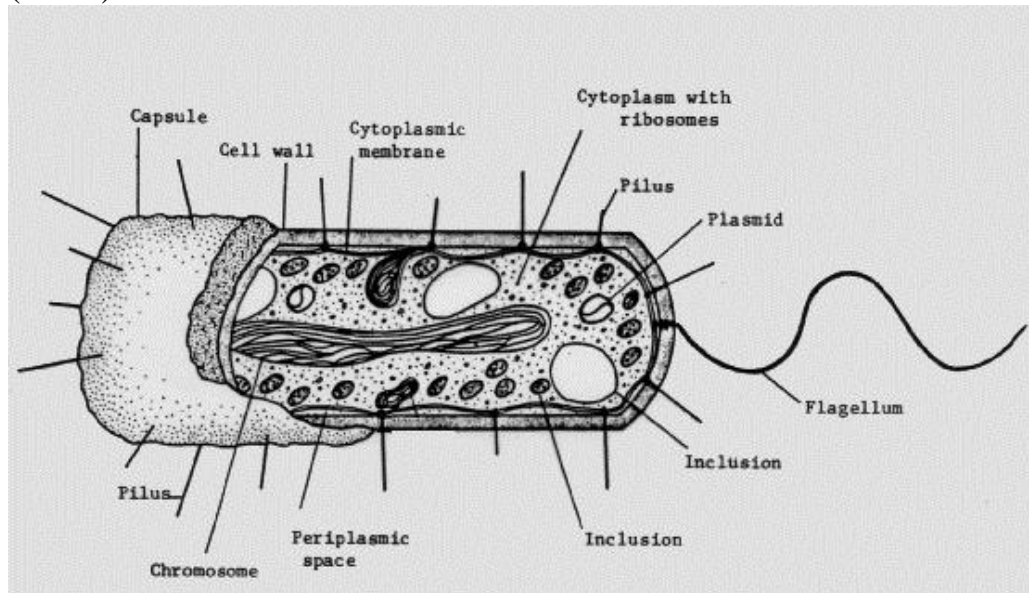


Prokaryotic Cell Architecture(bacteria)

Structurally, a bacterial cell (Figure below) has three architectural regions: **appendages** (attachments to the cell surface) in the form of **flagella** and **pili** (or **fimbriae**); a **cell envelope** consisting of a **capsule**, **cell wall** and **plasma membrane**; and a **cytoplasmic region** that contains the cell **chromosome** (DNA) and **ribosomes** and various sorts of **inclusions**.



Schematic drawing of a typical bacterial cell.

Appendages

Flagella

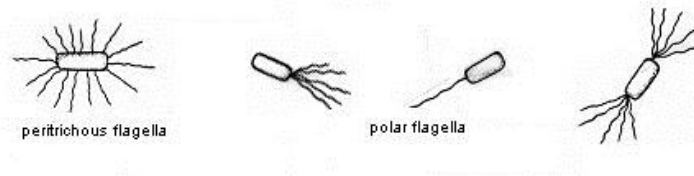
Flagella are filamentous protein structures attached to the cell surface that provide the swimming movement for most motile prokaryotes. The diameter of a prokaryotic flagellum is about 20 nanometers, well-below the resolving power of the light microscope. The flagellar filament is rotated by a motor apparatus in the plasma membrane allowing the cell to swim in fluid environments. Bacterial flagella are powered by proton motive force (chemiosmotic potential) established on the bacterial membrane, rather than ATP hydrolysis which powers eukaryotic flagella. About half of the bacilli and all of the spiral and curved bacteria are motile by means of flagella. Very few cocci are motile, which reflects their adaptation to dry environments and their lack of hydrodynamic design.



Salmonella enterica. . The enteric are motile by means of peritrichous flagella.

Flagella may be variously distributed over the surface of bacterial cells in distinguishing patterns, but basically flagella are either **polar** (one or more

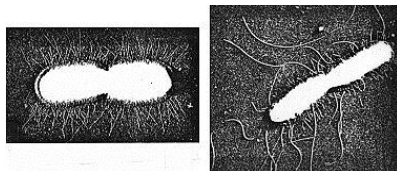
flagella arising from one or both poles of the cell) or **peritrichous** (lateral flagella distributed over the entire cell surface). Flagellar distribution is a genetically-distinct trait that is occasionally used to characterize or distinguish bacteria. For example, among Gram-negative rods, pseudomonads have polar flagella to distinguish them from enteric bacteria, which have peritrichous flagella.



