Introduction to Microbiology

- **Microbiology** is the study of microorganisms, a large and diverse group of microscopic organisms which must be viewed with a microscope that exist as single cells or cell clusters; it also includes viruses, which are microscopic but not cellular .

- **Importance of Microbiology:**

The importance of microbiology includes:

1. Used in biomedical research, creation of medicines, environmental applications and new research tools.
2. Bacteria are important for fixing N2 in a usable form for plants.
3. Bacteria and some fungi are important in decomposition and recycling of materials.
4. Industry applications of microbiology: food industry ,medicine, research and biotechnology.

History of Microbiology

The history of microbiology can be summarized in the following:

- **1660's Robert Hooke** observed microorganisms for the first time with a microscope and coined the term "cell".

- **1822-1895 Louis Pasteur** Defined pasteurization to prevent spoilage of food by bacteria and disproved the scientific dogma of "Spontaneous Generation". He defined "Germ Theory" and demonstrated that germs were responsible for disease.
- **1843-1910 Robert Koch** identified anthrax and developed agar growth medium. Koch's postulates was a systematic method to establish the microbial cause of disease.

- **The microorganisms include :-**

1- **Viruses:**

Viruses lack many of the attributes of cells, including the ability to replicate depending on themselves. But only when it infects a cell does a virus acquire the key attribute of a living system, reproduction.

A viral particle consists of a nucleic acid molecule, either DNA or RNA, enclosed in a protein coat, or **capsid** (sometimes itself enclosed by an envelope of lipids, proteins, and carbohydrates). Proteins—frequently glycoproteins—in the capsid determine the **specificity** of interaction of a virus with its host cell.
The capsid protects the nucleic acid and facilitates attachment and penetration of the host cell by the virus. Inside the cell, viral nucleic acid redirects the host's enzymatic machinery to functions associated with replication of the virus.

2- Prokaryotes

The primary distinguishing characteristics of the prokaryotes are their relatively small size, usually on the order of 1 μm in diameter, and the absence of a nuclear membrane. The DNA of almost all bacteria is a circle with a length of about 1 mm; this is the prokaryotic chromosome. Most prokaryotes have only a single chromosome. The specialized region of the cell containing DNA is termed the **nucleoid.**

-Bacteria & Archaebacteria: The Major Subdivisions within the Prokaryotes
A major success in molecular phylogeny has been the demonstration that prokaryotes fall into two major groups. Most investigations have been directed to one group, the **bacteria**. The other group, the **archaebacteria**, has received relatively little attention until recently, in part because many of its representatives are difficult to study in the laboratory. Some archaebacteria, for example, are killed by contact with oxygen, and others grow at temperatures exceeding that of boiling water. Before molecular evidence became available, the major subgroupings of archaebacteria seemed disparate. The **methanogens** carry out an anaerobic respiration that gives rise to methane; the **halophiles** demand extremely high salt concentrations for growth; and the **thermoacidophiles** require high temperature and acidity. It has now been established that these prokaryotes share biochemical traits such as cell wall or membrane components that set the group entirely apart from all other living organisms. An intriguing trait shared by **archaebacteria** and **eukaryotes** is the presence of **introns** within genes. The function of introns—segments of DNA that interrupts informational DNA within genes—is not established. What is known is that introns represent a fundamental characteristic shared by the DNA of archaebacteria and eukaryotes. This common trait has led to the suggestion that—just as mitochondria and chloroplasts appear to be evolutionary derivatives of the bacteria the eukaryotic nucleus may have arisen from an archaebacterial ancestor.

3- Eukaryotes: are members of the four following major groups: **algae**, **protozoa**, **fungi**, and **slime molds**. It should be noted that these groupings are not necessarily phylogenetic: Closely related organisms may have been categorized separately because underlying biochemical and genetic similarities may not have been recognized.

