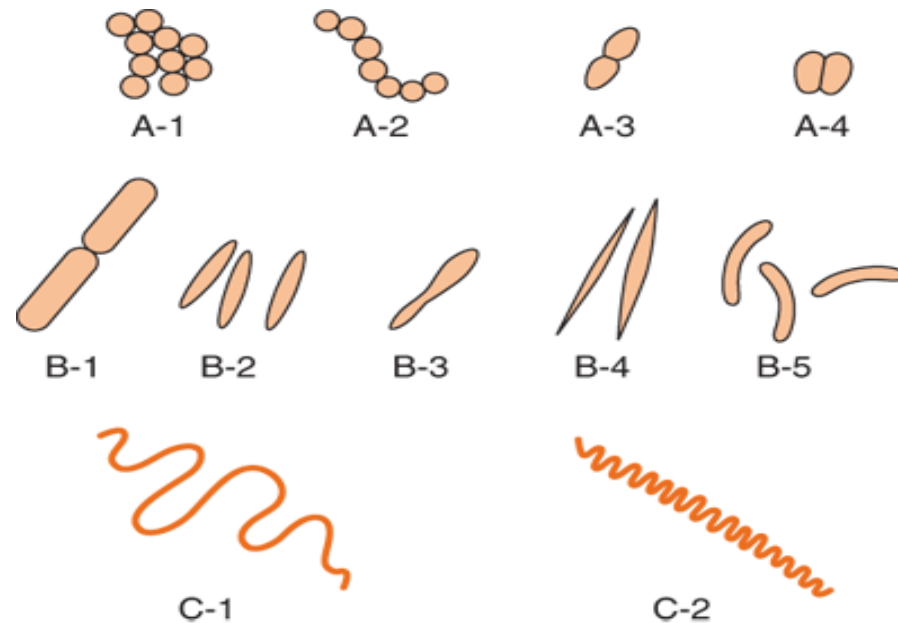


Chapter 5

Structure and Classification of Bacterial Cell

Dr. Ariyah Terasawat



Source: Warren Levinson: Review of Medical Microbiology and Immunology, 14th Edition, www.accessmedicine.com
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A: Cocci in clusters (e.g., *Staphylococcus*; A-1); chains (e.g., *Streptococcus*; A-2); in pairs with pointed ends (e.g., *Streptococcus pneumoniae*; A-3); in pairs with kidney bean shape (e.g., *Neisseria*; A-4).

B: Rods (bacilli): with square ends (e.g., *Bacillus*; B-1); with rounded ends (e.g., *Salmonella*; B-2); club-shaped (e.g., *Corynebacterium*; B-3); fusiform (e.g., *Fusobacterium*; B-4); comma-shaped (e.g., *Vibrio*; B-5).

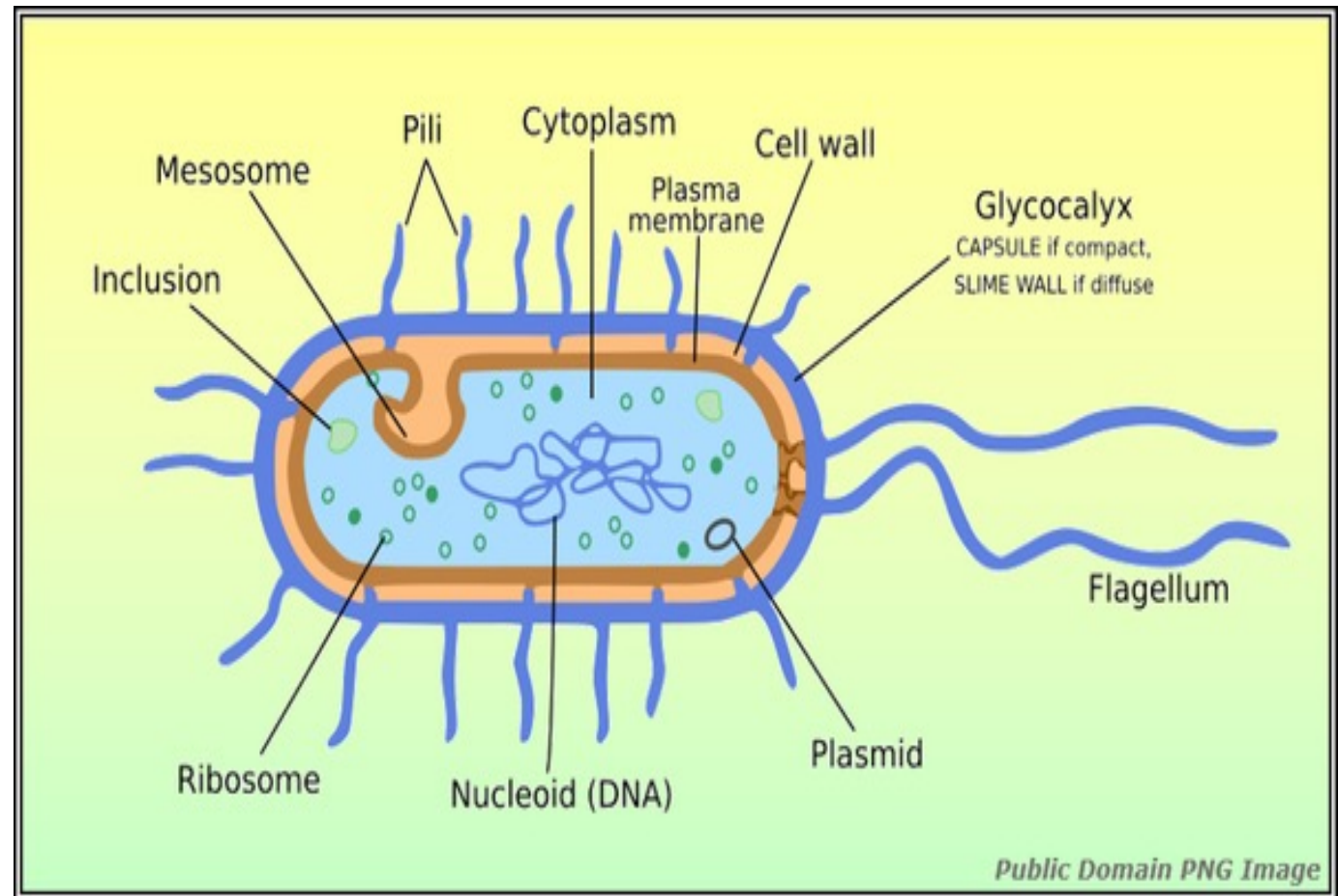
C: Spirochetes: relaxed coil (e.g., *Borrelia*; C-1); tightly coiled (e.g., *Treponema*; C-2).

Essential component:

- Cell wall
- Cell membrane
- Ribosome
- Nucleoid
- Mesosome
- Periplasm (in Gram negative bacteria)

• Non essential component:

- Capsule
- Pilli
- Flagella
- Spore
- Plasmid
- Glycocalyx
- Granules



Essential component:

Cell wall

Cell membrane

Ribosome

Nucleoid

Mesosome

Periplasm (in Gram negative bacteria)

Cell Wall:



The cell wall is the outermost component, common to all bacteria (except *Mycoplasma* species, which are bounded by a cell membrane, not a cell wall).



The cell wall is located external to the cytoplasmic membrane and is composed of peptidoglycan.