Fimbriae and Pili

Fimbriae and **Pili** are interchangeable terms used to designate short, hair-like structures on the surfaces of prokaryotic cells. Like flagella, they are composed of protein. Fimbriae are shorter and stiffer than flagella, and slightly smaller in diameter. Generally, fimbriae have nothing to do with bacterial movement (there are exceptions, e.g. twitching movement on *Pseudomonas*). There are two types of Pili:

Common Pili (often called fimbriae) are usually involved in specific adherence (attachment) of prokaryotes to surfaces in nature. In medical situations, they are major determinants of bacterial virulence. The **F or sex pilus**, mediates DNA transfer during conjugation and apparently stabilizes mating bacteria during the process of conjugation.



Gram-positive wall is a uniformly thick layer external to the plasma membrane. It is composed mainly of peptidoglycan (murein). The Gram-negative wall appears thin and multilayered. It consists of a relatively thin peptidoglycan sheet between the plasma membrane and a phospholipid-lipopolysaccharide outer membrane. The space between the inner (plasma) and outer membranes (wherein the peptidoglycan resides) is called the periplasm.

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 called a **slime layer** or a **biofilm**. A type of capsule found in bacteria called a **glycocalyx** or **microcapsule** is a very thin layer of tangled polysaccharide fibers on the cell surface. Capsules have **several functions** and often have multiple functions in a particular organism. Like fimbriae, capsules, slime layers, and glycocalyx often

mediate adherence of cells to surfaces. Capsules also **protect bacterial cells from engulfment** by predatory protozoa or white blood cells (phagocytes), or from attack by antimicrobial agents of plant or animal origin. Capsules in certain soil bacteria **protect cells from perennial effects of drying** or desiccation. Capsular materials (e.g. dextrans) may be overproduced when bacteria are fed sugars to become **reserves of carbohydrate** for subsequent metabolism. A classic example of biofilm construction in nature is the formation of **dental plaque** mediated by the oral bacterium, *Streptococcus mutans*. The bacteria adhere specifically to the pellicle of the tooth by means of a protein on the cell surface. The bacteria grow and synthesize a dextran capsule which binds them to the enamel and forms a biofilm some 300-500 cells in thickness. The bacteria are able to cleave sucrose (provided by the animal diet) into glucose plus fructose. The fructose is fermented as an energy source for bacterial growth. The glucose is polymerized into an extracellular dextran polymer that cements the bacteria to tooth enamel and becomes the matrix of dental plaque. The dextran slime can be depolymerized to glucose for use as a carbon source, resulting in production of lactic acid within the biofilm (plaque) that decalcifies the enamel and leads to dental caries or bacterial infection of the tooth.

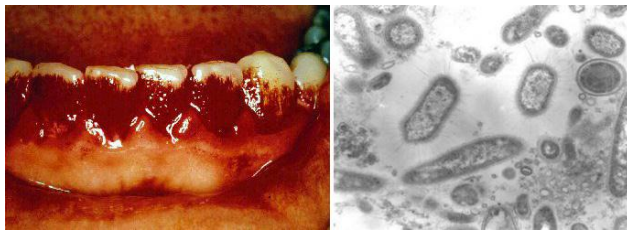


Figure 16 (Left) Dental plaque revealed by a harmless red dye.

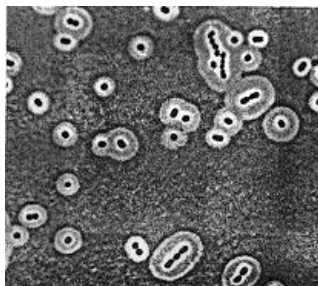


Figure 13. Bacterial capsules outlined by India ink viewed by light microscopy.

The 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, which is the essential and definitive

characteristic of a "cell". 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**.

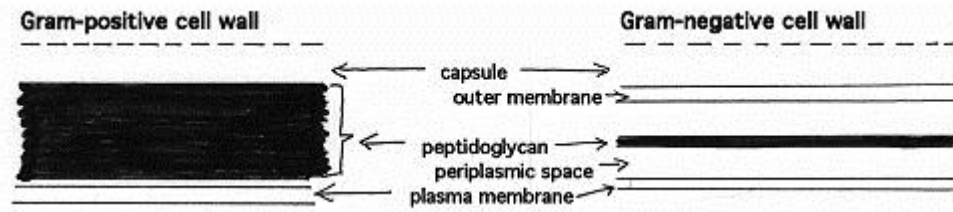


Figure 12. Profiles of the cell envelope the Gram-positive and Gram-negative bacteria.