-Algae

The term "algae" has long been used to denote all organisms that produce O2 as a product of photosynthesis. One major subgroup of these organisms the blue green bacteria, or cyanobacteria are prokaryotic and no longer are termed algae. This classification is reserved exclusively for photosynthetic eukaryotic organisms. All algae contain chlorophyll in the photosynthetic membrane of their subcellular chloroplast. Many algal species are unicellular microorganisms. Other algae may form extremely large multicellular structures. Kelps of brown algae sometimes are several hundred meters in length.

-Protozoa

Protozoa are unicellular nonphotosynthetic protists. The most primitive protozoa appear to be flagellated forms that in many respects resemble representatives of the algae. It seems likely that the ancestors of these protozoa were algae that became **heterotrophs**—the nutritional requirements of such organisms are met by organic compounds. Adaptation to a heterotrophic mode of life was sometimes accompanied by loss of chloroplasts, and algae thus gave rise to the closely related protozoa. Similar events have been observed in the laboratory to be the result of either mutation or physiologic adaptation. From **flagellated** protozoa appear to have evolved the **ameboid** and the **ciliated** types; intermediate forms are known that have flagella at one stage in the life cycle and pseudopodia (characteristic of the ameba) at another stage. A fourth major group of protozoa, the **sporozoa**, are strict parasites that are usually immobile; most of which reproduce sexually and asexually in alternate generations by means of spores.

-Fungi:

The fungi are nonphotosynthetic protists growing as a mass of branching, interlacing filaments ("**hyphae**") known as a **mycelium.** Although the hyphae exhibit cross-walls, the cross-walls are perforated and allow free passage of nuclei and cytoplasm. The entire organism is thus a **coenocyte** (a multinucleated mass of continuous cytoplasm) confined within a series of branching tubes. These tubes, made of polysaccharides such as chitin, are homologous with cell walls. The mycelial forms are called **molds;** a few types, **yeasts,** do not form a mycelium but are easily recognized as fungi by the nature of their sexual reproductive processes and by the presence of transitional forms.

BACTERIAL STRUCTURES

Despite their lack of complexity compared to eukaryotes, a number of eubacterial structures may be defined. Not all bacteria possess all of these components.

The bacterial cell has three architectural regions:

1- **Cell envelope** consisting of a **capsule**, **cell wall** and **plasma membrane**.
2- **Cytoplasmic region** that contains the cell **chromosome** (**DNA**) and **ribosomes** and various sorts of **inclusions.**
3- **Appendages** (attachments to the cell surface) in the form of **flagella** and **pili (or fimbriae)**.