Cell Walls of Gram-Positive

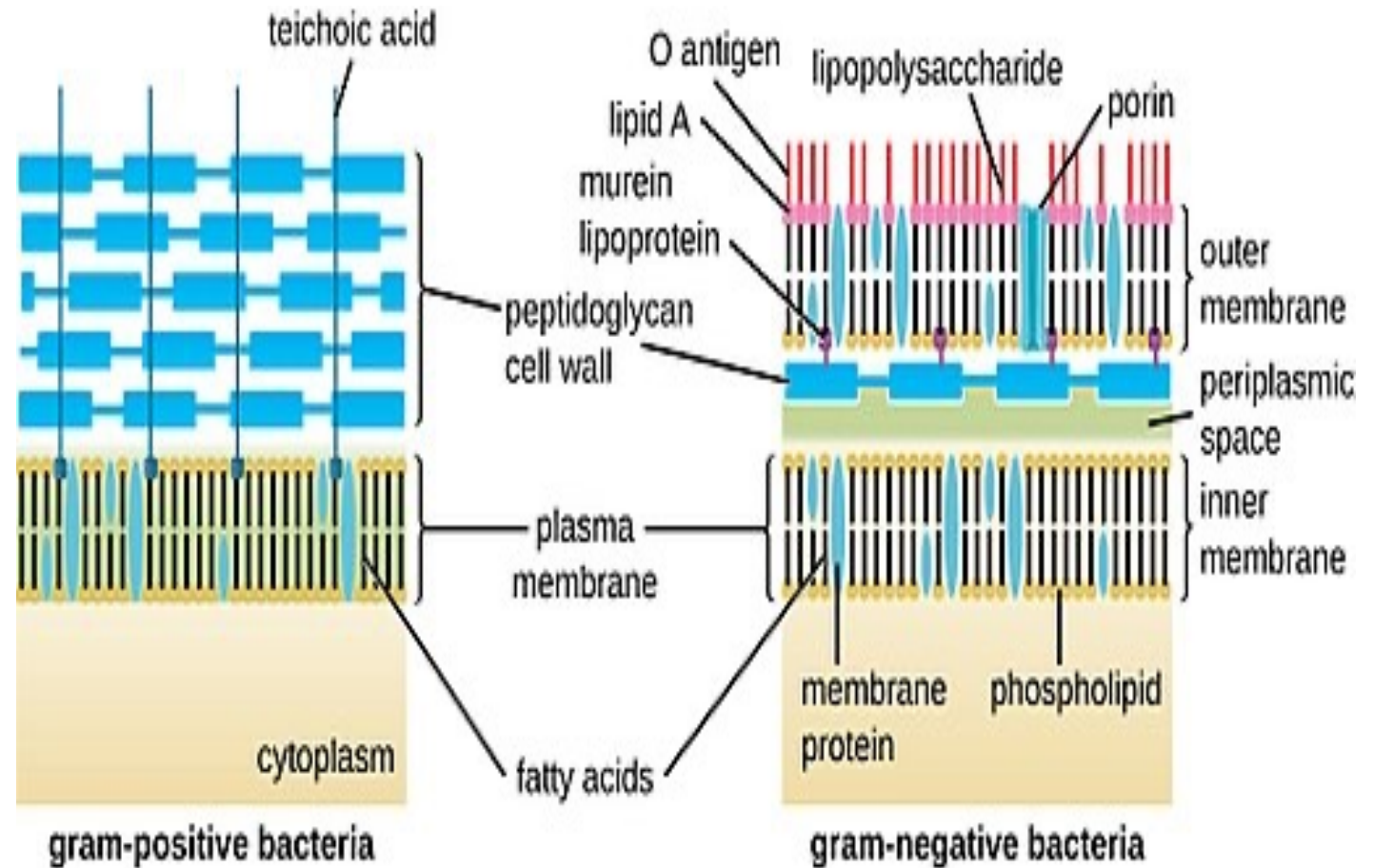
bacteria:

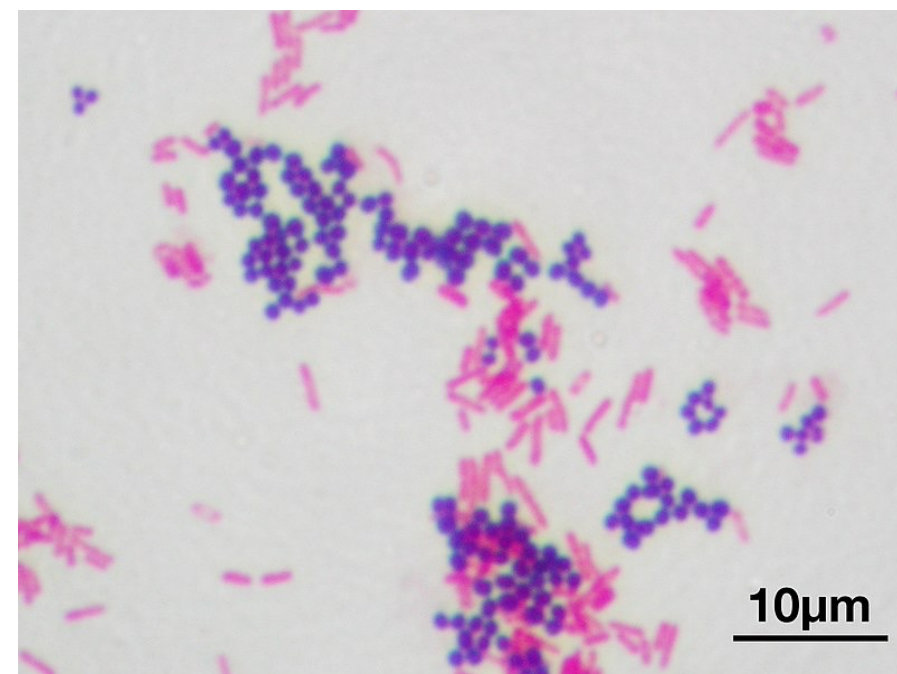
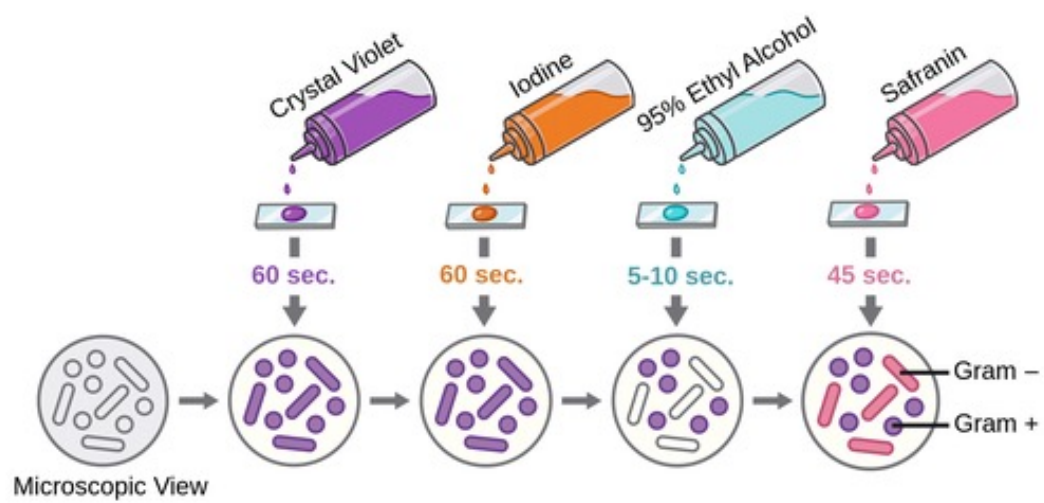
- The peptidoglycan layer:
 - up to 40 sheets of peptidoglycan,
 - comprising up to 50% of the cell wall material
- Many gram-positive bacteria also have fibers of teichoic acid that protrude outside the peptidoglycan.

Cell Walls of Gram-negative

bacteria:

- Peptidoglycan layer
 - one or two sheets,
 - comprising 5–10% of the cell wall material.
- Lipopolysaccharide,
- Lipoprotein, and
- Phospholipid.





Gram positive

- *Nocardia asteroides* is weakly acid-fast. The meaning of the term “weakly” is that if the acid-fast staining process uses a weaker solution of hydrochloric acid to decolorize than that used in the stain for Mycobacteria, then *N. asteroides* will not decolorize. However, if the regular strength hydrochloric acid is used, *N. asteroides* will decolorize.

