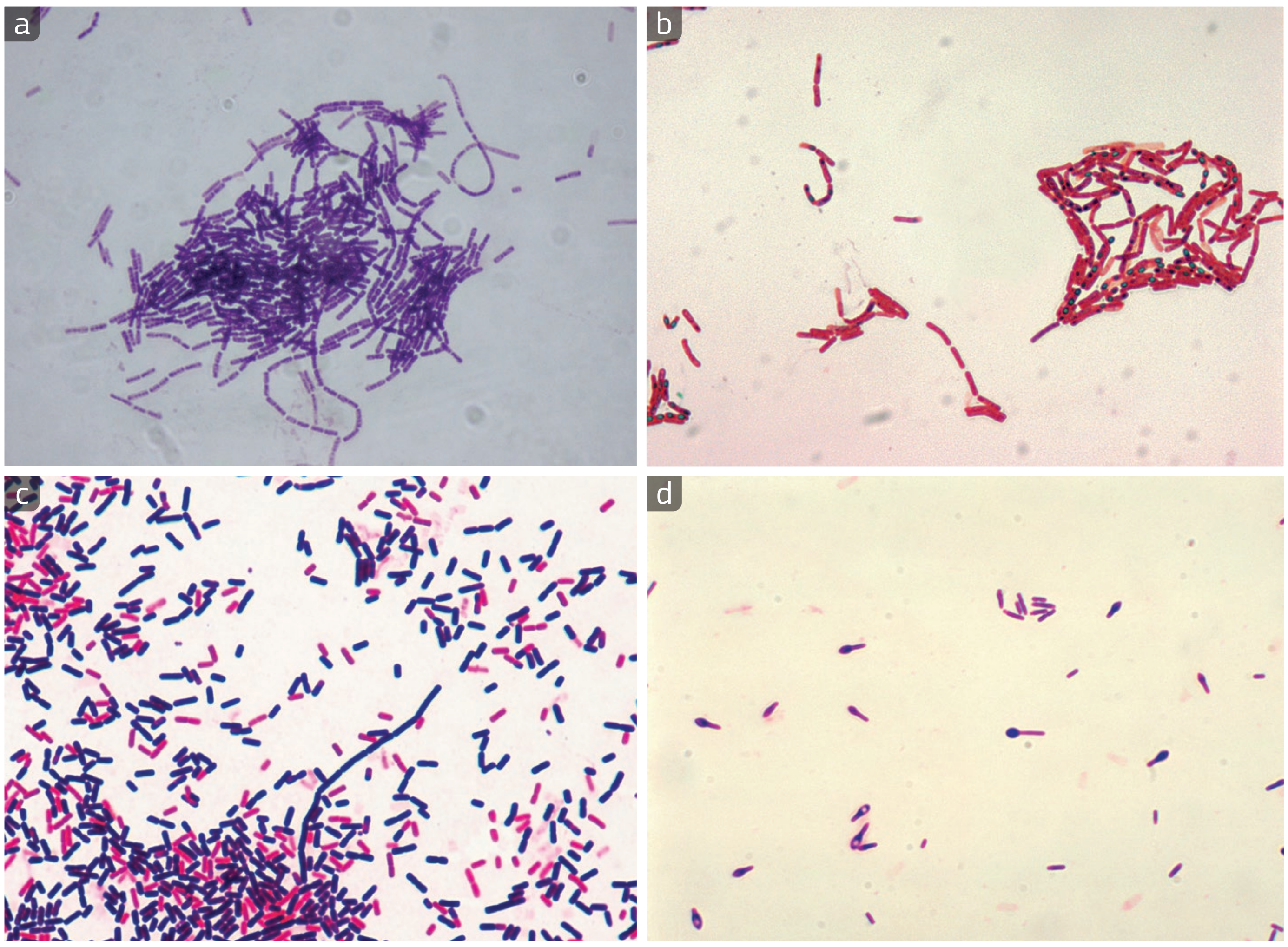
Firmicutes

The most representative genera in Firmicutes are *Bacillus* and *Clostridium,* which are obligate and facultative anaerobic bacteria, respectively [26]. These genera include important species of human and animal pathogens that produce resistant cell structures called endospores. Spores tolerate different types of stresses. For example, they are more resistant to heat than normal cells by a factor greater or equal to 10^5. Furthermore, they are 100 times or more resistant to ultraviolet radiation, and more tolerant to drought, antibiotics and disinfectants. Most *Bacillus* species, such as *B. cereus*, which causes contamination of food, are soil inhabitants. Due to their pathogenicity on some soil insects, some *Bacillus* species, including *B. popilliae*, *B. lentimorbus* and *B. thuringiensis*, have been successfully used in agriculture to control pests. *Bacillus* may also be dangerous: *Bacillus anthracis* is considered the most lethal biological weapon for human beings because it is the origin of anthrax (see box on page 108). Another