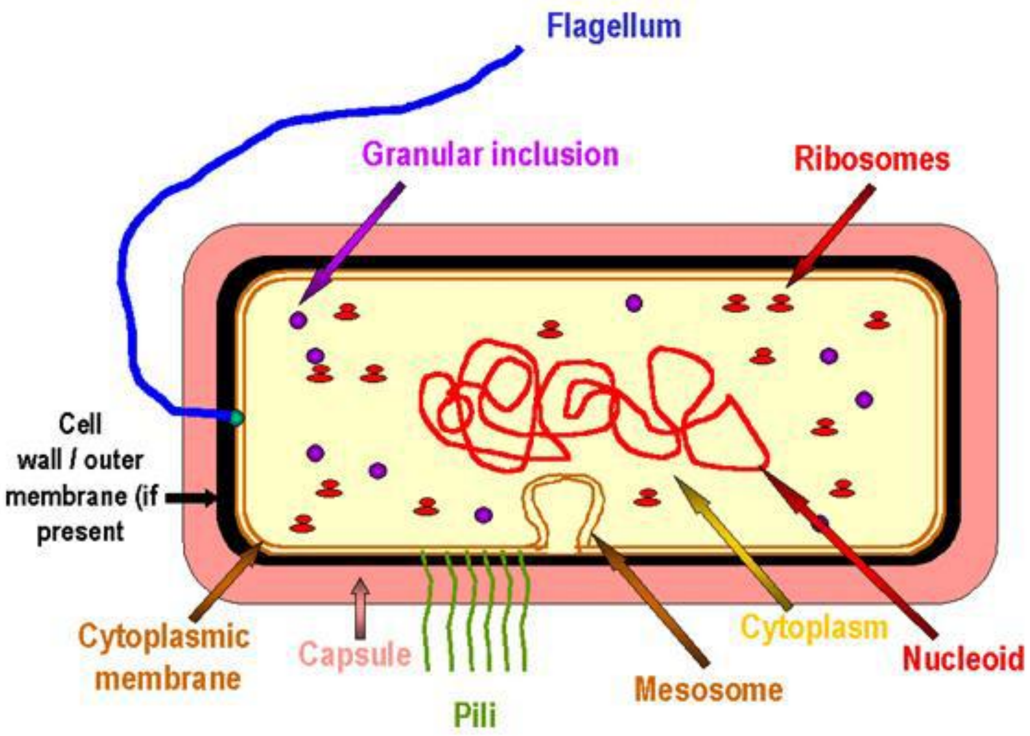


Figure. The prototype bacterial cell

***Cell envelope:-**

The **cell envelope** is a descriptive term for the several layers of material that envelope or enclose the protoplasm of the cell. The cell protoplasm (**cytoplasm**) is surrounded by the **plasma membrane**, a **cell wall** and a **capsule**. The cell wall itself is a layered structure in Gram-negative bacteria. All cells have a plasma membrane . Almost all prokaryotes have a cell wall to prevent damage to the underlying protoplast. Outside the cell wall, foremost as a surface structure, may be a polysaccharide **capsule** or **glycocalyx**.

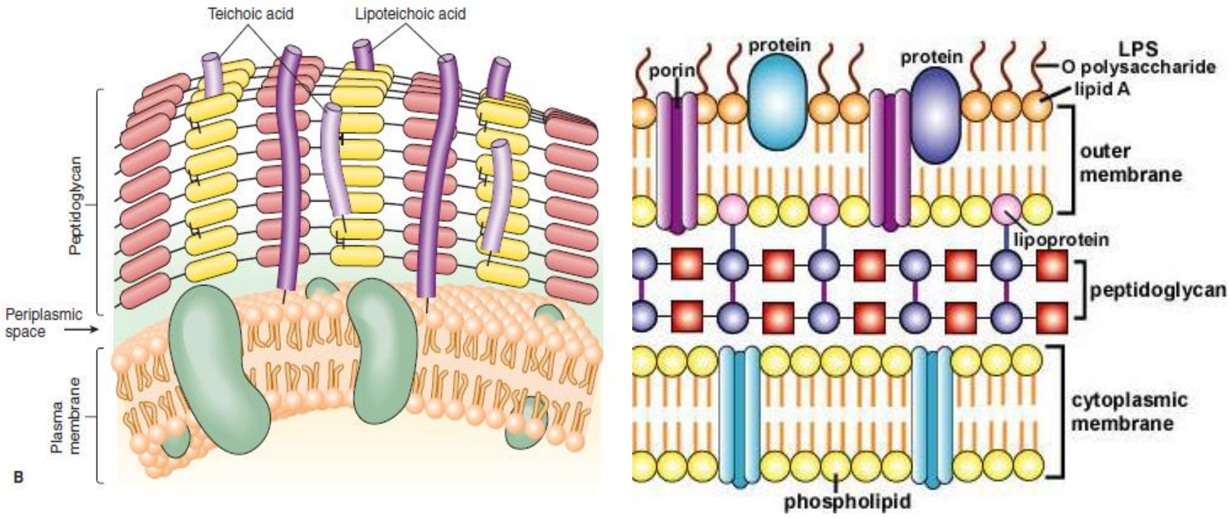
1-Capsules:- Most bacteria contain some sort of a polysaccharide layer outside of the cell wall polymer. In a general sense, this layer is called a capsule. A true capsule is a discrete detectable layer of polysaccharides deposited outside the cell wall. A less discrete structure or matrix which embeds the cells is a called a slime layer or a **glycocalyx** is a very thin layer of tangled polysaccharide fibres on the cell surface, often mediate adherence of cells to surfaces. Capsules have several functions and often have multiple functions. Capsules mediate **adherence** of cells to surfaces, **protect** bacterial cells from engulfment by phagocytes and invasiveness of pathogen.

