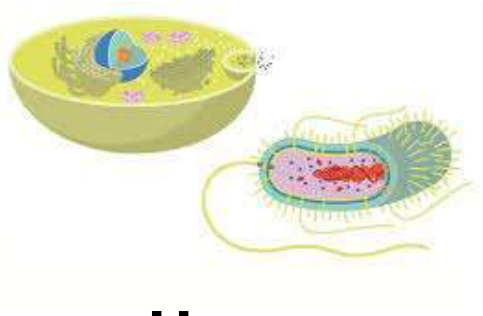

Prokaryotic & Eukaryotic cells

Lecture# 2

Pharmaceutical Microbiology

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EUKARYOTES & PROKARYOTES

- The names given to the two cell types derive from Greek words:

Procaryotic = 'before nucleus'

Eucaryotic = 'true nucleus'

- *Similarities:*

1. Cell contents bounded by a plasma membrane
2. Genetic information encoded on DNA
3. Ribosomes act as site of protein synthesis

EUKARYOTES & PROKARYOTES

- *Differences*

TABLE 1–3 Characteristics of Prokaryotic and Eukaryotic Cells

Characteristic	Prokaryotic Bacterial Cells	Eukaryotic Human Cells
DNA within a nuclear membrane	No	Yes
Mitotic division	No	Yes
DNA associated with histones	No	Yes
Chromosome number	One	More than one
Membrane-bound organelles, such as mitochondria and lysosomes	No	Yes
Size of ribosome	70S	80S
Cell wall containing peptidoglycan	Yes	No

Summary of differences between prokaryote and eukaryote cells

Prokaryotic cells	Eukaryote cells
Small cell (< 5μm)	Larger cells (> 10 μm)
Always unicellular	Often multicellular
No nucleus or any membrane bound organelles	Always have nucleus and membranes bound organelles.
DNA circular, without proteins	DNA is linear and associated with proteins to form chromatin.
Ribosomes are small 70S	Ribosomes are large 80S
No cytoskeleton	Always have cytoskeleton
Motility by rigid rotating flagellum made from flagellin	Motility by flexible waving cilia or flagella made from tubulins.
Cell division is by binary fission	Cell division is by meiosis and mitosis.
Reproduction is always asexual	Reproduction is sexual and asexual.

EUKARYOTES & PROKARYOTES

- (1) Eukaryotic cells contain **organelles, such as mitochondria** and lysosomes, and larger (80S) ribosomes, whereas prokaryotes contain no organelles and smaller (70S) ribosomes.

- (2) Most prokaryotes have a rigid external cell wall that contains **peptidoglycan, a polymer of amino acids and** sugars

  Whilst eukaryotic cells do not contain peptidoglycan. Either they are bound by a flexible cell membrane, or, in the case of

  fungi, they have a rigid cell wall with chitin, a homopolymer of *N-acetylglucosamine*

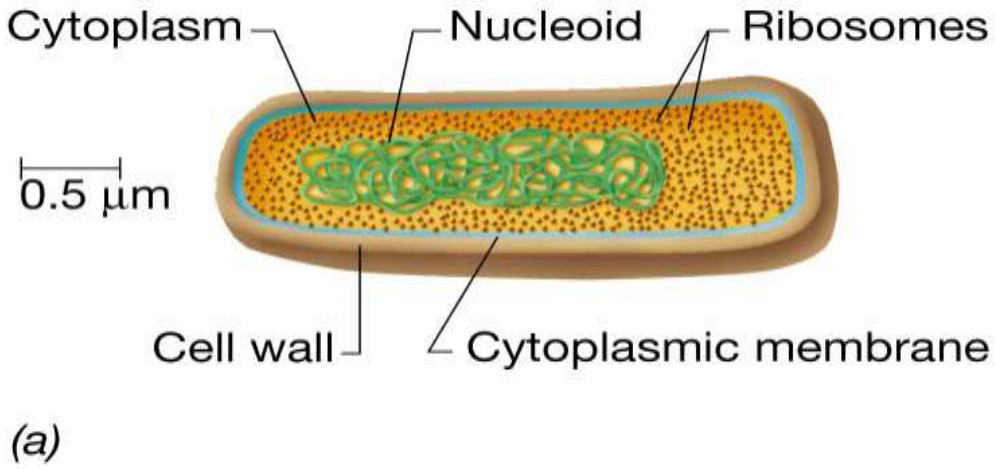
EUKARYOTES & PROKARYOTES

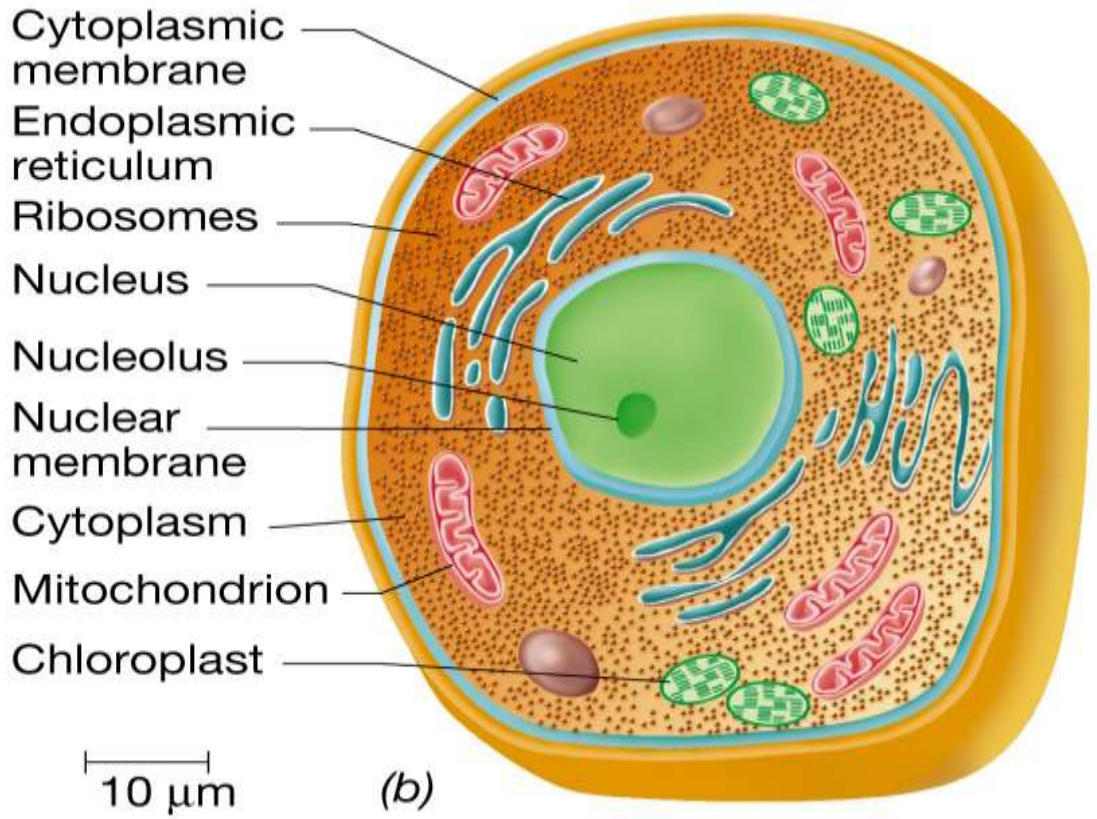
(3)Motility is another characteristic by which these organisms

can be distinguished. Most protozoa and some bacteria are motile, whereas fungi and viruses are nonmotile. The protozoa are a heterogeneous group that possess three different organs of locomotion: flagella, cilia, and pseudopods.

The motile bacteria move only by means of flagella

EUKARYOTES & PROKARYOTES





Prokaryote

Eukaryote

Comparison between organisms

TABLE 1–2 Comparison of Medically Important Organisms

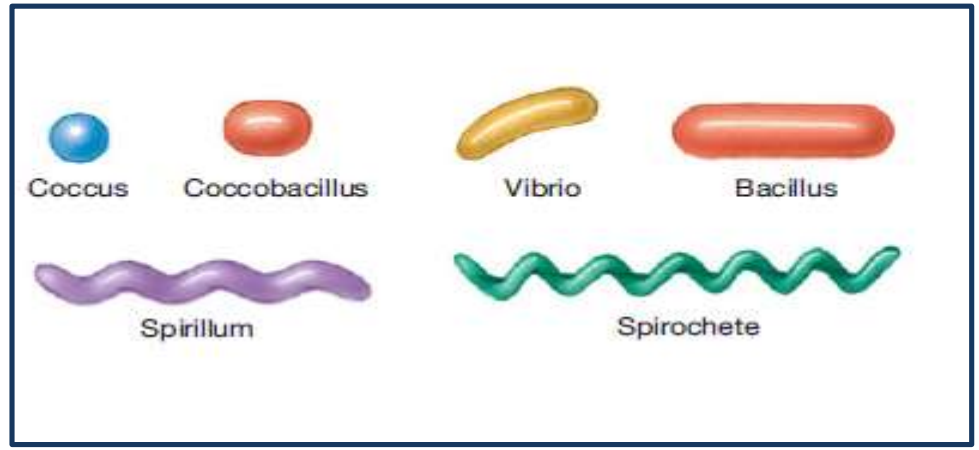
Characteristic	Viruses	Bacteria	Fungi	Protozoa and Helminths
Cells	No	Yes	Yes	Yes
Approximate diameter (μm)[1]	0.02–0.2	1–5	3–10 (yeasts)	15–25 (trophozoites)
Nucleic acid	Either DNA or RNA	Both DNA and RNA	Both DNA and RNA	Both DNA and RNA
Type of nucleus	None	Prokaryotic	Eukaryotic	Eukaryotic
Ribosomes	Absent	70S	80S	80S
Mitochondria	Absent	Absent	Present	Present
Nature of outer surface	Protein capsid and lipoprotein envelope	Rigid wall containing peptidoglycan	Rigid wall containing chitin	Flexible membrane
Motility	None	Some	None	Most
Method of replication	Not binary fission	Binary fission	Budding or mitosis[2]	Mitosis[3]

Bacteria

- Most bacterial cells are 0.5-2.0 µm (or 0.5-5.0 µm) in diameter
- They have a cytoplasmic membrane surrounded by a cell wall; a unique interlinking polymer called peptidoglycan makes the wall rigid.
- The simple prokaryotic cell plan includes no mitochondria, lysosomes, endoplasmic reticulum, or other organelles .
- Their cytoplasm contains only ribosomes and a double-stranded DNA chromosome.
- Bacteria have no nucleus, but all the chemical elements of nucleic acid and protein synthesis are present.