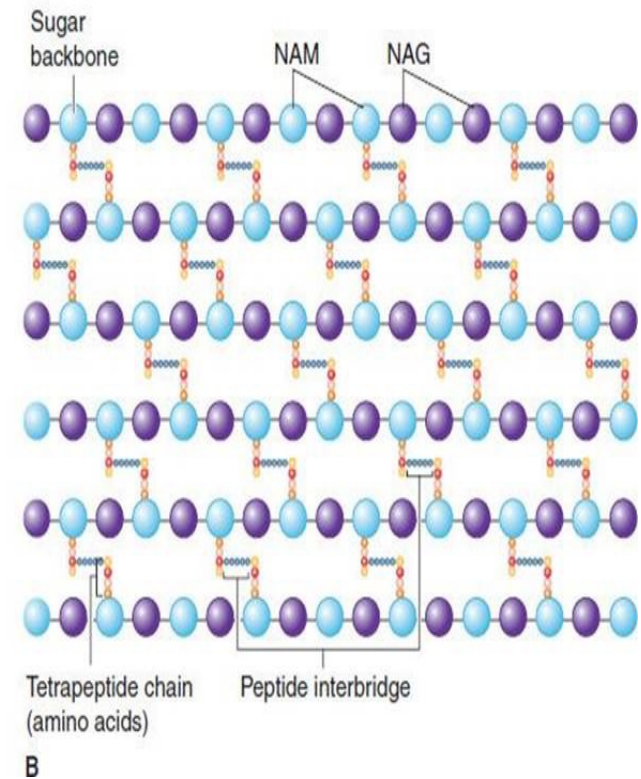
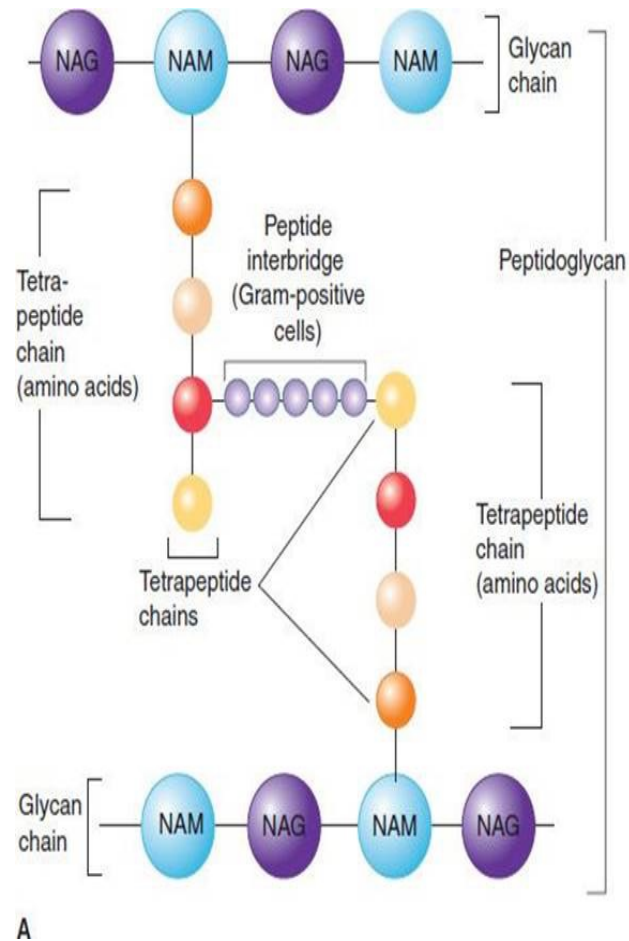
Peptidoglycan layer:

- Glycan chain

N-acetylmuramic acid and
N-acetylglucosamine molecules.

- Peptide chain

Tetrapeptide chain, and
Peptide interbridge

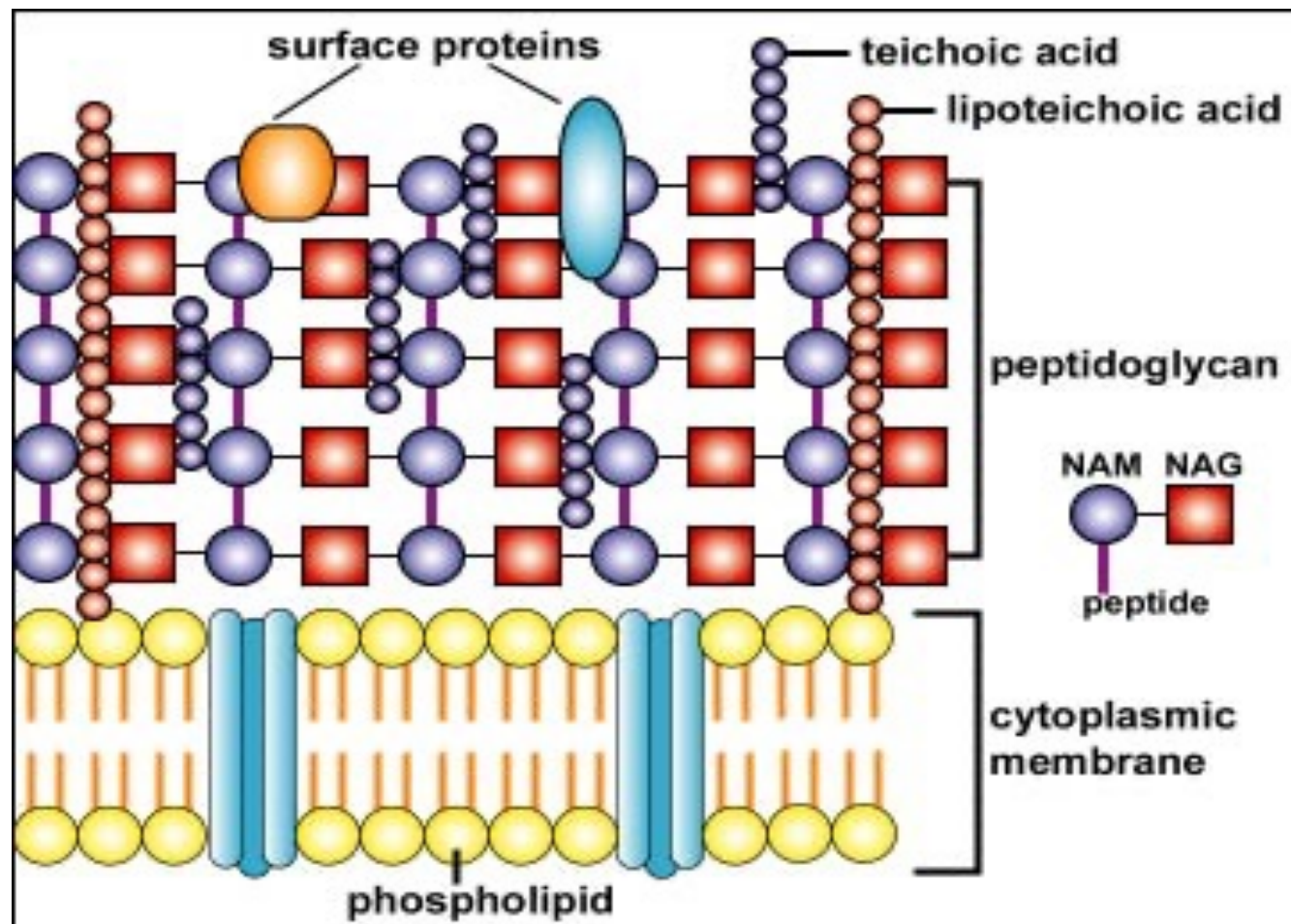


Function of peptidoglycan layer :

- Gives rigid support,
- maintaining the characteristic shape of the cell,
- and allows the cell to withstand media of low osmotic pressure, such as water.
- Site of action of penicillin and cephalosporins and is degraded by lysozyme.

Teichoic Acid

- Teichoic acids are fibers located in the outer layer of the **gram-positive** cell wall and extend from it.
- They are composed of polymers of either glycerol phosphate or ribitol phosphate. Some polymers of glycerol teichoic acid penetrate the peptidoglycan layer and are covalently linked to the lipid in the cytoplasmic membrane, they are called lipoteichoic acid; others anchor to the muramic acid of the peptidoglycan.

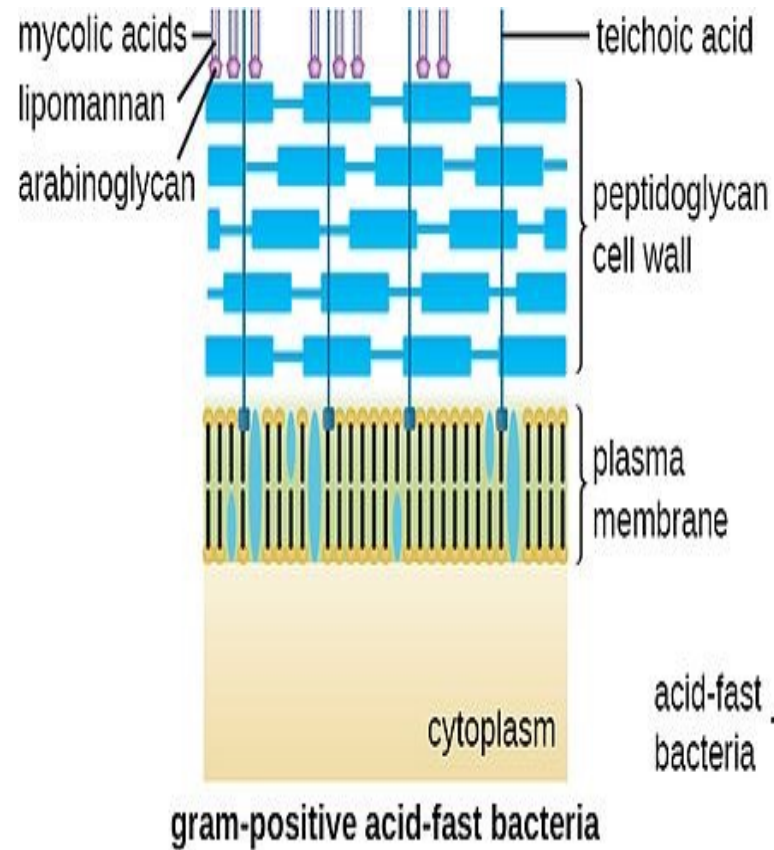


Function of teichoic acid:

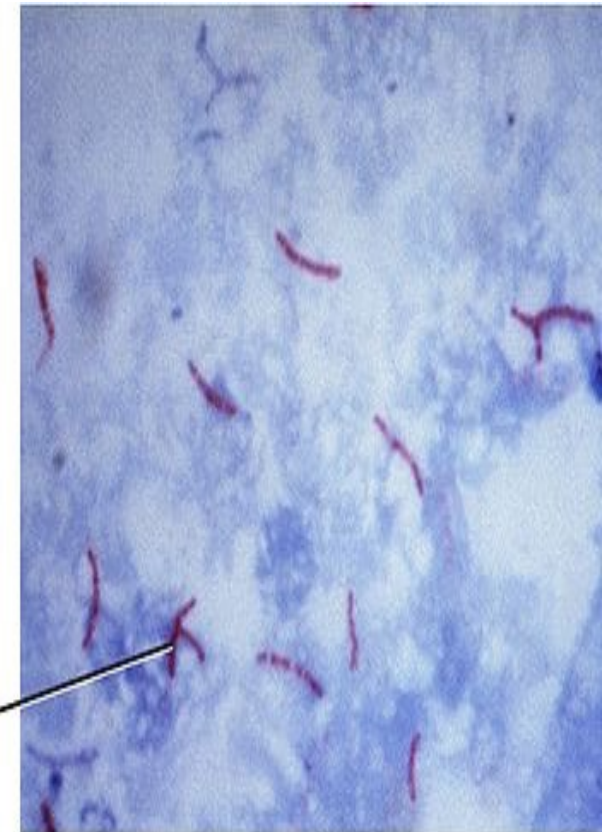
- May induce **inflammation** and **septic shock** when caused by certain gram-positive bacteria; that is, they activate the same pathways as does endotoxin (LPS) in gram-negative bacteria.
- Teichoic acids also mediate the attachment of staphylococci to mucosal cells.
- Teichoic acid Major **surface antigen** but rarely used in laboratory diagnosis.

- **Cell Walls of Acid-Fast Bacteria:**

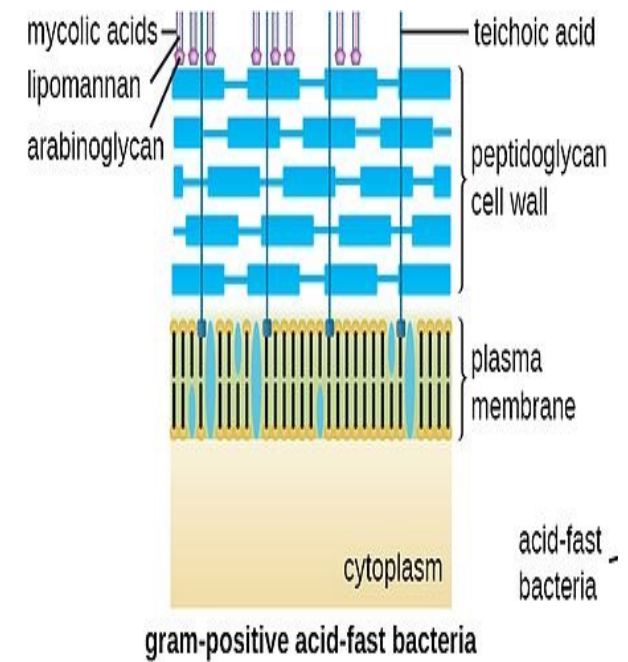
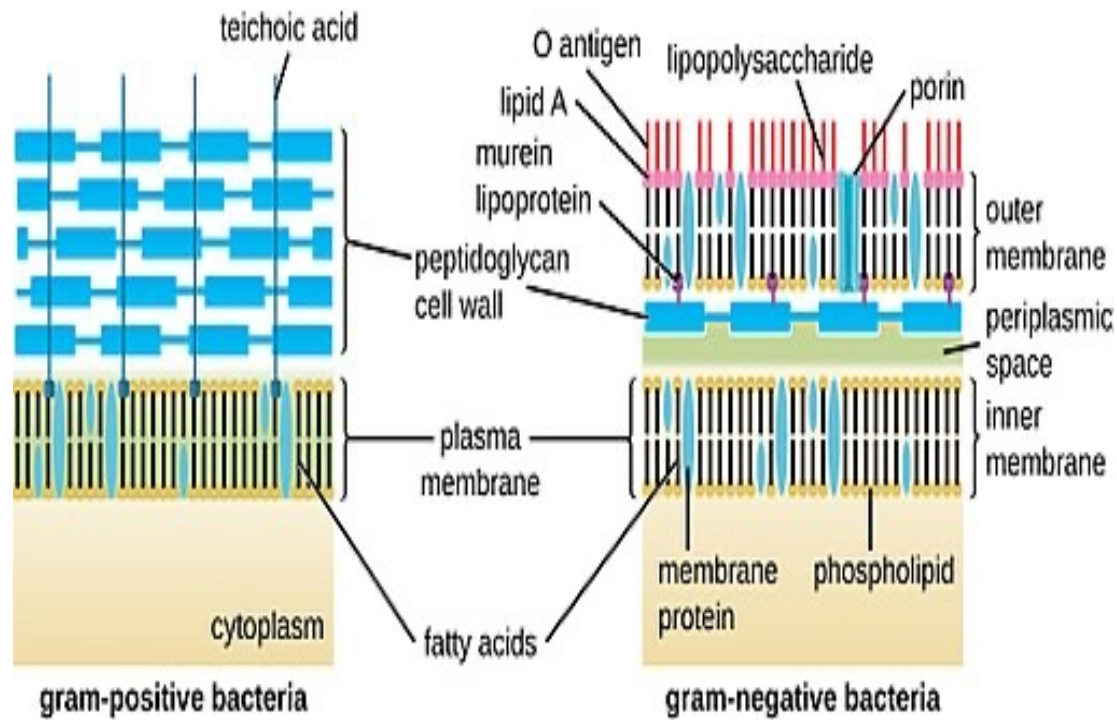
Mycobacteria (e.g., *Mycobacterium tuberculosis*) have an unusual cell wall, resulting in their inability to be Gram-stained because they resist decolorization with acid-alcohol after being stained with carbol fuchsin. This property is related to the high concentration of lipids, called mycolic acids.



(a)



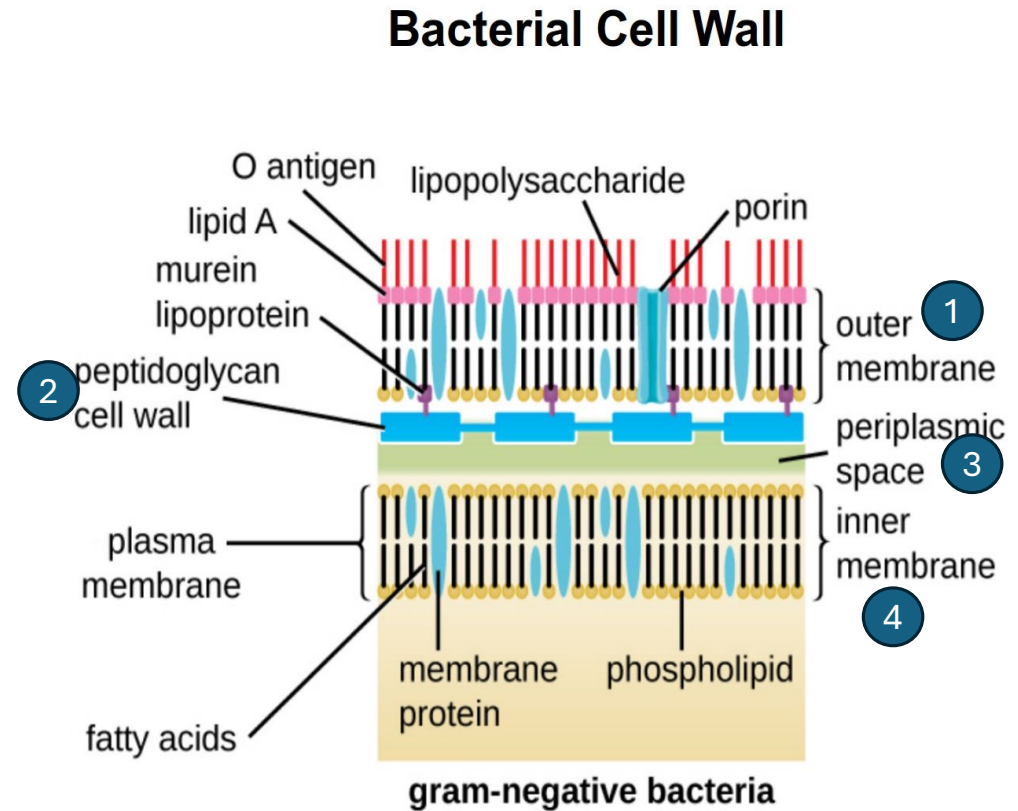
(b)