2-Cell wall:- Most prokaryotes have a rigid cell wall. The cell wall is an essential structure that **protects** the cell protoplast from mechanical damage and from osmotic rupture or lysis. Bacterial **murein** is a unique type of peptidoglycan. Peptidoglycan is a polymer of sugars (a glycan) cross-linked by short chains of amino acids (peptide). A layer of peptidoglycan consist of two types of alternating joined molecules called **N-acetylmuramic acid**, and **N-acetylglucose amine** and asset of identical tetrapeptide side chain. The glycan backbone of the peptidoglycan molecule can be cleaved by an enzyme called **lysozyme** that is present in animal serum, tissues and secretions, and in phagocyte granules. The **function** of lysozyme is to lyse (rupture) bacterial cells as a defense against bacterial pathogens.

Bacteria are classified as **gram positive** and **gram negative** according to the response to the gram staining procedure. In Gram-positive Bacteria the cell wall is thick (15-80 nanometers), consisting of several layers of peptidoglycan. It have a layer of **teichoic acids** on the outside of the peptidoglycan which are unique to the Gram-positive cell wall.

In the Gram-negative Bacteria the cell wall is relatively thin (10 nanometers) and is composed of a single layer of peptidoglycan surrounded by a membranous structure called the **outer membrane**. The outer membrane of Gram-negative bacteria invariably contains a unique component, l**ipopolysaccharide** (LPS or endotoxin), which is toxic to animals, **lipoprotein**, and **phospholipid**. Lying between the outer membrane layer and the cytoplasmic membrane is the **periplasmic space** which is the site in some species of enzymes (ex. beta lactamases) that degrade penicillin and other beta lactam drugs.

Bacterial lipopolysaccharides are toxic. When injected in small amounts LPS or endotoxin activates several host responses that lead to fever, inflammation and shock.



Gram + Gram -

Figure. Comparison of bacterial cell wall of Gram positive and Gram negative.

3- Plasma membrane:-

The **plasma membrane**, also called the **cytoplasmic membrane**, is the most dynamic structure of a prokaryotic cell. Its main function is a s a selective permeability barrier that regulates the passage of substances into and out of the cell. Bacterial membranes are composed of 40 % phospholipid and 60 % protein.

Functions of plasma membrane:-

1. Osmotic or permeability barrier

2. Location of transport systems for specific solutes (nutrients and ions)

3. Energy generating functions, involving respiratory and photosynthetic electron transport systems, establishment of proton motive force, and transmembranous, ATP-synthesizing ATPase