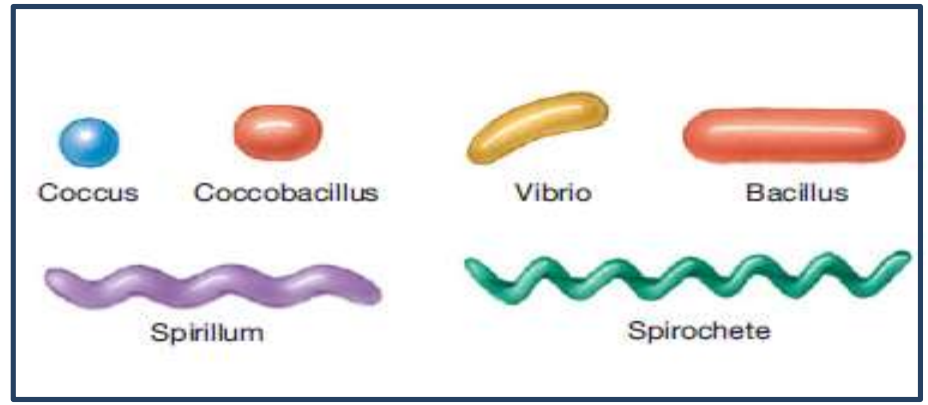
Bacteria- Shape

- The major morphologic forms are spheres, rods, bent or curved rods, and spirals .
- Spherical or oval bacteria are called **cocci (singular: coccus) and are typically arranged in clusters or chains.**
- **Rods are called bacilli (singular: bacillus) and may be straight or curved. Bacilli that are small and resembling cocci are often called coccobacilli.**



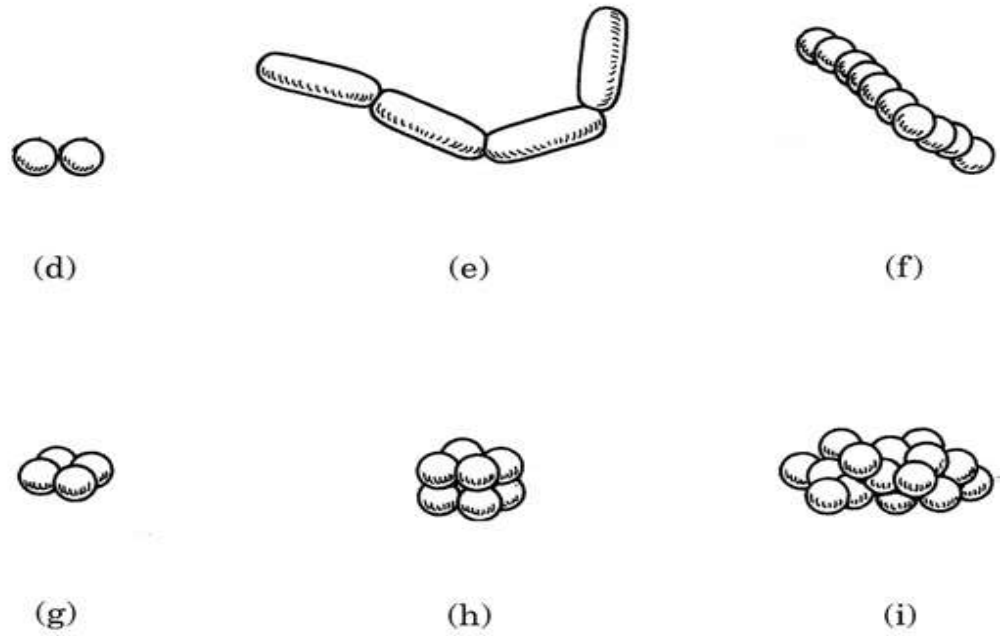
Bacteria- Shape

- Spiral bacteria have a variety of curved shapes.
- A comma-shaped bacterium is called **vibrio (vib’re-o);**
- **A rigid, wavy-shaped one, a spirillum** spiril’um; plural: *spirilla); and a corkscrew-shaped one, a* **spirochete (spi’ro-ket).**
- Some bacteria do not fit any of the preceding categories but rather have spindle shapes or irregular, lobed shapes



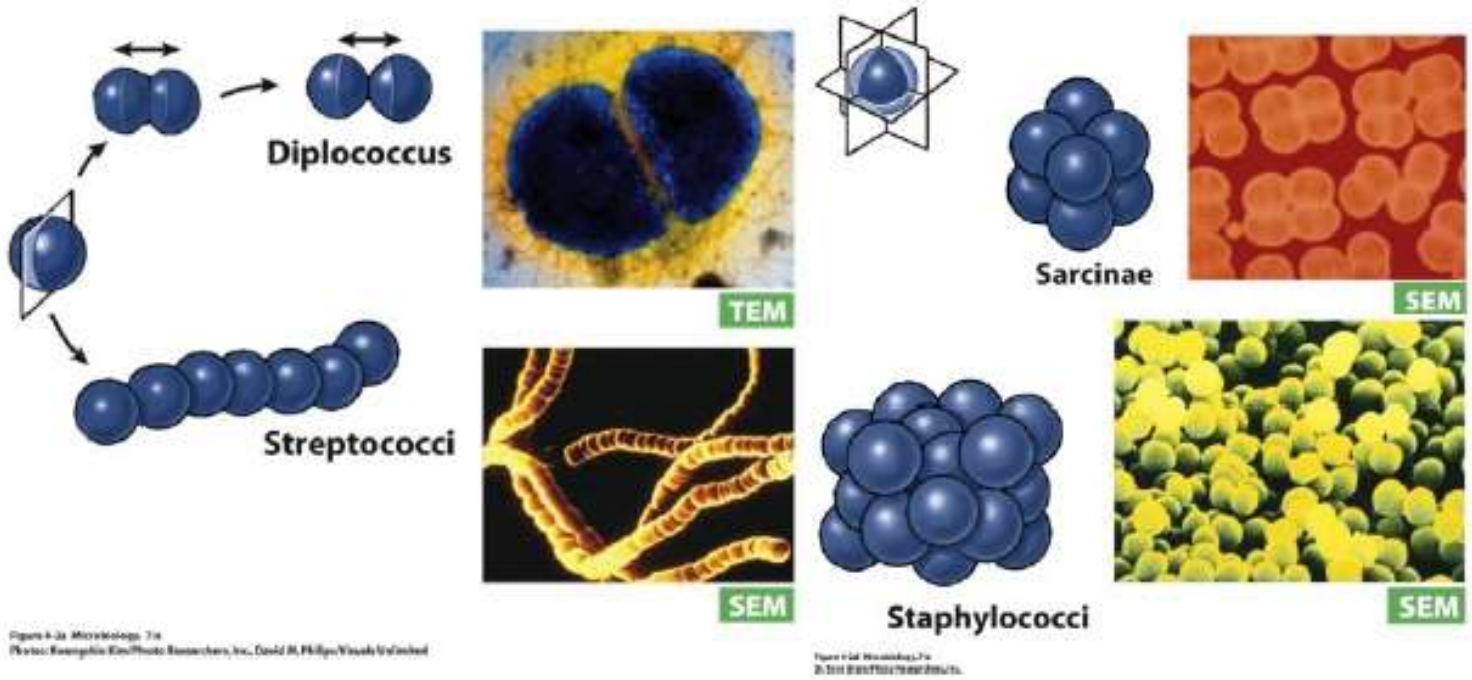
Bacteria- Arrangement

- Cocci can divide in one or more planes, or randomly. Division in one plane produces cells in pairs (indicated by the prefix **diplo-) or in chains (strepto-).**
- Random division planes produce grapelike clusters (**staphylo-**
- Bacilli divide in only one plane, but they can produce cells connected end-to-end (like train cars) or side-byside.
- Spiral bacteria are not generally grouped together



These basic shapes may join to form (d) pairs, (e and f) chains, (g) sheets, (h) packets or (i) irregular aggregates