Firmicute genus, *Paenibacillus,* includes important soil-living nitrogen fixers (see page 99). Nitrogen-fixing bacteria are also present in both *Bacillus* and *Clostridium* genera.

Bacteria as workers

- Many compounds are produced in large amounts by bacteria to be used for various purposes in industry and medicine. They can be a part of silk, cotton and rubber manufacturing. Bacteria also synthesise certain antibiotics, such as bacitracin and polymyxin.
- Bacteria are able to degrade complex compounds. For example, they break down the woody and tough tissues of jute, coconut, hemp and flax. They can also degrade hydrocarbons and clean up oil spills.

Bacteria can be used to 'eat' oil spills. (LSU)



(**a–b**) *Bacillus* and (**c–d**) *Clostridium* are the two most important genera of the phylum Firmicutes. *Bacillus* species are able to colonise a variety of habitats ranging from soil and insects to humans. *Clostridium* species from soil samples, manure and plant materials can be easliy grown and studied. (DS, LS, UCSFMC, GL)

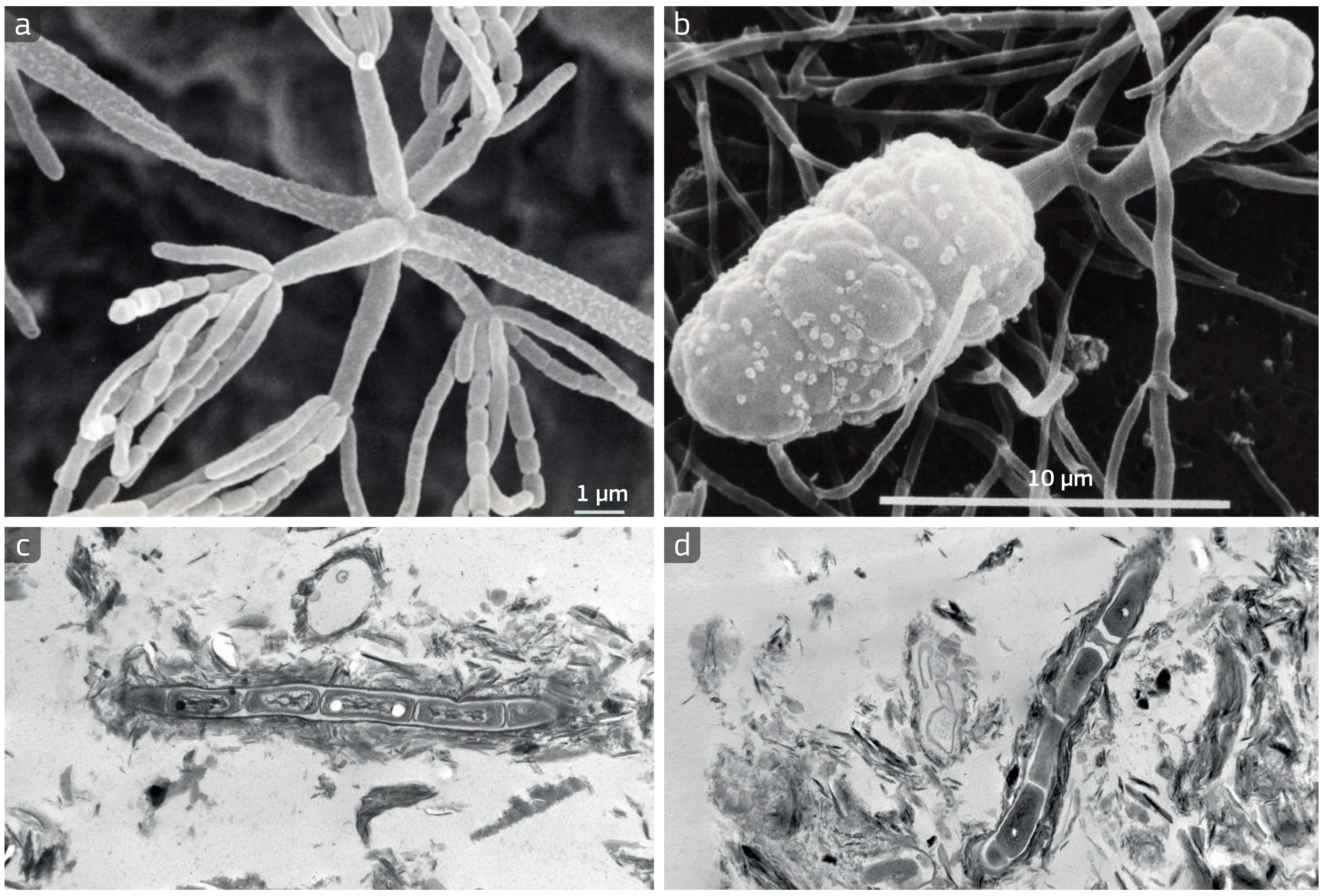
Endospores, what are they?