Gram negative

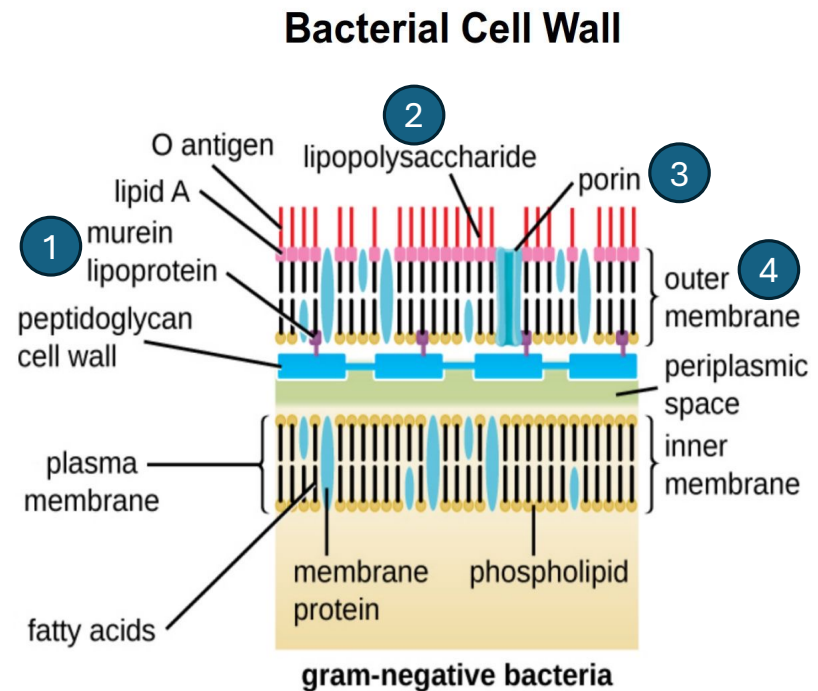
Gram negative

1. Outer membrane
2. Peptidoglycan
3. Periplasmic space
4. Inner membrane

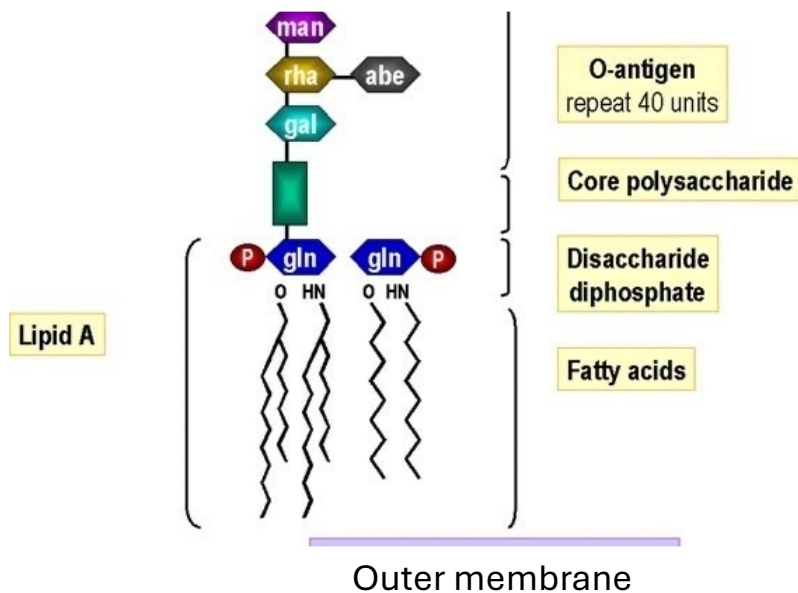


Outer membrane

- The outer membrane is present in gram negative bacteria that surround the cell wall of bacteria.
- This membrane is composed of :
 - lipoprotein
 - lipopolysaccharides
 - Porin
 - phospholipids.



Lipopolysaccharide



- A phospholipid called lipid A
- A core polysaccharide of five sugars linked through ketodeoxyoctulonate (KDO) to lipid A.
- An outer polysaccharide consisting of up to 25 repeating units of three to five sugars that is somatic, or O, antigen.

(Some bacteria, notably members of the genus *Neisseria*, have an outer lipooligosaccharide (LOS) containing very few repeating units of sugars).

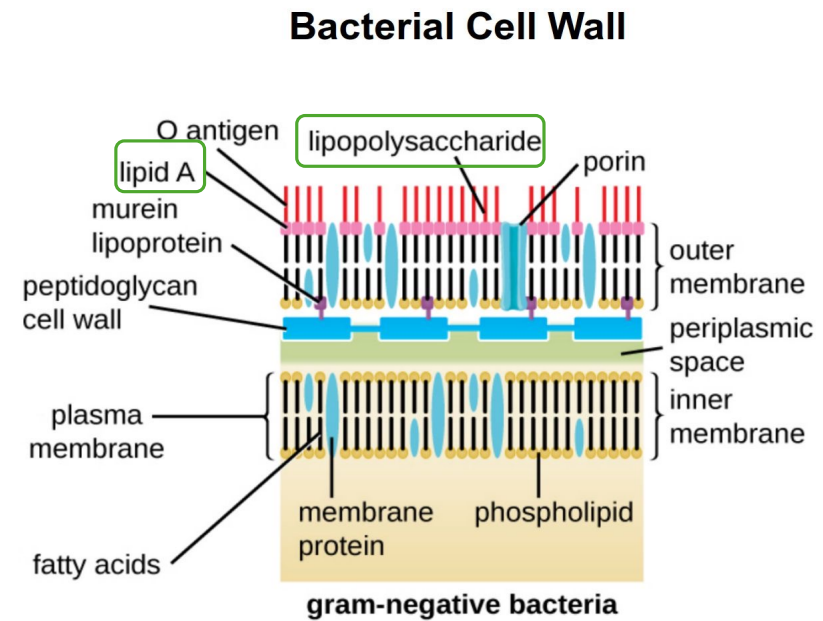
Function of lipopolysaccharide:

- **Lipid A :**

Toxic component of endotoxin

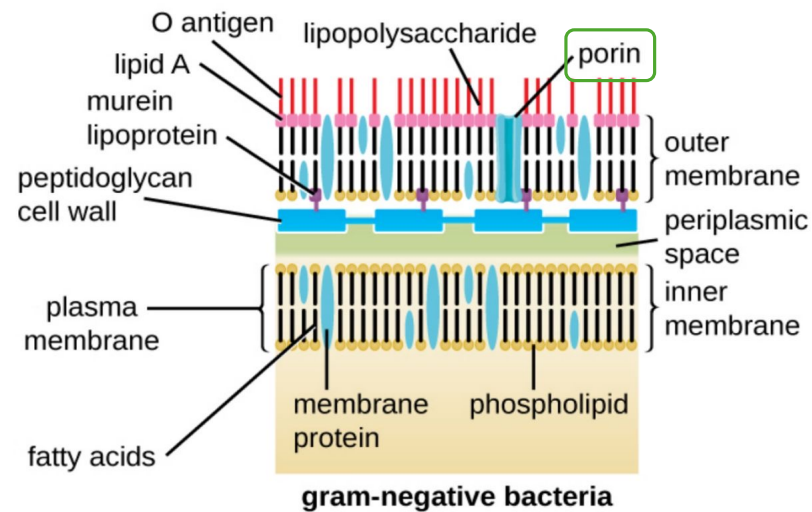
- **Polysaccharide :**

Major surface antigen used frequently in laboratory diagnosis.