Cell Wall

Most prokaryotes have a rigid **cell wall**. The cell wall is an essential structure that protects the cell **protoplast** (the region bound by and including the membrane) from mechanical damage and from osmotic rupture or **lysis**. Bacteria usually live in relatively dilute environments such that the accumulation of solutes inside the cell cytoplasm greatly exceeds the total solute concentration in the outside environment. Thus, the osmotic pressure against the inside of the plasma membrane may be the equivalent of 10-25 atmospheres. Since the membrane is a delicate, plastic structure, it must be restrained by an outside wall made of porous, rigid material that has high tensile strength. Such a material is **murein**, the ubiquitous component of bacterial cell walls.

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). All bacterial peptidoglycan contain **N-acetylmuramic acid**, which is the definitive component of murein. The profiles of the cell surface of bacteria, as seen with the electron microscope, are drawn in Figure 12.

In **Gram-positive Bacteria** (those that retain the purple crystal violet dye when subjected to the Gram-staining procedure) the cell wall is thick (15-80 nanometers), consisting of several layers of peptidoglycan. Running perpendicular to the peptidoglycan sheets are a group of molecules called **teichoic acids** which are unique to the Gram-positive cell wall. **Peptidoglycan** is a polysaccharide consisting of alternating amino sugars (**N-acetylglucosamine (NAG)** and **N-acetyl muramic acid (NAM)**) linked by Beta 1-4 bonds like those in cellulose. The Beta 1-4 bond can be broken by an enzyme called lysozyme that is present in your tears, saliva, blood, other bodily fluids and egg white. These polysaccharide chains are held together by peptide cross-links. These peptides that they may contain **D-amino acids**.

Antibiotics like penicillin and cephalosporin inhibit the enzymes that synthesise peptidoglycan. This is why these antibiotics are not effective against the eucaryotic cells of fungi and parasites.

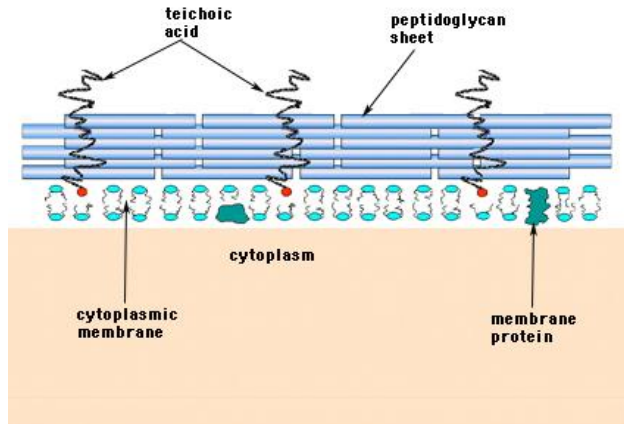


Figure 17. Structure of the Gram-positive bacterial cell wall. The wall is relatively thick and consists of many layers of peptidoglycan interspersed with teichoic acids that run perpendicular to the peptidoglycan sheets.

In the **Gram-negative Bacteria** (which do not retain the crystal violet in the Gram-stain procedure) 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, **lipopolysaccharide (LPS or endotoxin)**, which is toxic to animals. In Gram-negative bacteria the outer membrane is usually considered as part of the cell wall.

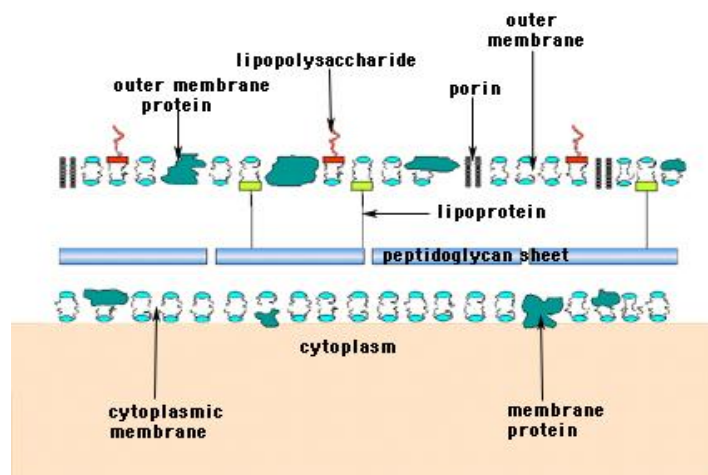


Figure 18. Structure of the Gram-negative cell wall. The wall is relatively thin and contains much less peptidoglycan than the Gram-positive wall. Also, teichoic acids are absent. However, the Gram negative cell wall consists of an outer membrane that is outside of the peptidoglycan layer. The outer membrane is attached to the peptidoglycan sheet by a unique group of lipoprotein molecules.