- Endospores can survive environmental assaults that would normally kill the bacterium. These stresses include high temperatures, high UV irradiation, desiccation and chemical damages. The extraordinary resistance properties of endospores make them of particular importance because they are not readily killed by many antimicrobial treatments.
- When favoured nutrients are exhausted, some Gram-positive bacteria may develop an extreme survival strategy: the formation of endospores.
- This complex development allows the bacterium to produce a highly resistant cell to preserve the cell's genetic material in times of extreme stress.
- The resilience of an endospore can be explained in part by its unique cellular structure. The outer coat surrounding the spore provides much of the chemical resistance. Beneath the coat there is a very thick layer called the cortex. Proper cortex formation is needed for dehydration of the spore core, which aids in resistance to high temperature. A germ cell wall is found under the cortex. This layer will become the cell wall of the bacterium after the endospore germinates. The inner membrane, under the germ cell wall, is a major permeability barrier against several potentially damaging chemicals. The centre of the endospore, the core, exists in a very dehydrated state and houses the cell's DNA.
- The process of forming an endospore is complex and requires several hours to complete.

Endospores of the Firmicutes (**a**) *Paenibacillus alvei* and (**b**) *Bacillus anthracis* are clearly visible (white spots and dark circle, respectively) inside bacterial cells. (TE, SZ)