Prokaryotes: Arrangement



Bacteria- Cell structure

1. Cell wall

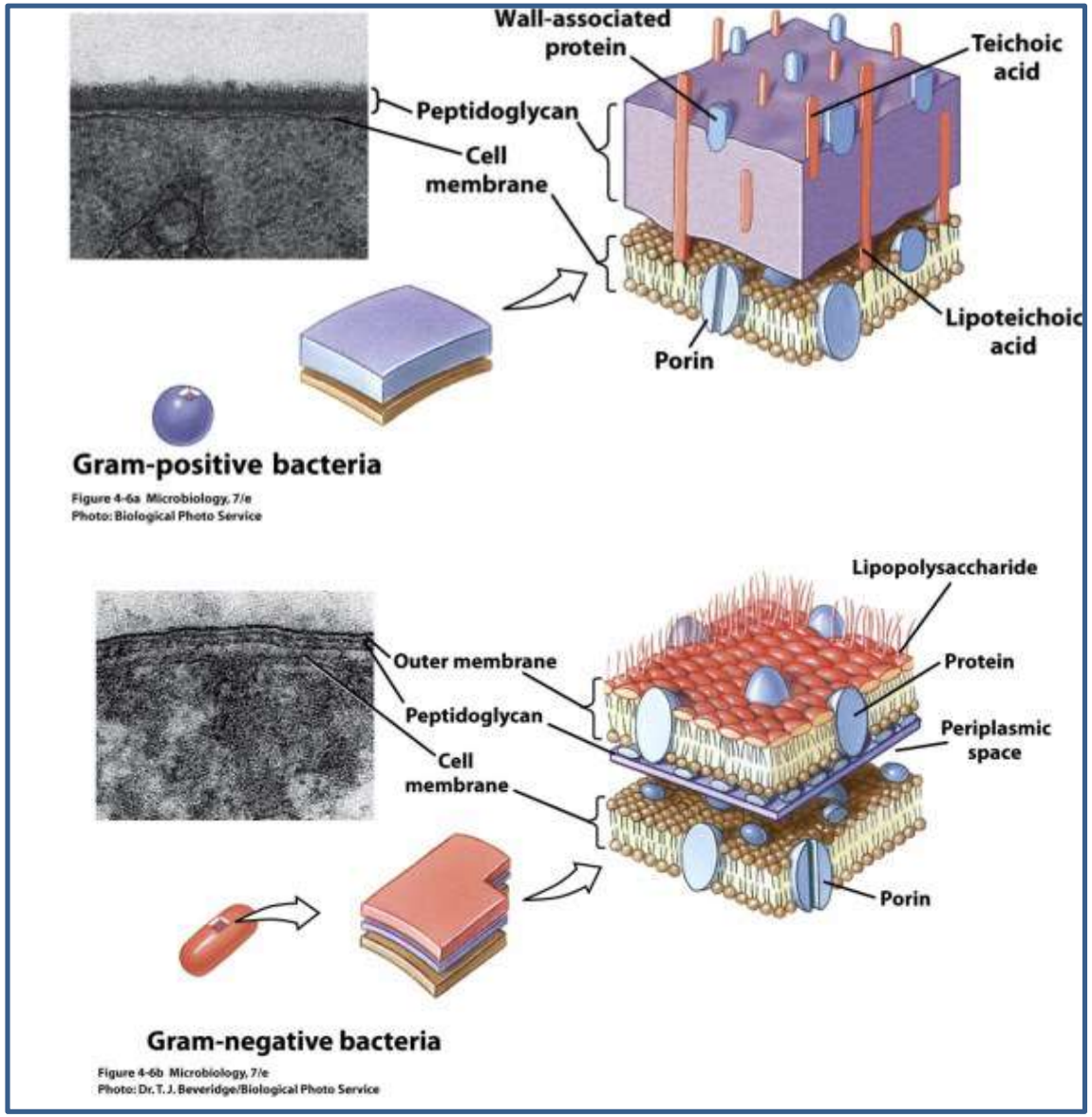
- thick, rigid cell wall, which maintains the integrity of the cell.
- The major component of the cell wall is a substance unique to bacteria, called *peptidoglycan (murein).*

- It maintains the characteristic shape of the cell
- It protects the cell from chemical and physical assault, while still permitting the rapid exchange of nutrients and metabolic by products required for rapid growth and division.

Structure of Bacterial Cells: Cell Wall

- Components of the bacterial cell wall:

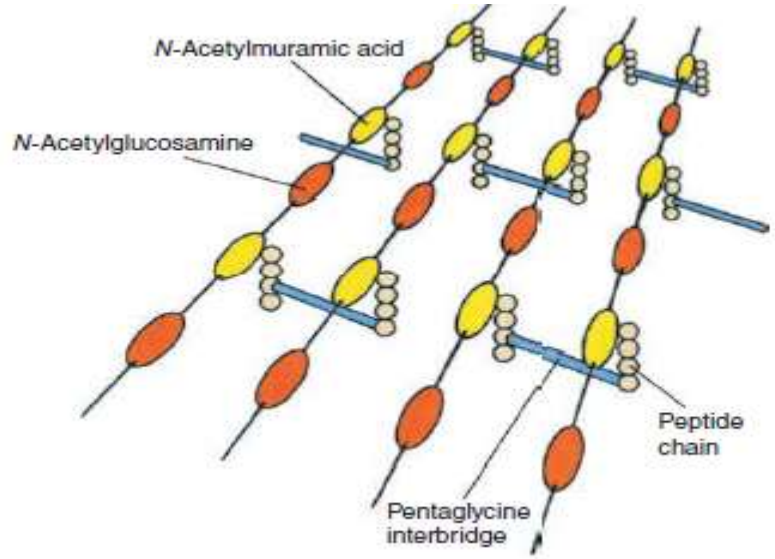
1. Peptidoglycan
2. Outer membrane (G-ve)
3. Periplasmic space



Bacteria-Cell wall component

- This is a high molecular

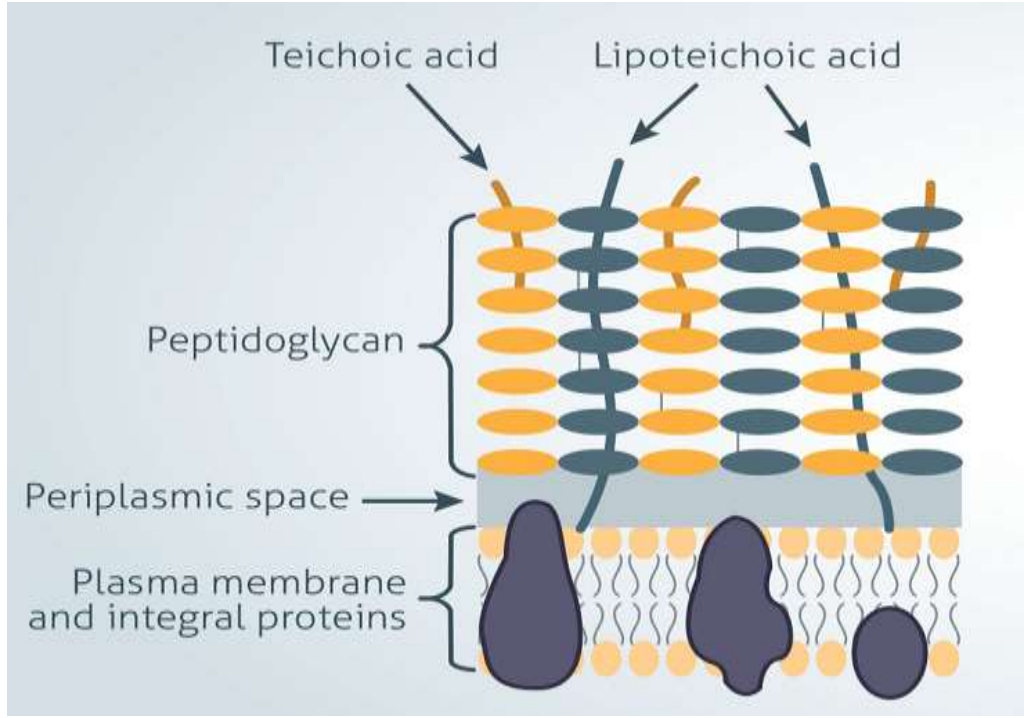
weight polymer whose basic subunit is made up of three parts: *N-acetylglucosamine, N-acetylmuramic acid and a short peptide chain*



schematic diagram of one model of peptidoglycan. Shown are the polysaccharide chains.

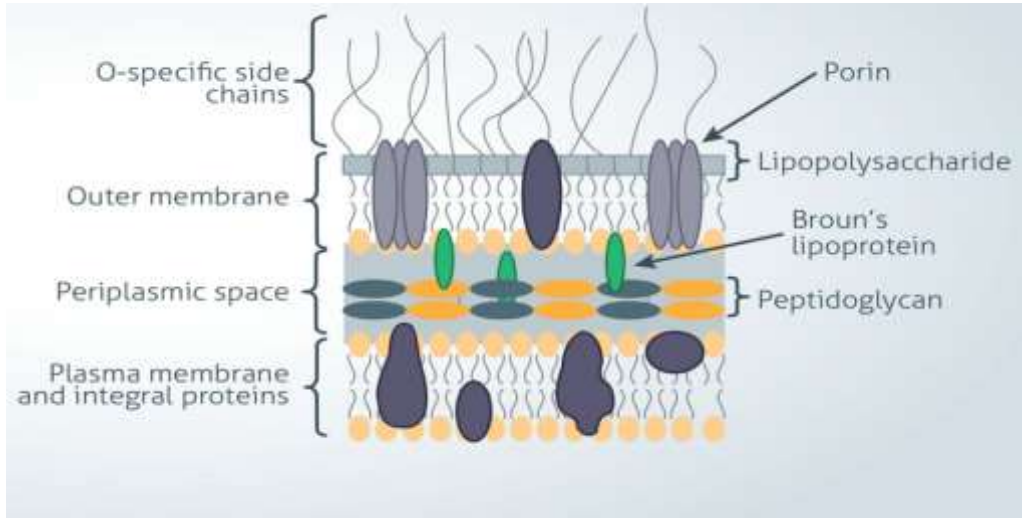
Gram positive cell wall

- Contains thick layers of peptidoglycan to support the cell membrane and provide a place of attachment for other molecules.
- The cell walls also contain chains of **teichoic acid** which helps in maintaining cell shape and play a role in proper cell division.