4. Synthesis of membrane lipids (including lipopolysaccharide in Gram-negative cells)

5. Synthesis of murein (cell wall peptidoglycan)

6. Assembly and secretion of extracytoplasmic proteins

7. Location of specialized enzyme system

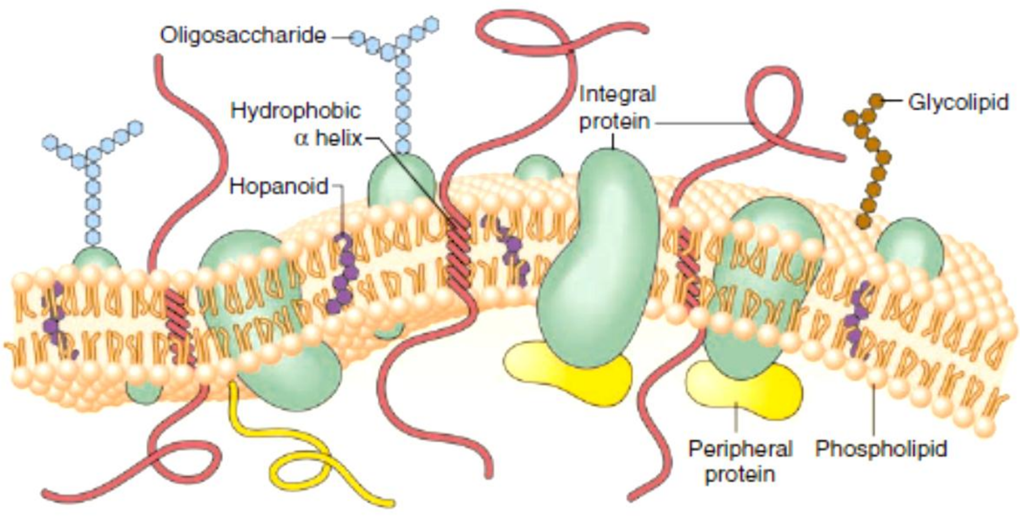


Figure : Bacterial plasma membrane structure.

Wall-less forms of Bacteria

When bacteria are treated with enzymes that are lytic for the cell wall (e.g. lysozyme or antibiotics that interfere with biosynthesis of peptidoglycan), wall-less bacteria are often produced. Usually these treatments generate non-viable organisms. Wall-less bacteria that can not replicate are referred to as **spheroplasts** (when an outer membrane is present) or **protoplasts** (if an outer membrane is not present). Occasionally wall-less bacteria that can replicate are generated by these treatments (L forms).

* **Cytoplasmic Region**:

The cytoplasmic constituents of bacterial cells invariably include the prokaryotic chromosome (nucleoid), ribosomes, and several hundred proteins and enzymes.

1- Chromosome:-

The chromosome is typically one large circular molecule of DNA, more or less free in the cytoplasm. The cell chromosome is the genetic control center of the cell which determines all the properties and functions of the bacterium.

2- Plasmids:-

The plasmids are extrachromosomal pieces of DNA double strand, circular DNA. Plasmid are not essential for the life of cell. They may confer certain properties like toxigenicity, virulence and drug resistance.

3- Ribosome:-

Ribosomes are involved in the process of translation (protein synthesis), The ribosomes of prokaryotes are smaller than cytoplasmic ribosomes of eukaryotes. Prokaryotic ribosomes are 70S in size, being composed of 30S and 50S subunits. The 80S ribosomes of eukaryotes are made up of 40S and 60S subunits. ribosomes the site of action of many antibiotics that inhibit bacterial but not human protein synthesis. It consist from **protein** and **RNA**.