Actinobacteria

Actinobacteria is a phylum of Gram-positive bacteria that have a highly diverse morphology, ranging from micrococci (spherical) and rods to branched filaments that resemble fungal hyphae (see box on page 39) [26]. The bacterial filaments are narrow (diameter from 0.5 to 2 µm) and can be short and rudimentary or extensively branched. Throughout their life cycles, Actinobacteria may combine these different forms. Their reproduction is by fragmentation of hyphae or through the production of spores. The spores may be of several types (e.g. arthrospores, very primitive spore type, formed through the breaking up of hyphal filaments in *Streptomyces* and zoospores, motile and flagellate spores, in *Spirillospora* and *Actinoplanes*). Spores are produced (from one to several in chains) on hyphae, in spore-producing structures (sporangia) or vesicles. The ecological niche of most Actinobacteria is the aerobic zone in soil. A striking feature of Actinobacteria is the production of extracellular enzymes that degrade complex macromolecules commonly found in soils (e.g. casein, starch, chitin, cellulose and lignocellulose). Furthermore, they synthesise and excrete thousands of metabolites, such as antibiotics. For example, Selman Waksman, one of the most important soil microbiologists, won the Nobel Prize for Medicine in 1952 for his discovery of streptomycin produced by bacteria of the genus *Streptomyces*. In addition to streptomycin, Streptomyces are capable of producing a wide variety of antibiotics with numerous properties: antibacterial, antifungal, antiviral, antitumor, antiparasitic, insecticide and weed controlling. Actinobacteria also includes the nitrogen-fixing bacteria of the genus *Frankia*, which form root symbioses with plants of eight botanical families (e.g. Betulaceae – see page 43). Other species belonging to the genera *Streptomyces* and *Corynebacterium* are plant pathogens. Animal pathogens are found among the genera *Corynebacterium*, *Actinomyces*, *Nocardia*, *Thermoactinomyces* and *Mycobacterium*. Among them, the *Mycobacterium avium-intracellulare-scrofulaceum* stands out as being lethal for people who have contracted the human immunodeficiency virus (HIV).



Scanning electron images show (**a**) branching filaments (hyphae) of *Streptomyces verticillus* with spores at their ends and (**b**) two young spore-producing structures (sporangia) developed from a single hypha of a *Frankia* species. (**c**-**d**) Actinobacteria among soil particles. (TH/MHA/SAJ, DDB/HAL/SAJ, FW)

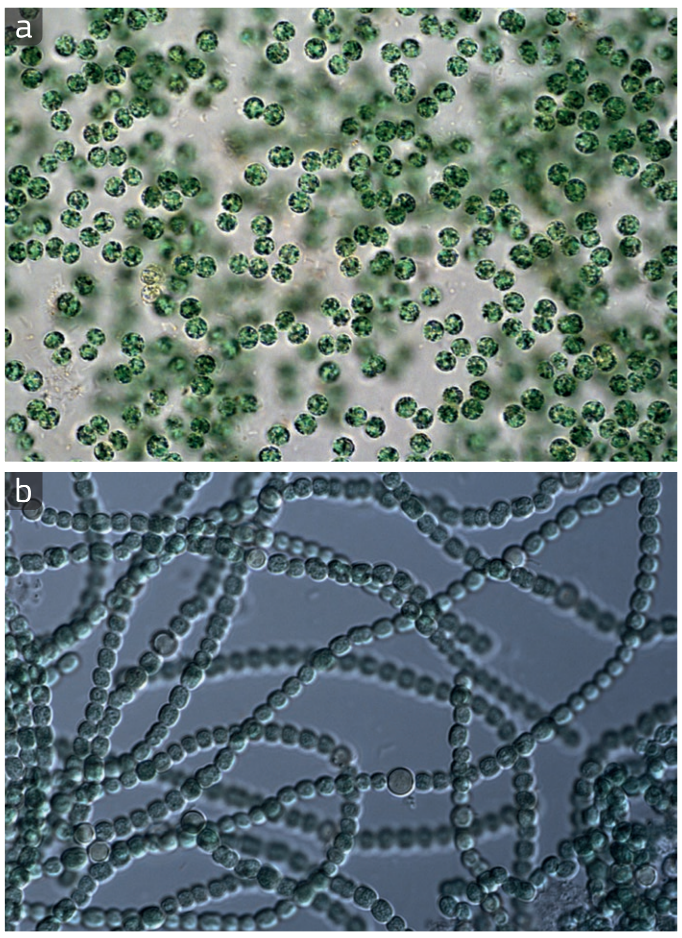
Why does the air smell of soil after rain?