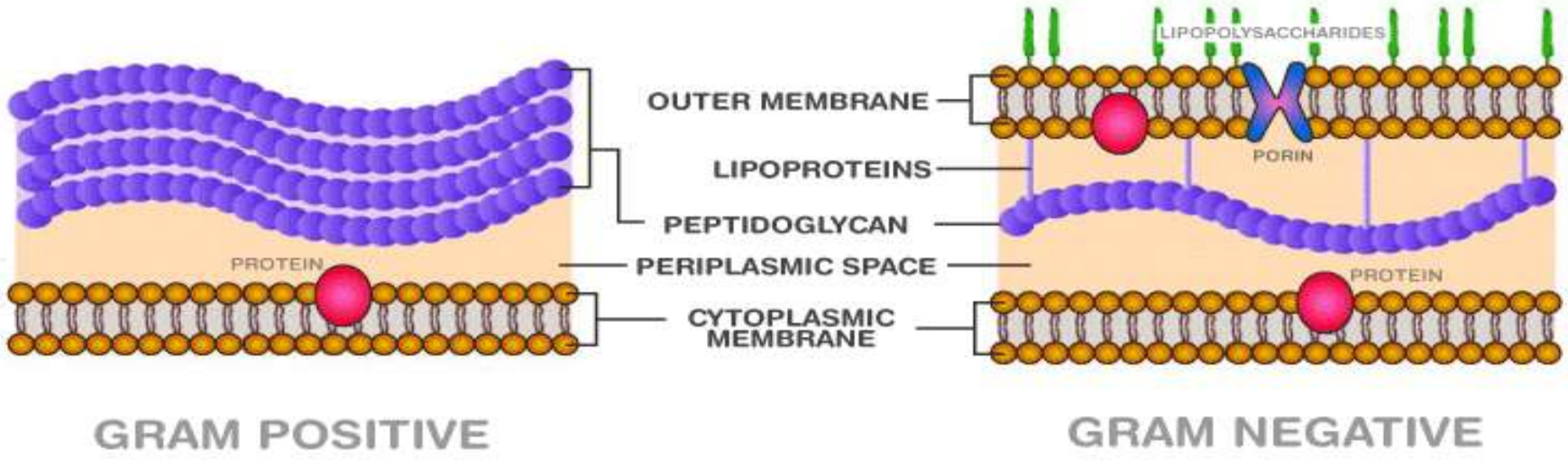
Gram negative cell wall

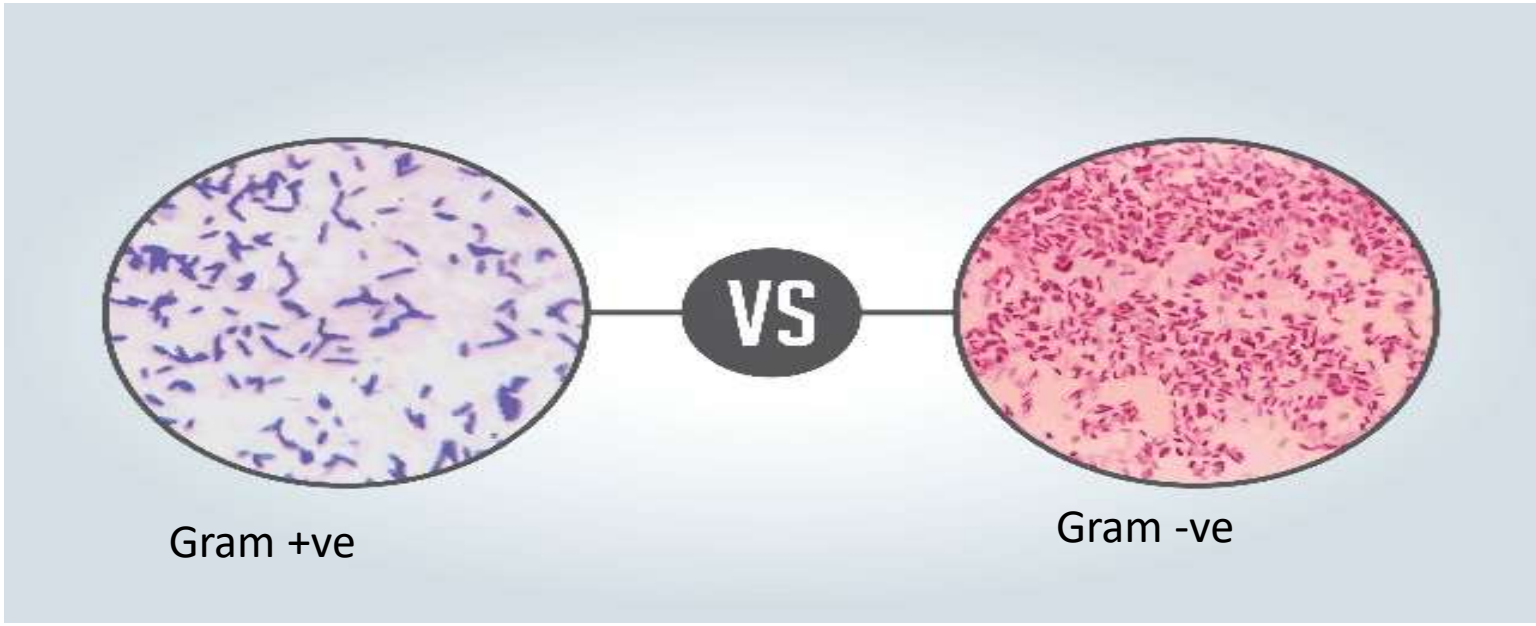
- The Gram negative bacterial cell wall is composed of a single thin layer peptidoglycan.
- A gel-like matrix called periplasmic space is presented
- An outer membrane containing lipopolysaccharide (LPS)



GRAM-POSITIVE AND GRAM-NEGATIVE BACTERIA

BYJU'S The Learning App





Gram stain

All purple

1. Crystal violet (1 minute)
Drain, rinse

All purple;
iodine acts as mordant
to set stain

2. Iodine (1 minute)
Drain, rinse

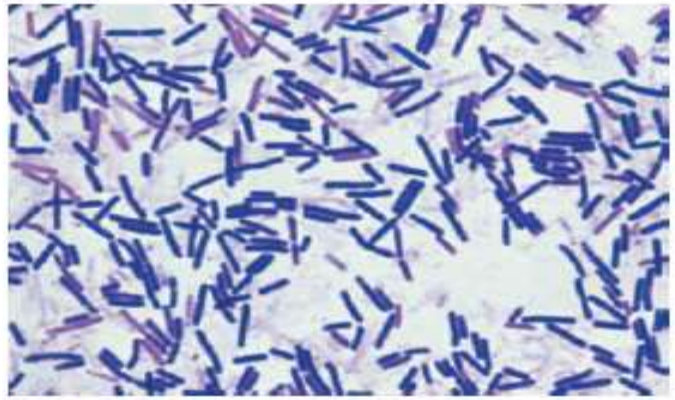
Gram + cocci = purple
Gram − rods = clear

3. Decolorize with alcohol
(one quick rinse);
immediately after, rinse with water

Gram + cocci = purple
Gram − rods = red (pink)

4. Safranin (30–60 seconds)
Drain, rinse, blot

(a)



(b)

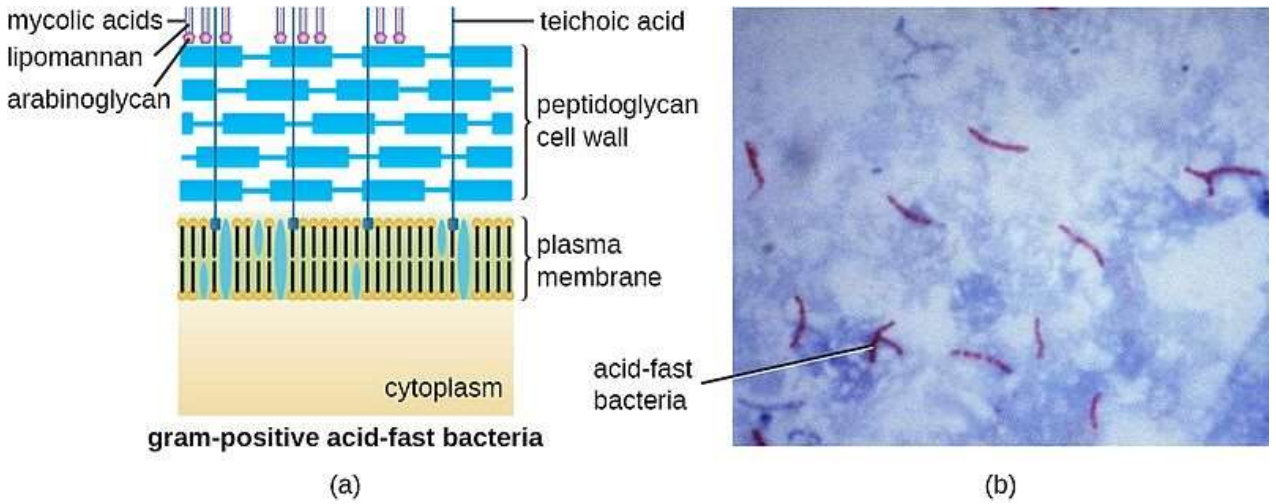
FIGURE 3.30 The Gram stain.
(a) Steps in Gram staining.
(b) Gram-positive cells retain the purple color of crystal violet, whereas Gram-negative cells are decolorized with alcohol and subsequently pick up the red color of the safranin counterstain. *(CNRI/Science Source)*

Gram status is important in medicine; the presence or absence of a cell wall will change the bacterium's susceptibility to some antibiotics.

Acid-Fast Bacteria

•Acid-fast cells contain a large amount of lipids and waxes in their cell walls **primarily mycolic acid.**