Of special interest as a component of the Gram-negative cell wall is the **outer membrane**, a discrete bilayered structure on the outside of the peptidoglycan

sheet (see Figure 12 above and Figure 19 below). For the bacterium, the outer membrane is first and foremost a permeability barrier, but primarily due to its lipopolysaccharide content, it possesses many interesting and important characteristics of Gram-negative bacteria. The outer membrane superficially resembles the plasma membrane except the outer face contains a unique type of Lipopolysaccharide referred to by medical microbiologists as **endotoxin** because of its toxic effects in animals.

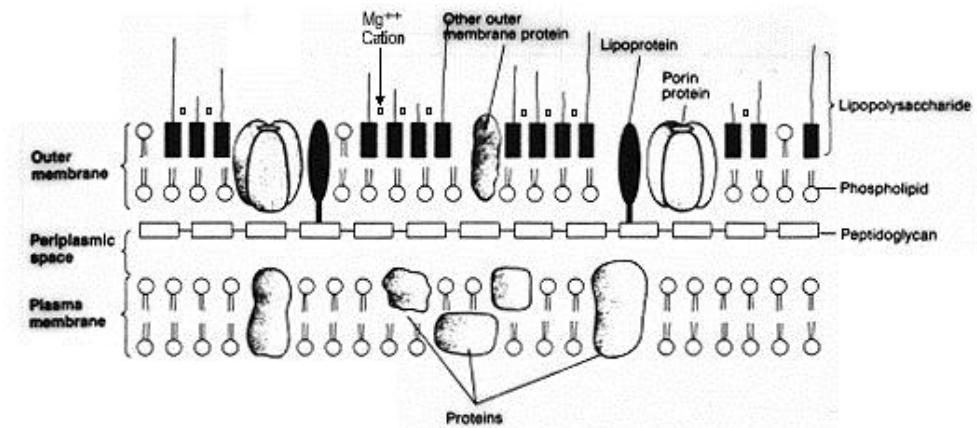


Figure 23. Schematic illustration of the outer membrane, cell wall, and plasma membrane of a Gram-negative bacterium. Lipopolysaccharide (LPS or endotoxin) is located on the outer face of the outer membrane.

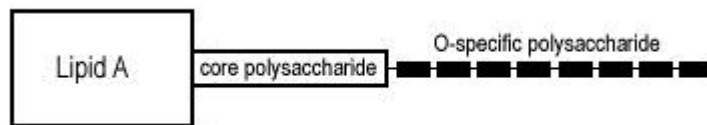


Figure 24. Structure of bacterial lipopolysaccharide or endotoxin. The lipo- part (Lipid A) is the toxic portion of the molecule. It causes fever, inflammation, hemorrhage and shock in animals. The -polysaccharide part of the molecule is responsible for antigenic properties of the bacterium, which influences how the animal immune system will respond.

Endotoxins may play a role in infection by any Gram-negative bacterium. The toxic component of endotoxin (LPS) is Lipid A. The O-specific polysaccharide may provide for adherence or resistance to phagocytosis, in the same manner as fimbriae and capsules. The O polysaccharide (also referred to as the O antigen) also accounts for multiple antigenic types (serotypes) among Gram-negative bacterial pathogens.

The Gram stain and bacterial cell walls

A correlation between Gram stain reaction and cell wall properties of bacteria is summarized in Table 5. The Gram stain procedure contains a "destaining" step wherein the cells are washed with an acetone-alcohol mixture. The lipid content of the Gram-negative wall probably affects the outcome of this step so that

Gram-positive cells retain a primary stain while Gram-negative cells are destained.

Table shows Correlation of the Grams stain with cell wall properties of Bacteria.

Property	Gram-positive	Gram-negative
Thickness of wall	thick (20-80 nm)	thin (10 nm)
Number of layers	1	2
Peptidoglycan (murein) content	>50%	10-20%
Teichoic acids in wall	present	absent
Lipid and lipoprotein content	0-3%	58%
Protein content	0	9%
Lipopolysaccharide content	0	13%

Cell Wall-less Forms

A few bacteria are able to live or exist without a cell wall. The **mycoplasmas** are a group of bacteria that lack a cell wall. Mycoplasmas have sterol-like molecules incorporated into their membranes and they are usually inhabitants of osmotically-protected environments (contain a high concentration of external solute).

The Cytoplasmic Membrane

The **cytoplasmic membrane** of bacterial cells is a delicate and plastic structure that completely encloses the cell cytoplasm (or protoplasm). The bacterial membrane is composed of 40 percent phospholipid and 60 percent protein. The phospholipids are amphoteric molecules, meaning they have a water-soluble hydrophilic region (the glycerol "head") attached to two insoluble hydrophobic fatty acid "tails". In water, such molecules naturally form the molecular bilayer characteristic of membranes. The fatty acid tails from one layer face towards the fatty acid tails of the second layer ("likes dissolve like"), and the glycerol heads naturally turn towards the water (Figure 25 and 26). Dispersed throughout the bilayer are various structural and enzymatic proteins which carry out most membrane functions. This arrangement of proteins and phospholipids forms what is called the **fluid mosaic membrane** as illustrated in Figure 27.