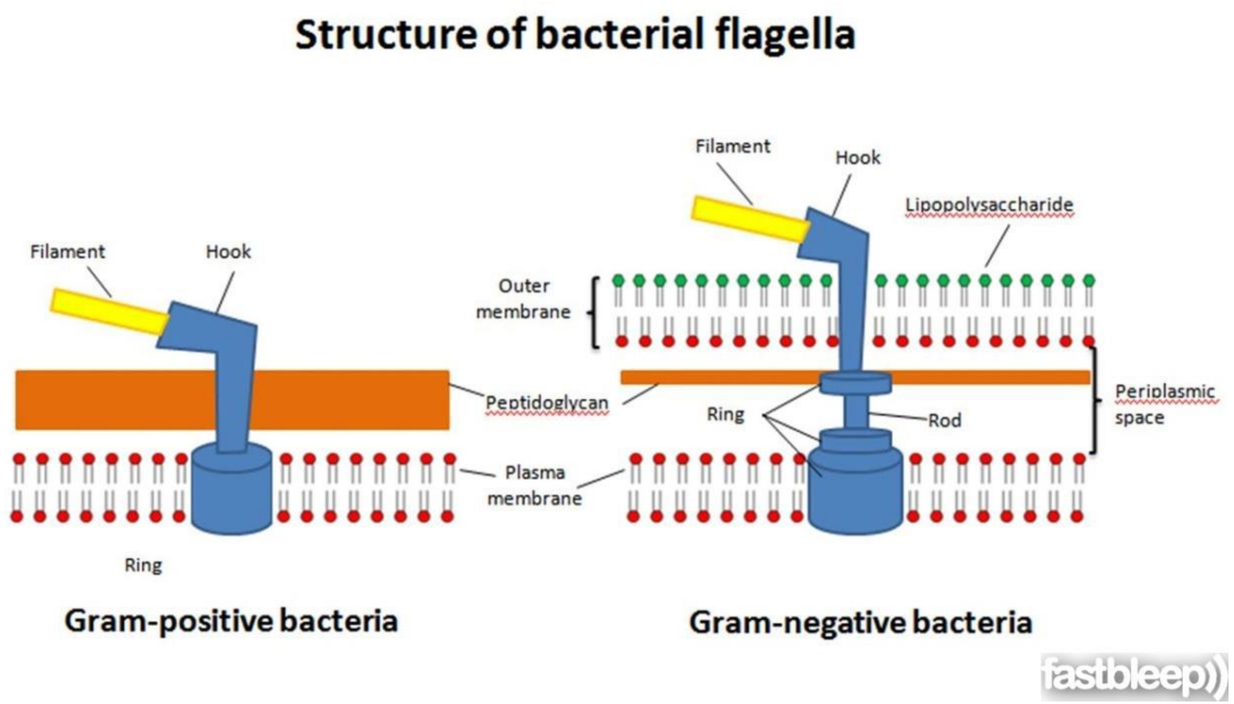
4- Inclusions:-

Inclusions are distinct granules that may occupy a substantial part of the cytoplasm. Inclusion granules are usually reserve materials of some sort. For example, carbon and energy reserves may be stored as glycogen.

*__Appendages__

1- Flagella:-

Flagella are filamentous protein structures attached to the cell surface that provide the swimming movement for most motile prokaryotes. The flagellar filament is rotated by a motor apparatus in the plasma membrane allowing the cell to swim in fluid environments. The flagellar apparatus consists of several distinct proteins: a system of rings imbedded in the cell envelope (**the basal body),** a **hook-like structure** near the cell surface, and the **flagellar filament** composed of polymerized protein called **flagellin.**



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Some bacteria has single flagella called **monotrichus** or has flagellum at each end of the cell called **amphitrichus**, or multiple flagella at one end called **lophotrichus** or flagella distributed over the entire body called **peritrichus.**