• Acid fast bacteria are usually members of the genus Mycobacterium or Nocardia

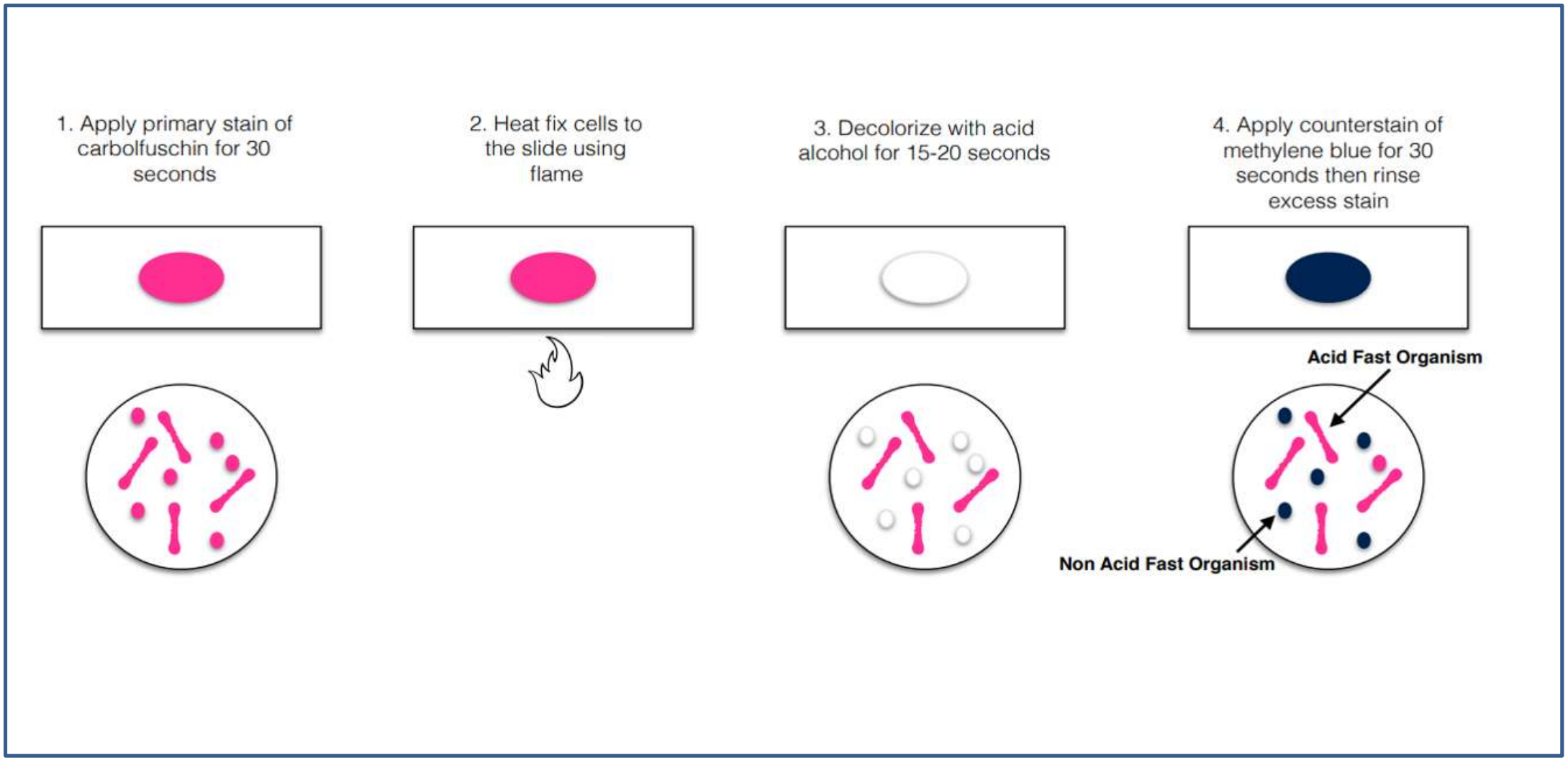


Ziehl-Neelsen staining

- It is used to stain species of *Mycobacterium tuberculosis* that do not stain with the standard laboratory staining procedures like Gram staining.

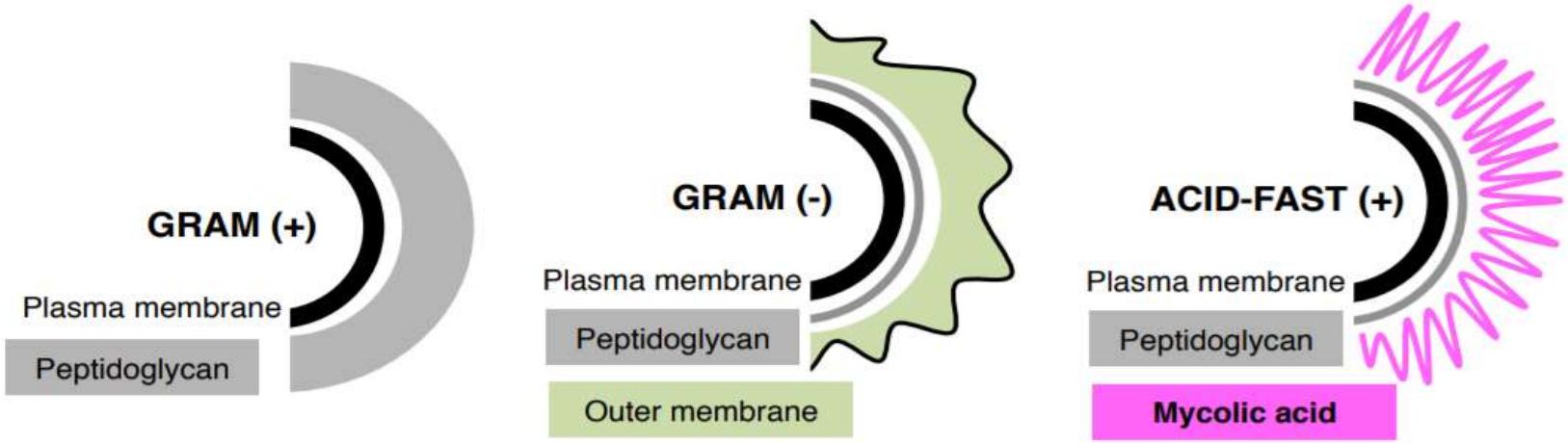
- The stains used are the red colored Carbol fuchsin that stains the bacteria and a counter stain like Methylene blue

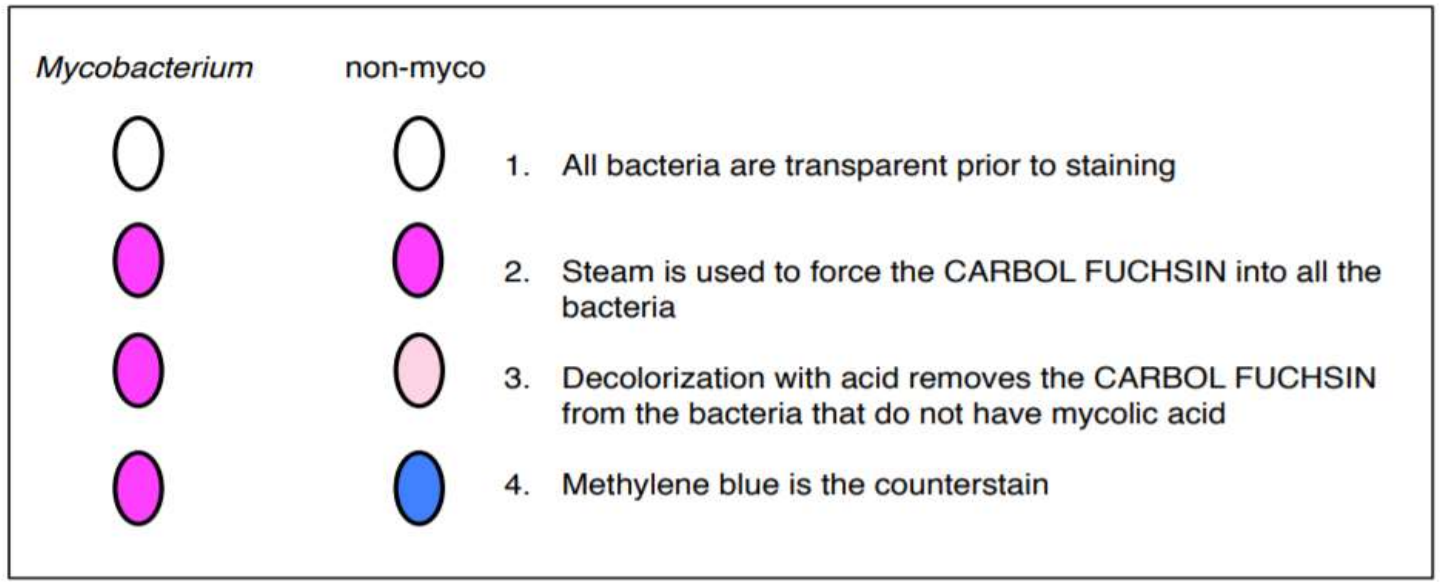
Acid fast stain



Acid- fast bacteria staining

- Primary stain binds cell wall mycolic acids
- Intense decolorization does not release primary stain from the cell wall of AFB
- Color of AFB-based on primary stain
- Counterstain provides contrasting background


GRAM (+)
Plasma membrane
Peptidoglycan
GRAM (-)
Plasma membrane
Peptidoglycan
Outer membrane
ACID-FAST (+)
Plasma membrane
Peptidoglycan
Mycolic acid

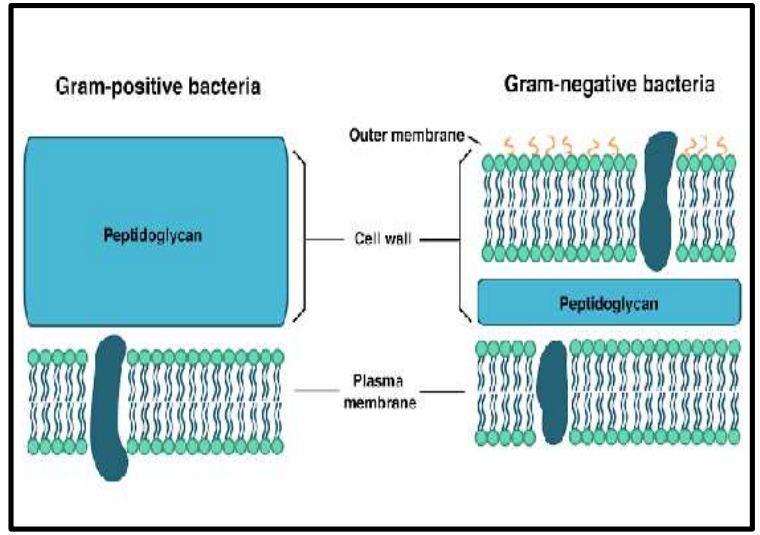

Mycobacterium
non-myco
1. All bacteria are transparent prior to staining
2. Steam is used to force the CARBOL FUCHSIN into all the bacteria
3. Decolorization with acid removes the CARBOL FUCHSIN from the bacteria that do not have mycolic acid
4. Methylene blue is the counterstain

Outer membrane

- The **outer membrane,** found primarily in **Gram-negative bacteria**, is a bilayer membrane.

- It is attached to peptidoglycan by **lipoproteins**
- It acts as a **coarse sieve and has little control over the movement of substances in & out of cells** However, it does control the transport of certain proteins from the environment.

- Proteins called porins form channels through the outer membrane.



Structure of Bacterial Cells

- **Lipopolysaccharide (LPS), also called endotoxin, is an** important part of the outer membrane and can be used to identify Gram-negative bacteria. It is an integral part of the cell wall and is not released until the cell walls of dead bacteria are broken down.