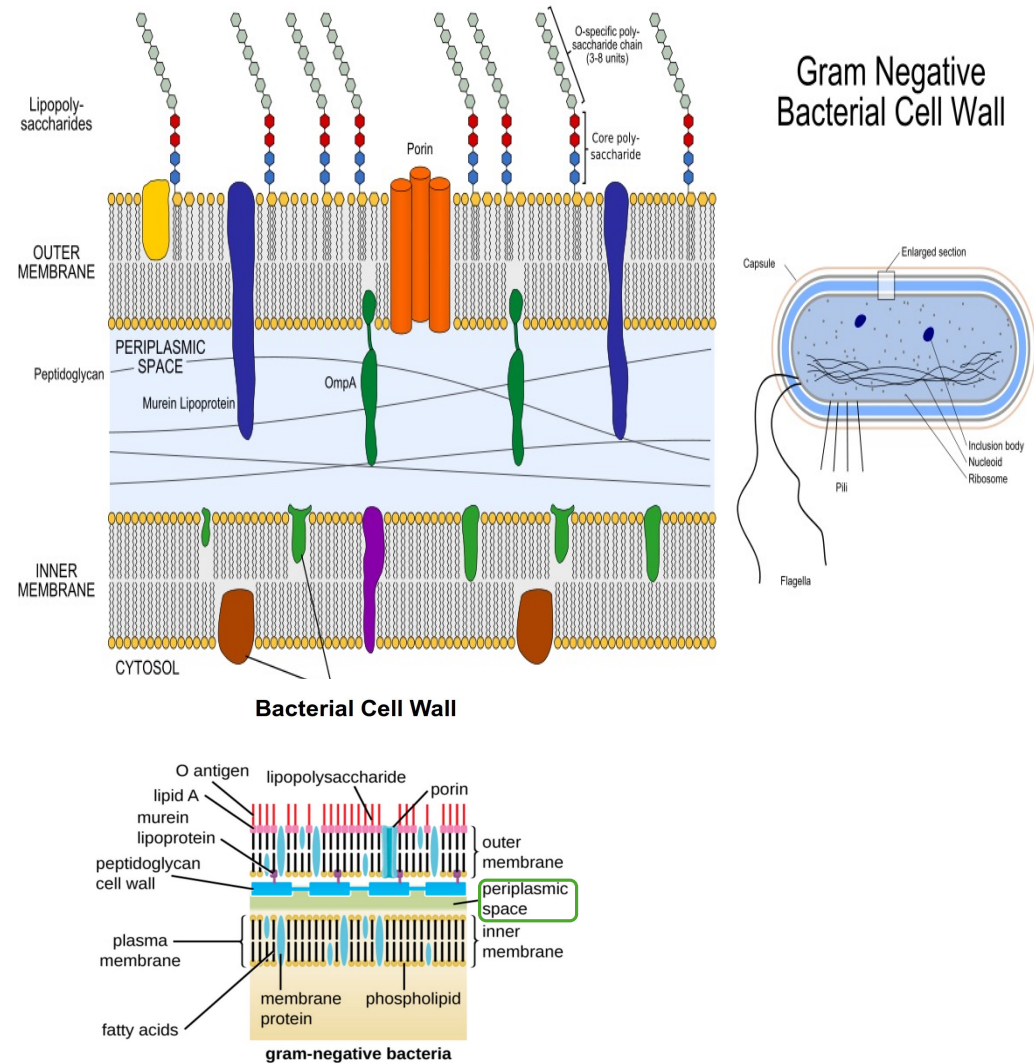
- **Porin proteins** in the outer membrane of gram-negative bacteria act **as a channel** to allow the entry of essential substances such as sugars, amino acids, vitamins, and metals as well as many antimicrobial drugs such as penicillins.

Bacterial Cell Wall



Periplasmic space:

- Between the outer-membrane layer and the cytoplasmic membrane in gram-negative bacteria is the periplasmic space, which is the site, in some species, of enzymes called β -lactamases that degrade penicillins and other β -lactam drugs.
- The periplasmic space is approximately 20–40% of the cell volume. It contains the peptidoglycan layer and a gel-like solution of proteins.



- **Contents:**

- binding proteins for specific substrates (eg, amino acids, sugars, vitamins, and ions),
- hydrolytic enzymes (eg, alkaline phosphatase and 5'-nucleotidase),
- detoxifying enzymes (eg, β -lactamase and aminoglycoside-phosphorylase).
- membrane-derived oligosaccharides.

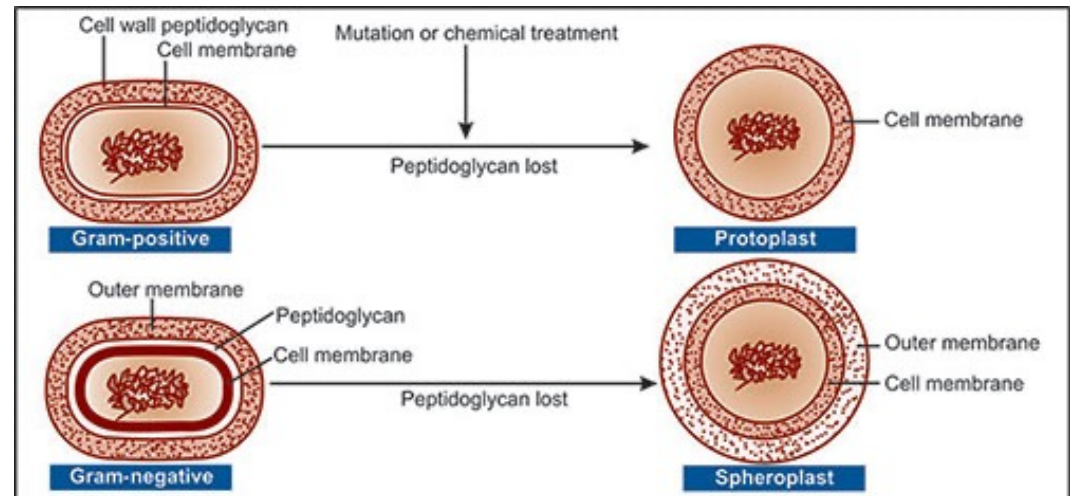
L-form bacteria \ L-phase bacteria \ L-phase variants \ cell wall-deficient (CWD) bacteria

- Bacteria that their peptidoglycan is destroyed or lost by various factors but they can survive under highly osmotic environment.