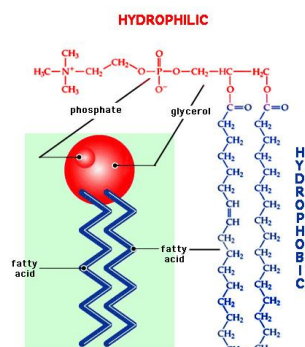


Figure 25. Molecular structure of a phospholipid, the building block of membranes. Inset - the usual depiction of a membrane phospholipid containing a phosphatidyl-glycerol "head" attached to two fatty acid "tails".

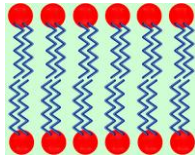


Figure 26. Organization of phospholipids in aqueous solution to form a bilayer. The hydrophilic phosphatidyl glycerols form the inner and outer faces of the membrane. The fatty acids orient themselves towards one another to form the hydrophobic interior of the membrane.

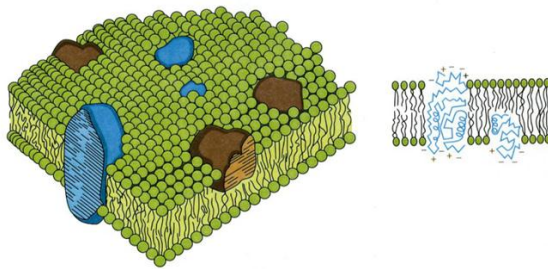


Figure 27. Fluid mosaic model of a biological membrane. In aqueous environments membrane phospholipids arrange themselves in such a way that they spontaneously form a fluid bilayer. Membrane protein may be either structural or functional. Proteins may be permanently or transiently associated with one side or the other of the membrane or built into the bilayer, or they may span the bilayer forming transport channels through the membrane.

Functions of the Cytoplasmic Membrane

Permeability Barrier

The cell membrane is the most dynamic structure in the cell. Its main function is as a **permeability barrier** that regulates the passage of substances into and out of the cell. The plasma membrane is the definitive structure of a cell since it sequesters the molecules of life in the cytoplasm, separating it from the outside environment. The bacterial membrane freely allows passage of water and a few small uncharged molecules (less than molecular weight of 100 daltons), but it does not allow passage of larger molecules or any charged substances except when monitored by proteins in the membrane called **transport systems**.

Transport of Solutes

The presence of **transport systems** in the membranes allows the bacteria to accumulate solutes and chemical precursors of cell material inside their cytoplasm at concentrations which greatly exceed the concentrations in the environment. Remember, most bacteria live in relatively dilute environments (e.g. a lake or stream) where the concentration of the business molecules of life is greater inside of the cell than in the environment. Hence, the bacterial cells must transport their nutrients from the environment and maintain a higher concentration of solutes inside the cell than outside the cell. This comes at a price. To concentrate a substance against the environmental gradient using a membrane transport system always costs energy in one form or another.

Bacteria have a variety of types of transport systems which can be used alternatively in various environmental situations. The most important transport systems are called **active transport systems** since they require energy and concentrate substances inside of the cell. At least 80 percent of the molecules needed in the cytoplasm are taken up by the process of **active transport**. Active transport systems are mediated by proteins in the membrane called **carrier proteins** or "permeases" that are generally quite specific for the substances that they will transport. All active transport systems require energy to operate. Some use chemical energy derived from ATP; others use **proton motive force (pmf)**, which is derived from the establishment of a charge and a pH gradient on opposite sides of the membrane.