- LPS consists of polysaccharides and **lipid A.**

- It is these repeating units that are used to identify different Gram-negative bacteria. The lipid A portion is responsible for the toxic properties that make any Gram negative infection a potentially serious medical problem

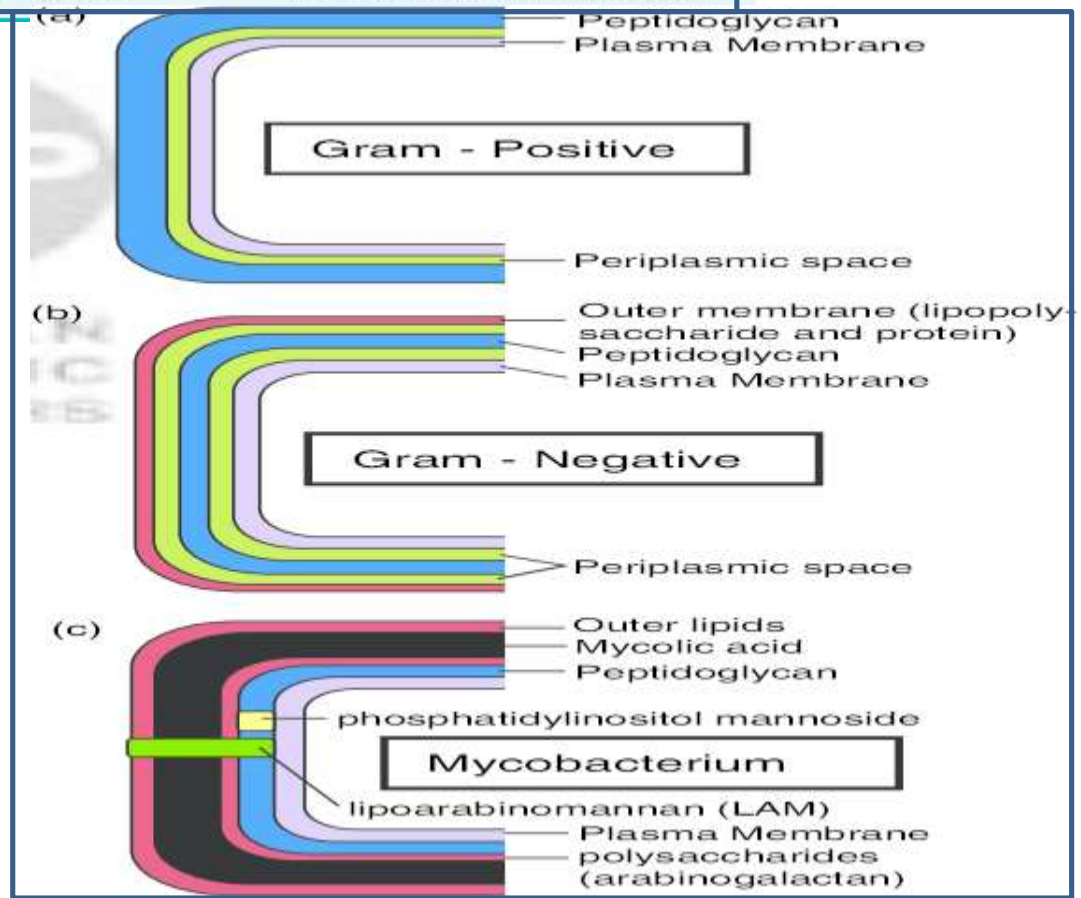
Periplasmic Space

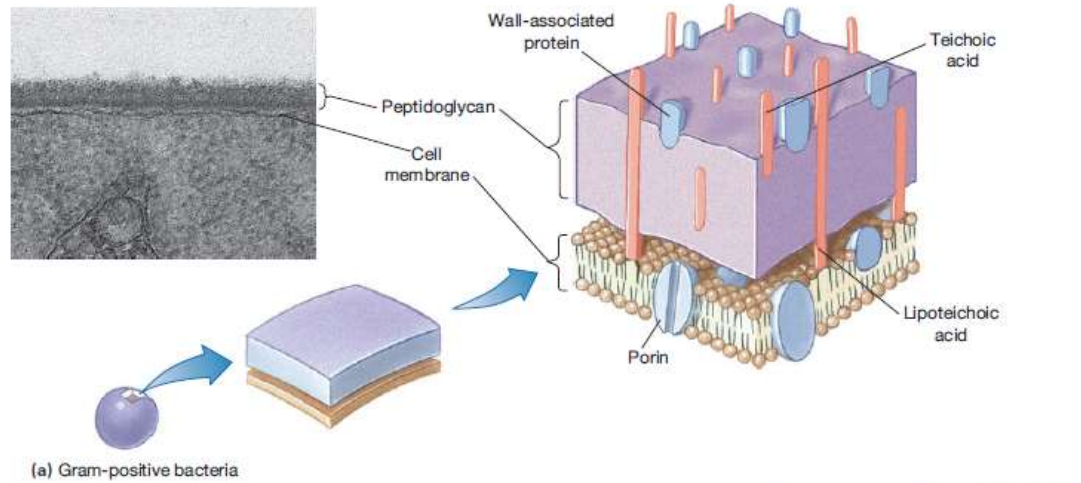
- Is most easily observed by electron microscopy of Gram-negative bacteria.
- It represents a very active area of cell metabolism.
- This space contains not only the cell wall peptidoglycan but also many digestive enzymes and transport proteins that destroy potentially harmful substances and transport metabolites into the bacterial cytoplasm.

TABLE 4.2 Characteristics of the Cell Walls of Gram-Positive, Gram-Negative, and Acid-Fast Bacteria

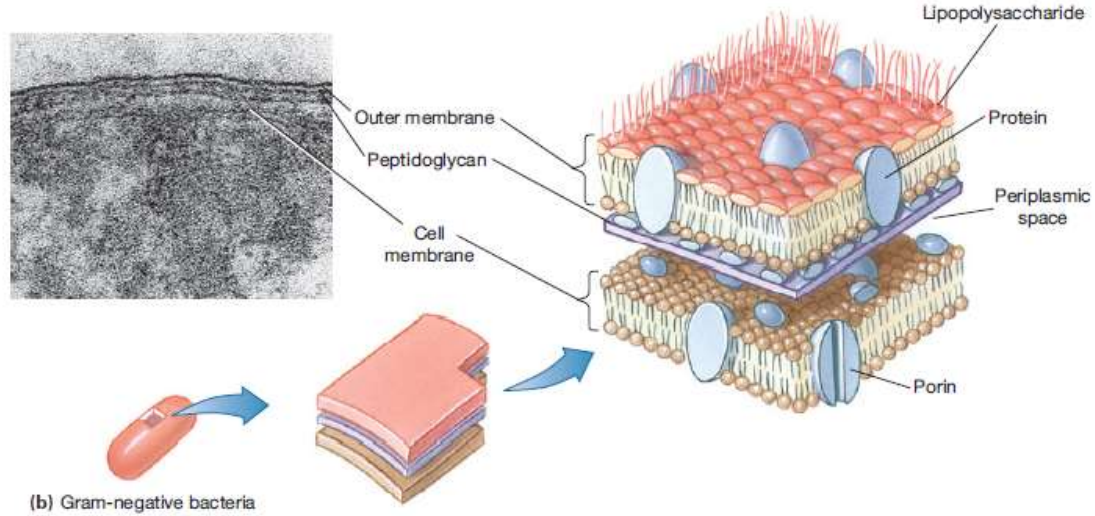
Characteristic	Gram-Positive Bacteria	Gram-Negative Bacteria	Acid-Fast Bacteria
Peptidoglycan	Thick layer	Thin layer	Relatively small amount
Teichoic acid	Often present	Absent	Absent
Lipids	Very little present	Lipopolysaccharide	Mycolic acid and other waxes and glycolipids
Outer membrane	Absent	Present	Absent
Periplasmic space	Absent	Present	Absent
Cell shape	Always rigid	Rigid or flexible	Rigid or flexible
Results of enzyme digestion	Protoplast	Spheroplast	Difficult to digest
Sensitivity to dyes and antibiotics	Most sensitive	Moderately sensitive	Least sensitive
Examples	*Staphylococcus aureus*	*Escherichia coli*	*Mycobacterium tuberculosis*

Summary

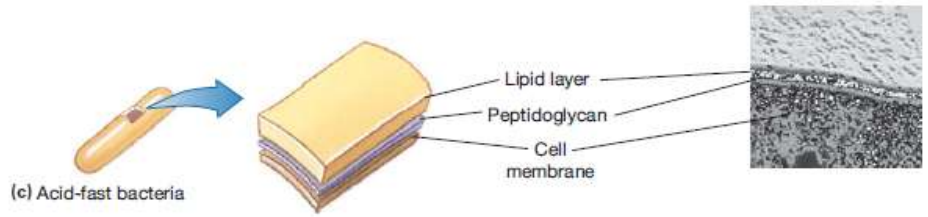




(a) Gram-positive bacteria



(b) Gram-negative bacteria



(c) Acid-fast bacteria

Cytoplasmic (Plasma) membrane

- Thin layer 5-10 nm, separates cell wall from cytoplasm.
- Acts as a semi-permeable membrane: controls the inflow and outflow of metabolites.
- Composed of lipoproteins with small amounts of carbohydrates.
- The main function of the cell membrane is to regulate the movement of materials into and out of a cell by transport mechanisms. It synthesizes cell wall components, assists with DNA replication, secretes proteins, carries on respiration, and captures energy as ATP. It also contains bases of appendages called *flagella*

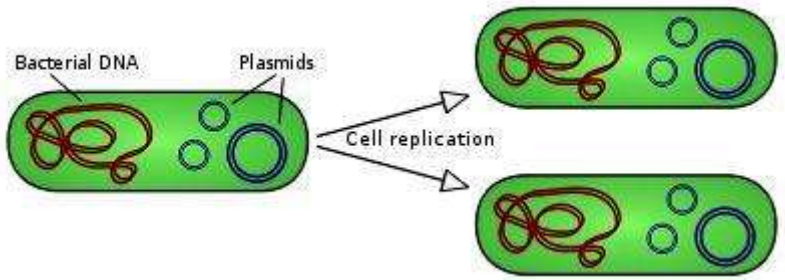
Other Cytoplasmic Components

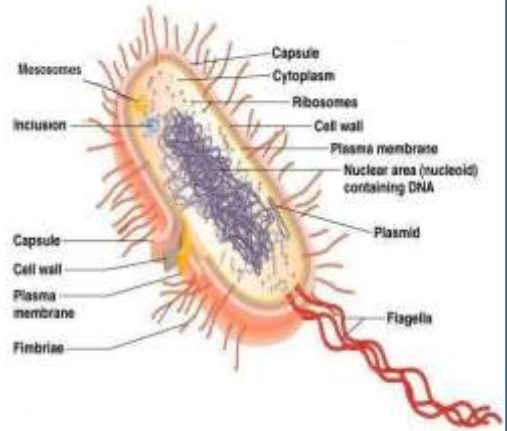
1. Ribosomes : Are of RNA and protein. often grouped in long chains called **polyribosomes and their main action is protein synthesis**.