- Type

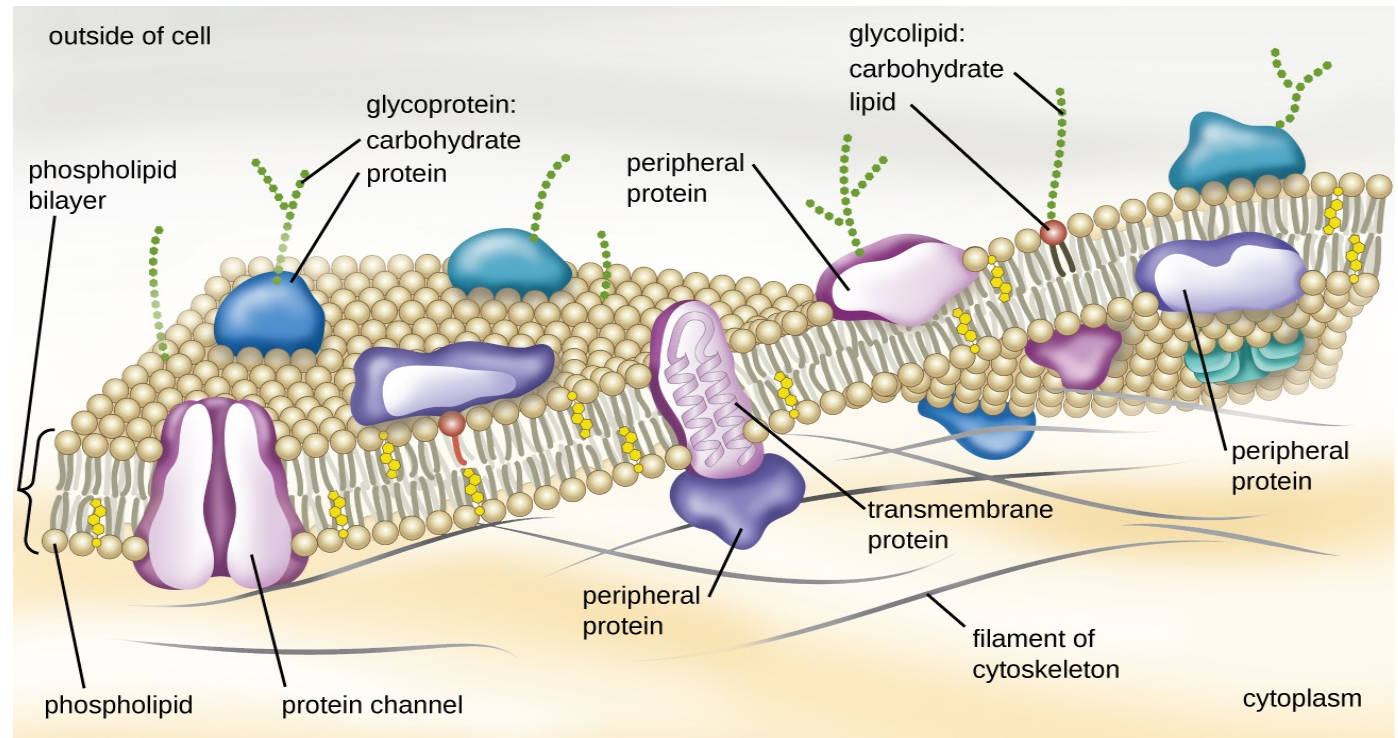
Protoplast: G + bacteria

Spheroplast: G - bacteria



Cytoplasmic Membrane

- Composed of a phospholipid bilayer similar to eukaryotic cells.
- They are chemically similar, but eukaryotic membranes contain sterols, whereas prokaryotes generally do not.
- The only prokaryotes that have **sterols** in their membranes are members of the genus *Mycoplasma*.

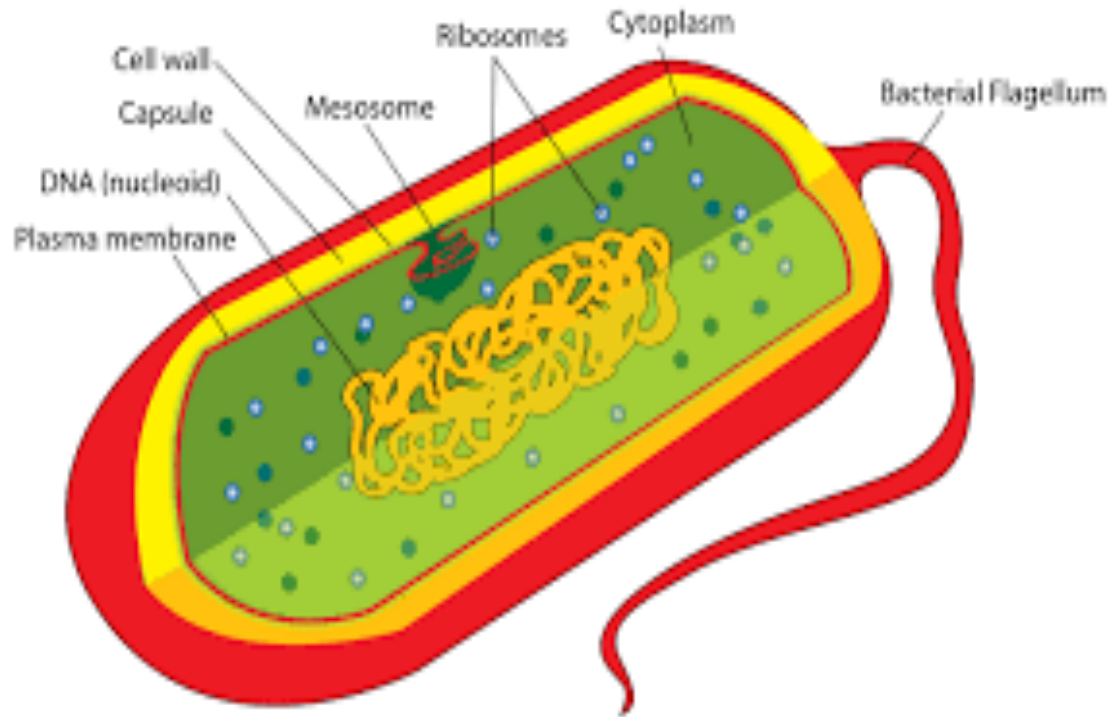


The membrane has four important functions:

- (1) Semipermeable membrane
- (2) active transport of molecules into the cell
- (3) energy generation by oxidative phosphorylation,
- (4) synthesis of precursors of the cell wall, and
- (5) secretion of enzymes and toxins.

Cytoplasm

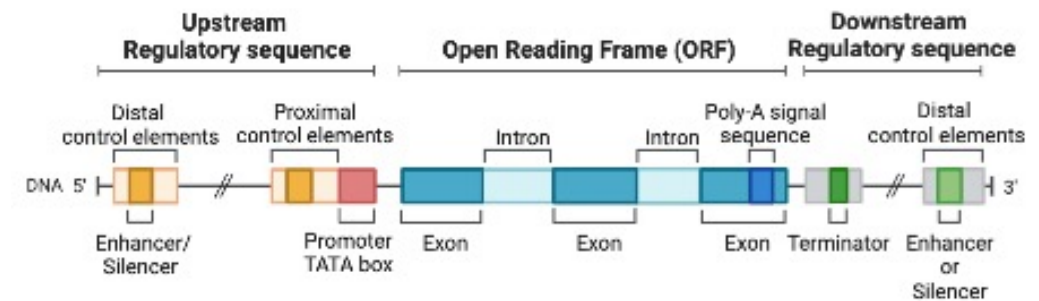
- matrix that contains ribosomes, nutrient granules, metabolites, and plasmids.
- An inner, nucleoid region composed of DNA.



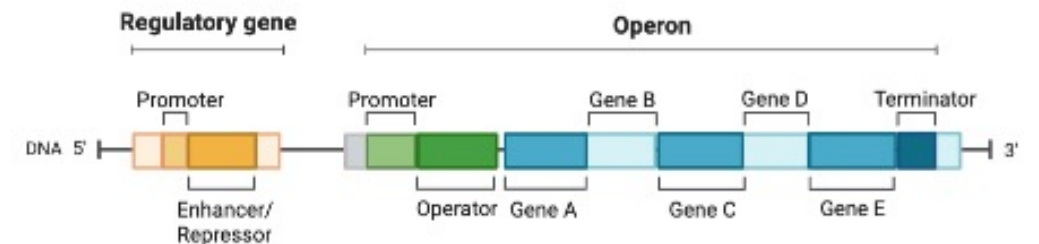
Nucleoid

- The nucleoid is the area of the cytoplasm in which DNA is located.
- It is a single, circular molecule, contains **no nuclear membrane**, no nucleolus, **no mitotic spindle**, and **no histones**.
- One major difference between bacterial DNA and eukaryotic DNA is that bacterial DNA has **no Introns**, whereas eukaryotic DNA does.