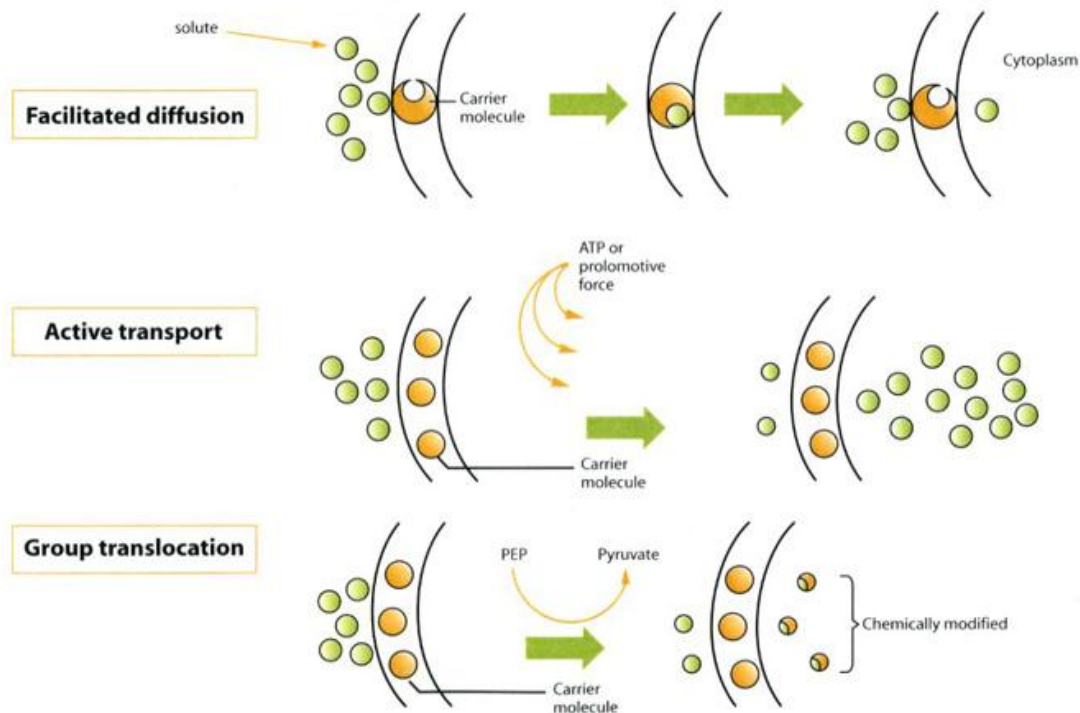


Figure 28. Operation of bacterial transport systems. Bacterial transport systems are operated by membrane proteins, also called carriers or permeases. Facilitated diffusion is a carrier-mediated system that does not require energy and does not concentrate solutes against a gradient. Active transport systems use energy and are able to concentrate molecules against a concentration gradient. Group translocation systems also use chemical energy during transport but they are distinguished from active transport because they modify the solute during its passage across the membrane. Most solutes in bacteria are transported by active transport systems.

Besides **transport proteins** that selectively mediate the passage of substances into and out of the cell, bacterial membranes may also contain **sensing proteins** that measure concentrations of molecules in the environment or **binding proteins** that translocate signals from the environment to genetic and metabolic machinery in the cytoplasm.

Generation of Energy

Unlike eukaryotes, bacteria don't have intracellular organelles for energy producing processes such as respiration or photosynthesis. Instead, the

cytoplasmic membrane carries out these functions. The membrane is the location of **electron transport systems (ETS)** used to produce energy during photosynthesis and respiration, and it is the location of an enzyme called **ATP synthetase (ATPase)** which is used to synthesize ATP.

When the electron transport system operates, it establishes a pH gradient across of the membrane due to an accumulation of protons (H^+) outside and hydroxyl ion (OH^-) inside. Thus the outside is acidic and the inside is alkaline. Operation of the ETS also establishes a charge on the membrane called **proton motive force (pmf)**. The outer face of the membrane becomes charged positive while inner face is charged negative, so the membrane has a positive side and a negative side, like a battery. The pmf can be used to do various types of work including the rotation of the flagellum, or active transport as described above. The pmf can also be used to make ATP by the membrane ATPase enzyme which consumes protons when it synthesizes ATP from ADP and phosphate. The connection between electron transport, establishment of pmf, and ATP synthesis during respiration is known as **oxidative phosphorylation**; during photosynthesis, it is called **phosphorylation**.

In bacteria, the **photosynthetic pigments** that harvest light energy for conversion into chemical energy are also located in the membrane. Membranes may contain other **enzymes** involved in many metabolic processes such as cell wall synthesis, septum formation, membrane synthesis, DNA replication, CO_2 fixation and ammonia oxidation. The predominant functions of bacterial membranes are listed in the table below.

Table 6. Functions of the prokaryotic 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 ATP-synthesizing ATPase
4. Synthesis of membrane lipids (including lipopolysaccharide in Gram-negative cells)
5. Synthesis of murein (cell wall peptidoglycan)
6. Coordination of DNA replication and segregation with septum formation and cell division
7. Location of specialized enzyme systems, such as for CO_2 fixation, nitrogen fixation

The Cytoplasm The cytoplasm of bacterial cells consists of an aqueous solution of three groups of molecules: macromolecules such as proteins (enzymes), DNA, mRNA and tRNA; small molecules that are energy sources,

precursors of macromolecules, metabolites or vitamins , various inorganic ions and cofactors . The cytoplasm of prokaryotes is more gel-like than that of eukaryotes and the processes of cytoplasmic streaming, which are evident in eukaryotes, do not occur.