2. Nuclear Region :

- No nucleolus- No nuclear membrane
- single, circular double stranded DNA
- Many bacteria exchange genetic information carried on plasmids

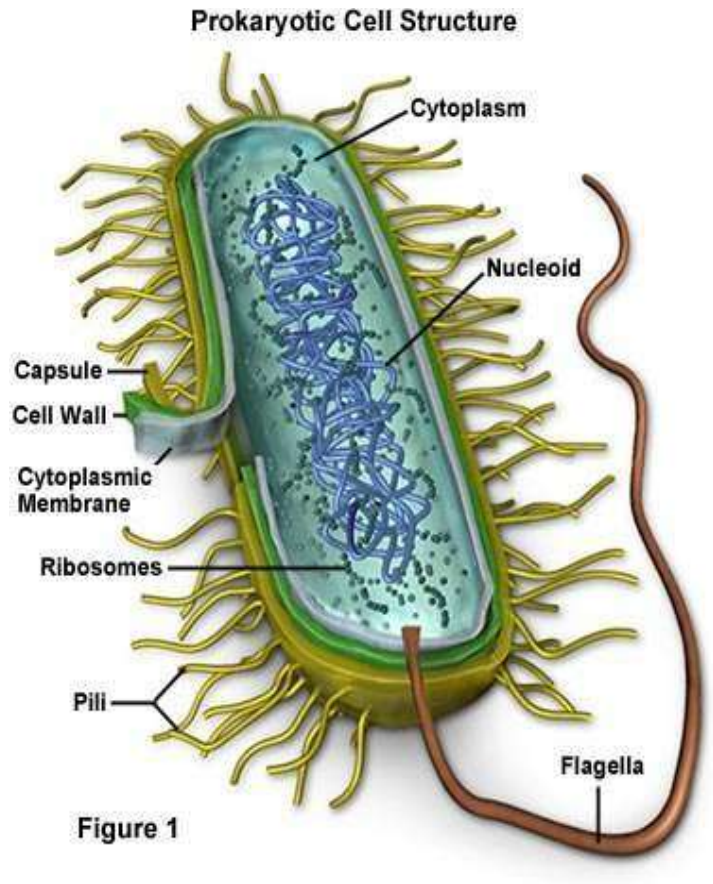
(small, specialized genetic elements capable of self-replication)





External Structure

- **Bacteria also often have these features:**
- **flagella-----movement**
- **pili--------- attachment** not involved in movement.
- **capsule------** Protects bacteria from lytic enzymes and Inhibits phagocytosis



Types of pili

1. Conjugation-pili: long pili, found in some groups of bacteria, attach 2 bacteria & DNA is transferred through them, a process called conjugation (like sexual reproduction). Results in transfer of antibiotic resistance among bacteria.

2. Attachment pili or fimbriae: short, attach bacteria to surfaces or air-water interface**. Contributes to the pathogenicity of certain bacteria-ability to produce disease**-by enhancing colonizationon surface of cells of other organisms

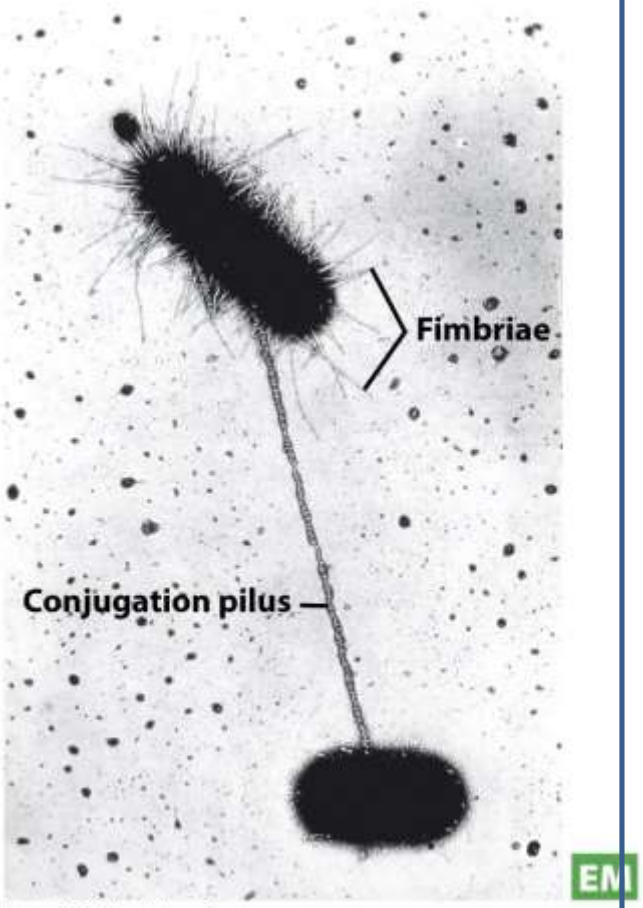
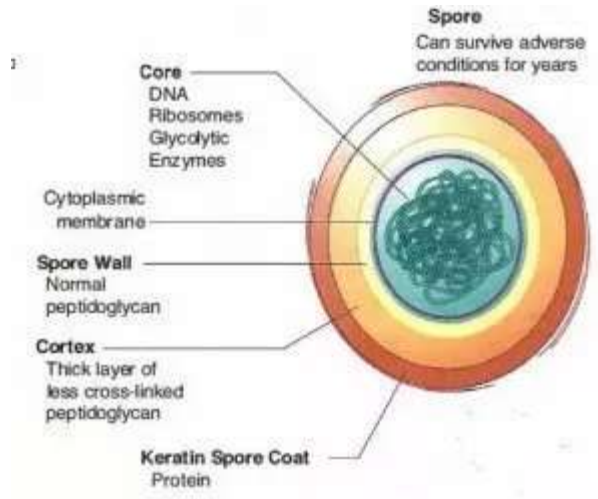


Figure 4-16 Microbiology, 7/e
Courtesy Charles C. Brinton, Jr., and Judith Carnahan

Structure of Bacterial Cells: SPORES

- To enhance survival during periods of environmental hostility some gram-positive rods form of a dormant cell called an endospore inside the original cell.
- Endospores can be released from the original cell as free spores.
- Spores are the most resistant life forms known.
- Sporulation: a process where bacteria prepare for the possibility of future adverse conditions



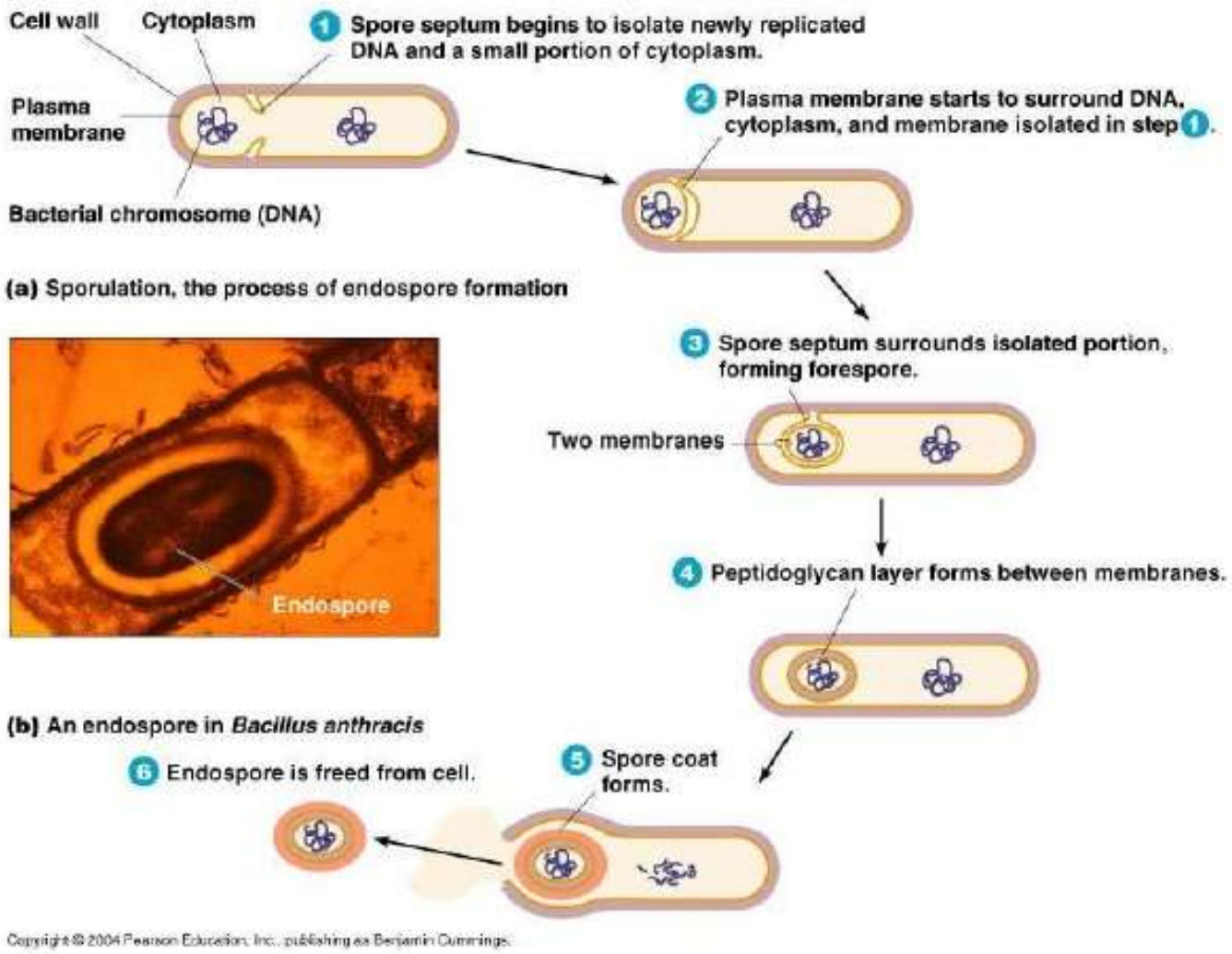
Examples of Spore forming Bacteria-

Spores formed by only two genera of Gram-positive rods are of medical importance.