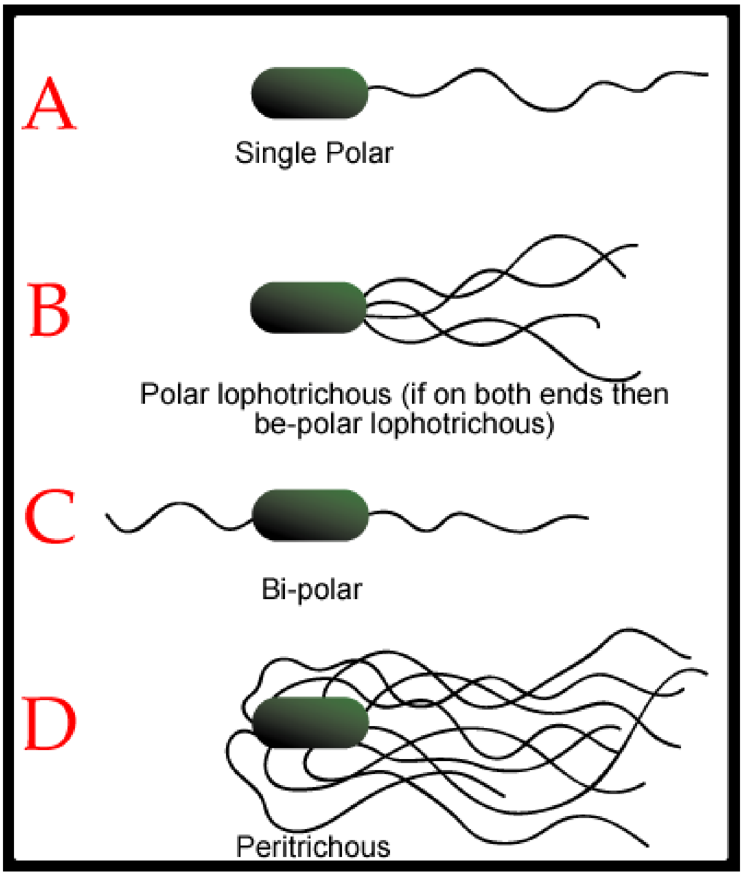

Figure. Types of flagella .

2-Pili (synonym: fimbriae)

The Pili are hair-like projections of the cell. Some are involved in sexual conjugation and others allow adhesion to host epithelial surfaces in infection.

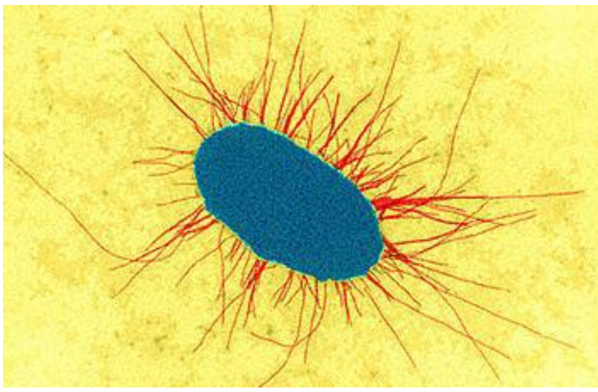

Figure. E. coli with fimbriae.

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-Endospores (spores)

These are a dormant form of a bacterial cell produced by certain bacteria when starved; the actively growing form of the cell is referred to as vegetative. The spore is resistant to adverse conditions (including high temperatures and organic solvents). The spore cytoplasm is dehydrated and contains **calcium dipicolinate (dipicolinic acid).**

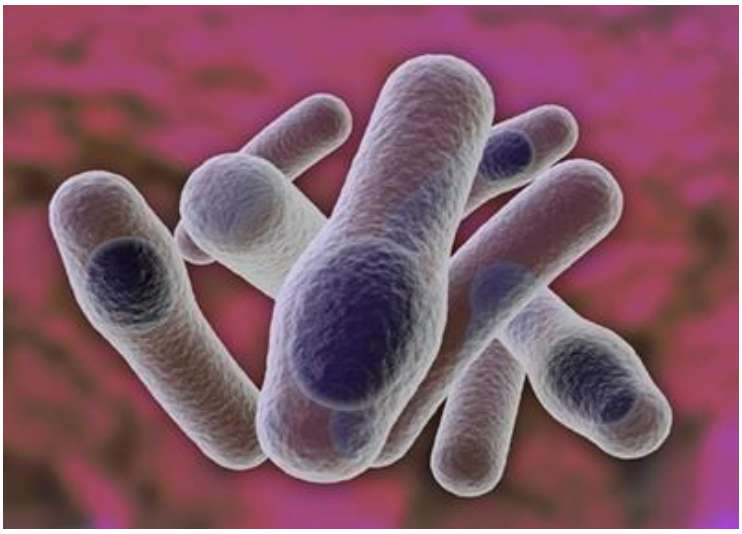

Figure. Endospores.