The cytoplasmic constituents of bacterial cells invariably include the **prokaryotic chromosome** (nucleoid), **ribosomes**, and several hundred **proteins and enzymes**. The chromosome is typically one large circular molecule of **DNA**, more or less free in the cytoplasm. Prokaryotes sometimes possess smaller extra chromosomal pieces of DNA called **plasmids**. The total DNA content of a prokaryote is referred to as the cell **genome**. The cell chromosome is the genetic control center of the cell which determines all the properties and functions of the bacterium. During cell growth and division, the prokaryotic chromosome is replicated in to make an exact copy of the molecule for distribution to progeny cells.

The distinct granular appearance of prokaryotic cytoplasm is due to the presence and distribution of **ribosomes**. The ribosomes of prokaryotes are smaller than cytoplasmic ribosomes of eukaryotes. Prokaryotic ribosomes are 70S in size, being composed of 30S and 50S subunits. Ribosomes are involved in the process of translation (protein synthesis)

Inclusions

Often contained in the cytoplasm of prokaryotic cells is one or another of some type of inclusion granule. **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 (a polymer of glucose) or as polybetahydroxybutyric acid (a type of fat) granules.....

A bacterial structure sometimes observed as an inclusion is actually a type of dormant cell called an **endospore**. Endospores are formed by a few groups of **Bacteria** as intracellular structures, but ultimately they are released as free endospores. Biologically, endospores are a fascinating type of cell. Endospores exhibit no signs of life, being described as **cryptobiotic**. They are highly resistant to environmental stresses such as high temperature (some endospores can be boiled for hours and retain their viability), irradiation, strong acids, disinfectants, etc. They are probably the most durable cell produced in nature. Although cryptobiotic, they retain viability indefinitely such that under appropriate environmental conditions, they germinate back into vegetative cells. Endospores are formed by vegetative cells in response to environmental signals that indicate a limiting factor for vegetative growth, such as exhaustion of an essential nutrient. They germinate and become vegetative cells when the

environmental stress is relieved. Hence, endospore-formation is a mechanism of survival rather than a mechanism of reproduction.

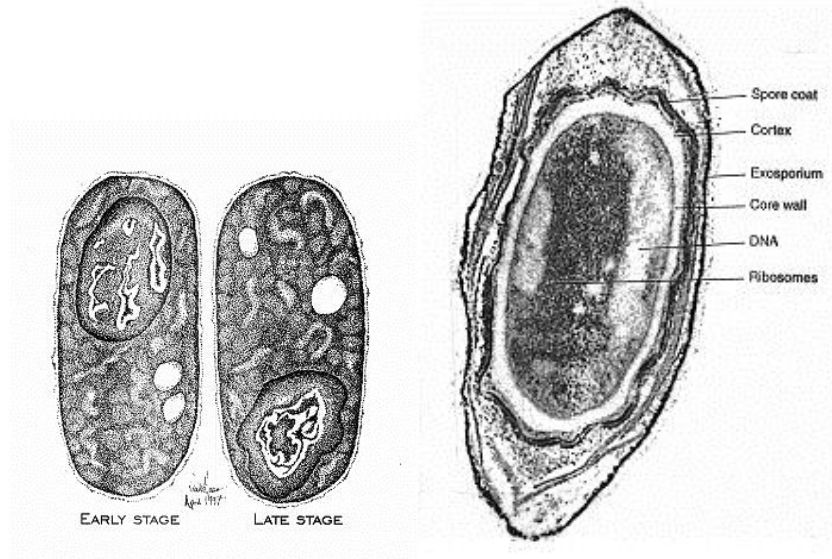


Figure 33. Early and late stages of endospore formation. Electron micrograph of a bacterial endospore

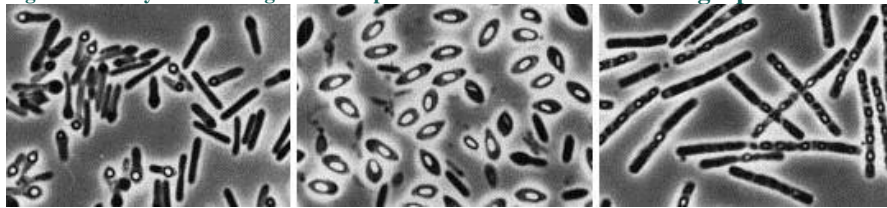


Figure 34. Bacterial endospores. Phase microscopy of sporulating bacteria demonstrates the refractility of endospores, as well as characteristic spore shapes and locations within the mother cell.