Bacillus spp

Clostridium spp

Sporulation Process



Medical significance of sporulation

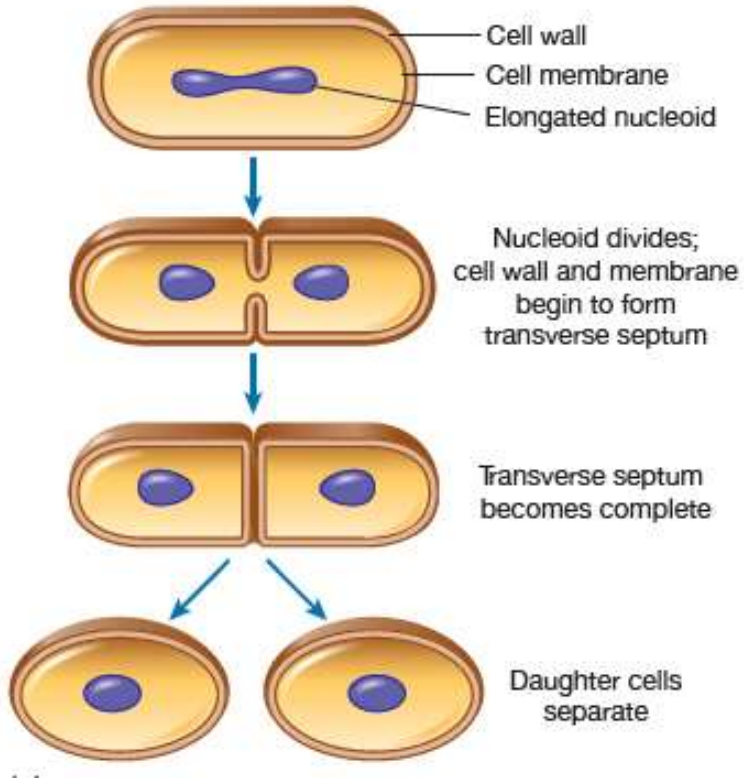
- Spores can survive for many years in soil and other inanimate objects.
- Wound contaminated with soils can be infected with spores and cause diseases such as tetanus, gas gangrene.
- Spores are highly resistant to many chemicals, including most disinfectants and antibiotics.
- Only solution designated as sporicidal will kill spores.
- Spores of these organisms can remain viable for many years and are generally not killed by boiling, but they can be killed by autoclaving (that is, subjecting the spores to temperatures above 120^{o}C at elevated pressure).

Cell Division

- In **binary fission, a cell duplicates its components**

  and divides into two cells.

- In continuously dividing cells, DNA synthesis also is continuous and replicates the bacterial chromosome shortly before the cell divides.

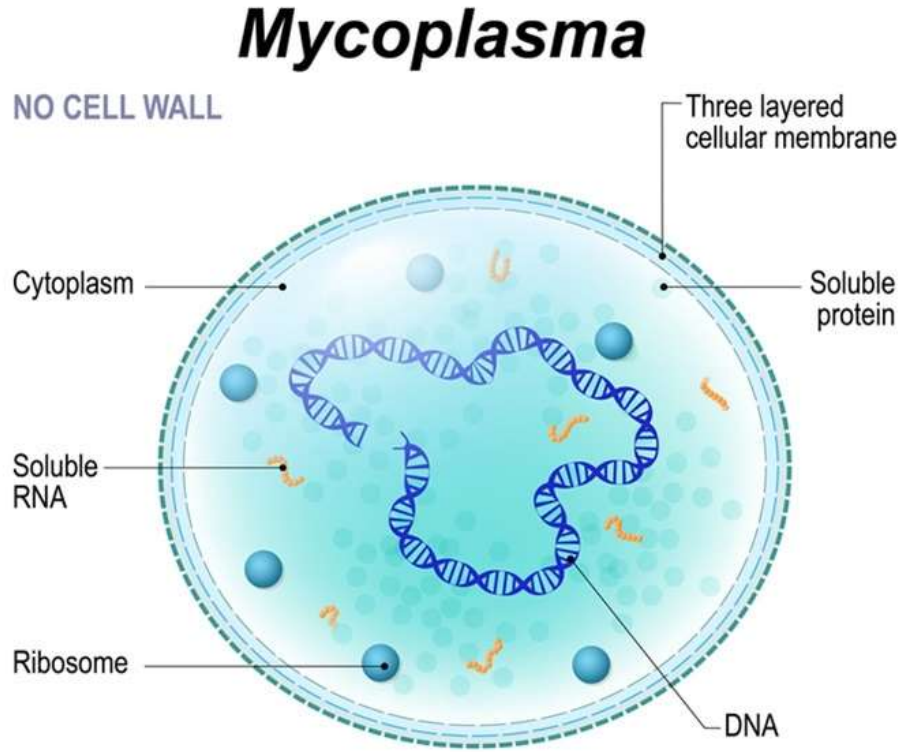


Types of bacteria

A. Typical bacteria

B. Atypical bacteria:

include groups of organisms such as Mycoplasma, Chlamydia, and Rickettsia that, although prokaryotic, lack significant characteristic structural components or metabolic capabilities that separate them from the larger group of typical bacteria



Types of bacteria –energy production

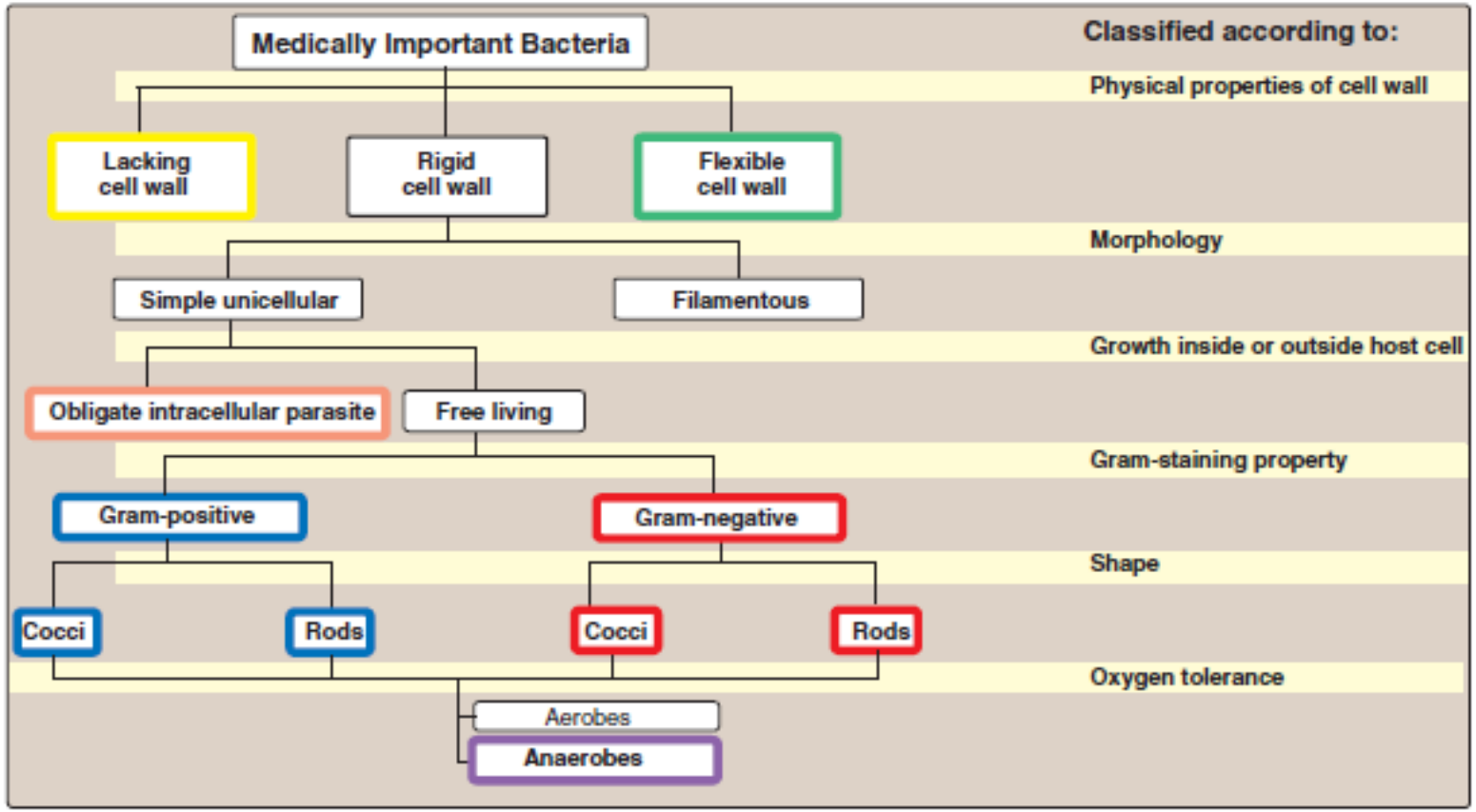
- **Aerobic bacteria: needs oxygen for survival and energy production.**

Example: Pseudomonas aeruginosa.

- **Anaerobic bacteria: cannot live in the presence of oxygen.**

Example: ***Clostridium tetani***

Hierarchical classification of clinically important bacteria according to six distinguishing characteristics.



Classification of Microbes:

Classification of Bacteria:

Medically important Bacteria are divided into three groups:

1-Lacking Cell Walls: Example: *Mycoplasma*

2-Flexible (have Cell Walls):

Example: Spirochetes (*Treponema*).

3-Rigid Cell Walls:

A- Filamentous bacteria: (antibiotic producers).

Example: *Streptomyces*.

B- Simple Unicellular:

1-Obligate intracellular parasite:

Example: (*Rickettsia , Chlamydia*).

2-Free-living bacteria:

Stages of the bacterial growth cycle

- **Because bacteria reproduce** by binary fission (one becomes two, two become four, four become eight, etc.), the number of cells increases exponentially with time (the exponential, or log, phase of growth).
- Eventually, growth slows and ceases entirely (stationary Phase) as nutrients are depleted, and toxic waste products accumulate. Most cells in a stationary phase are not dead