- The earthy smell after it rains is linked to Actinobacteria.
- In particular, the molecule responsible for the aroma is known as geosmin.
- Geosmin is produced by the Gram-positive bacterium *Streptomyces*, a genus of Actinobacteria, and released when these microorganisms die.
- The human nose is extremely sensitive to geosmin and is able to detect it at very low concentrations.
- Geosmin is also responsible for the earthy taste of beetroots.

Different species of Actinobacteria can be identified by growing them on artificial substrates made with jelly-like substances and nutrients such as oatmeal (see pages 64-65). Different colours and shapes allow the distinction of different species. (PT/FIIRV)

Cyanobacteria

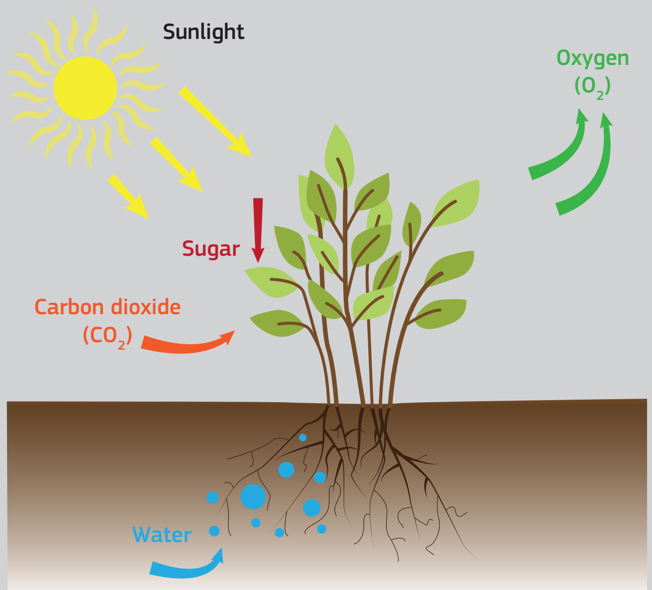
Cyanobacteria is a group of bacteria that are able to obtain their energy through photosynthesis. This is possible due to the presence of chlorophyll, which is also found in other photosynthetic organisms, such as algae and plants. Being photosynthetic, they manufacture their own food. This has caused them to be dubbed 'blue-green algae', though they have no relationship to any of the various eukaryotic algae. They are considered one of the most diverse groups of prokaryotes as they vary from unicellular to complex filamentous or branched forms. In some cases they have highly differentiated cells that carry out different functions, so they may be considered as truly multicellular organisms. Cyanobacteria have the distinction of being the oldest known fossils, more than 3.5 thousand million years old, in fact. The cyanobacteria have been tremendously important in shaping the course of evolution and ecological change throughout Earth's history. Indeed, the atmospheric oxygen that we depend on was generated by numerous cyanobacteria through photosynthesis. Furthermore, the photosynthetic structure of plant cells, the chloroplast, evolved from cyanobacterial ancestors. Cyanobacteria also contribute to the health and growth of many plants in another way: they have the ability to convert inert atmospheric nitrogen into ammonia (nitrogen fixation) that plants can use (see page 105). This process cannot occur in the presence of oxygen, so nitrogen is fixed in specialised cells called heterocysts. These cells have an especially thickened wall that contains an anaerobic environment. Cyanobacteria also form symbiotic relationships with many fungi, forming complex symbiotic organisms known as lichens (see page 42).



Cyanobacteria can have different forms: (**a**) unicellular and (**b**) filamentous. In the filaments it is possible to see bigger cells, called heterocysts, where nitrogen fixation takes place. (KSI)

What is photosynthesis?

- Photosynthesis is a process used by plants, algae and cyanobacteria to convert sunlight energy into chemical energy.
- This chemical energy is stored in carbohydrate molecules, such as sugars, which are produced from carbon dioxide and water;
- Oxygen is released as a waste product.
- Photosynthesis maintains atmospheric oxygen levels and supplies most of the energy necessary for life on Earth.



Schematic of photosynthesis. The sugars produced are stored in or used by plants, whereas oxygen is released into the atmosphere. (JRC)