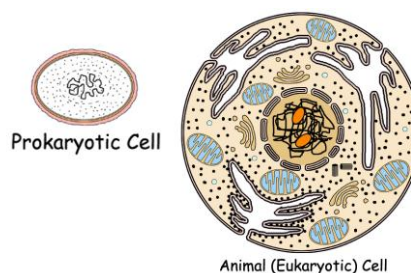
Summary to remember: Table shows Characteristics of typical bacterial cell structures.

Structure	Function(s)	Predominant chemical composition
Flagella	Swimming movement	Protein
Pili:		
Sex pilus	Mediates DNA transfer during conjugation	Protein
Common pili or fimbriae	Attachment to surfaces; protection against phagotrophic engulfment	Protein
Capsules (includes "slime layers" and glycocalyx)	Attachment to surfaces; protection against phagocytic engulfment, occasionally killing or digestion; reserve of nutrients or protection against desiccation	Usually polysaccharide; occasionally polypeptide
Cell wall		

Gram-positive bacteria	Prevents osmotic lysis of cell protoplast and confers rigidity and shape on cells	Peptidoglycan (murein) complexed with teichoic acids
Gram-negative bacteria	Peptidoglycan prevents osmotic lysis and confers rigidity and shape; outer membrane is permeability barrier; associated LPS and proteins have various functions	Peptidoglycan (murein) surrounded by phospholipid protein-lipopolsaccharide "outer membrane"
Plasma membrane	Permeability barrier; transport of solutes; energy generation; location of numerous enzyme systems	Phospholipid and protein
Ribosomes	Sites of translation (protein synthesis)	RNA and protein
Inclusions	Often reserves of nutrients; additional specialized functions	Highly variable; carbohydrate, lipid, protein or inorganic
Chromosome	Genetic material of cell	DNA
Plasmid	Extrachromosomal genetic material	DNA

Prokaryotic and Eukaryotic Cells

Cells in our world come in two basic types, prokaryotic and eukaryotic. "Karyose" comes from a Greek word which means "kernel," as in a kernel of grain. In biology, we use this word root to refer to the nucleus of a cell. "Pro" means "before," and "eu" means "true," or "good." So "Prokaryotic" means "before a nucleus," and "eukaryotic" means "possessing a true nucleus." This is a big hint about one of the differences between these two cell types. Prokaryotic cells have no nuclei, while eukaryotic cells do have true nuclei. This is far from the only difference between these two cell types, however. Here's a simple visual comparison between a prokaryotic cell and a eukaryotic cell: If we take a closer look at the comparison of these cells,



we see the following differences:

1. Eukaryotic cells have a true nucleus, bound by a double membrane. Prokaryotic cells have no nucleus. The purpose of the nucleus is to sequester the DNA-related functions of the big eukaryotic cell into a smaller chamber, for the purpose of increased efficiency. This function is unnecessary for the prokaryotic cell, because its much smaller size means that all materials within the cell are relatively close together. Of course, prokaryotic cells do have DNA and DNA functions. Biologists describe the central region of the cell as its "nucleoid" (-oid=similar or imitating), because it's pretty much where the DNA is located. But note that the nucleoid is essentially an imaginary "structure." There is no physical boundary enclosing the nucleoid.
2. Eukaryotic DNA is linear; prokaryotic DNA is circular (it has no ends).
3. Eukaryotic DNA is complexed with proteins called "histones," and is organized into chromosomes; prokaryotic DNA is "naked," meaning that it has no histones associated with it, and it is not formed into chromosomes. Though many are sloppy about it, the term "chromosome" does not technically apply to anything in a prokaryotic cell. A eukaryotic cell contains a number of chromosomes; a prokaryotic cell contains only one circular DNA molecule and a varied assortment of much smaller circlets of DNA called "plasmids." The smaller, simpler prokaryotic cell requires far fewer genes to operate than the eukaryotic cell.
4. Both cell types have many, many ribosomes, but the ribosomes of the eukaryotic cells are larger and more complex than those of the prokaryotic cell. Ribosomes are made out of a special class of RNA molecules (ribosomal RNA, or rRNA) and a specific collection of different proteins. A eukaryotic ribosome is composed of five kinds of rRNA and about eighty kinds of proteins. Prokaryotic ribosomes are composed of only three kinds of rRNA and about fifty kinds of protein.
5. The cytoplasm of eukaryotic cells is filled with a large, complex collection of organelles, many of them enclosed in their own membranes; the prokaryotic cell contains no membrane-bound organelles which are independent of the plasma membrane. This is a very significant difference, and the source of the vast majority of the greater complexity of the eukaryotic cell. There is much more space within a eukaryotic cell than within a prokaryotic cell, and many of these structures, like the nucleus, increase the efficiency of functions by confining them within smaller spaces within the huge cell, or with communication and movement within the cell.

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