Eukaryotic Gene Structure

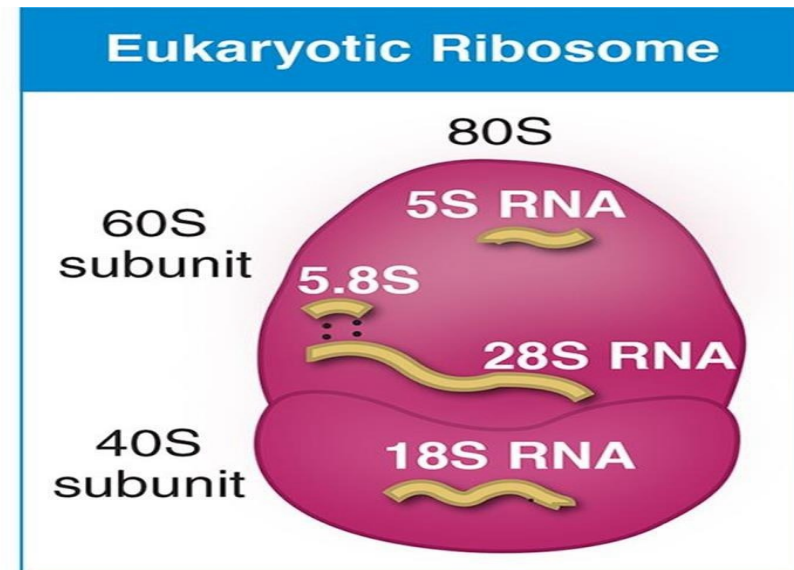
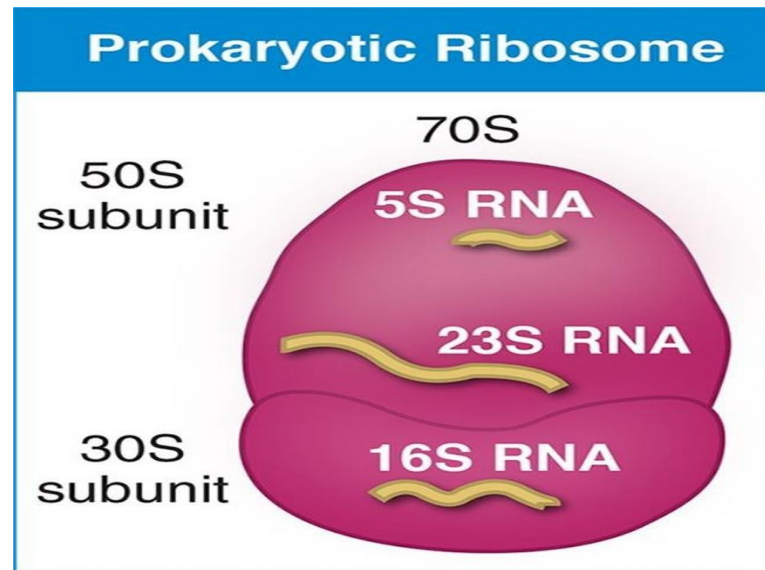


Prokaryotic Gene Structure



Ribosomes

- Bacterial ribosomes are 70S in size, with 50S and 30S subunits, whereas eukaryotic ribosomes are 80S in size, with 60S and 40S subunits.

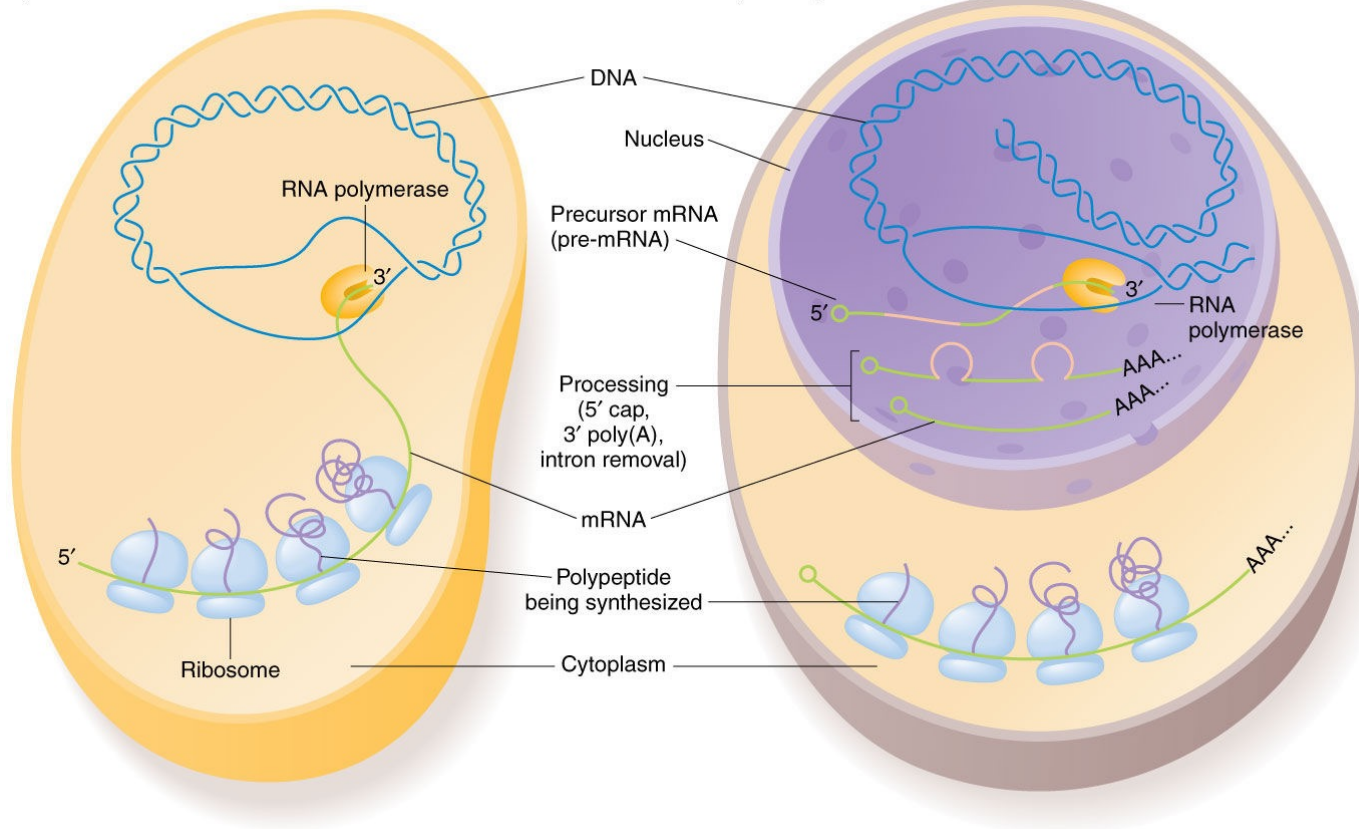


Functions:-

- Site of protein synthesis.
- Site of selective action of several antibiotics that inhibit bacterial, but not human, protein synthesis.

a) Bacterium

b) Eukaryote

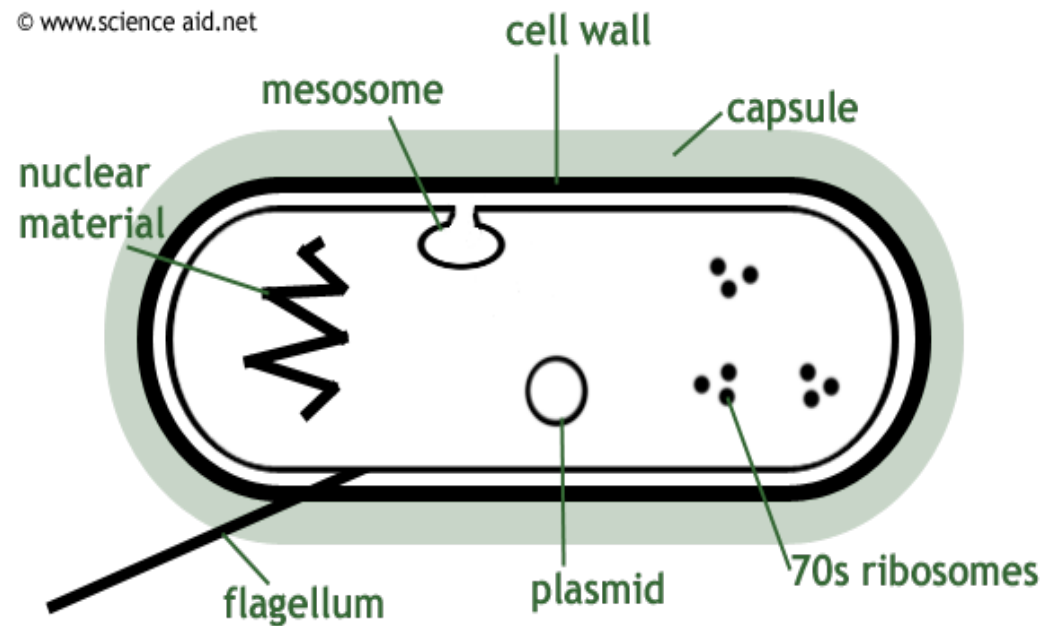


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Prokaryote cell nucleoid: Prokaryote cell (right) showing the nucleoid in comparison to a eukaryotic cell (left) showing the nucleus.

Mesosomes

- Mesosomes are specialized structures formed by convoluted invaginations of cytoplasmic membrane.
- Types:
 - septal mesosome
 - lateral mesosome.



- **Functions-**

- Helps in the synthesis of the cell wall
- Helps in replication of DNA.
- They also help in the equal distribution of chromosomes into the daughter cells.

Non-essential component:

Capsule

Pili

Flagella

Spore

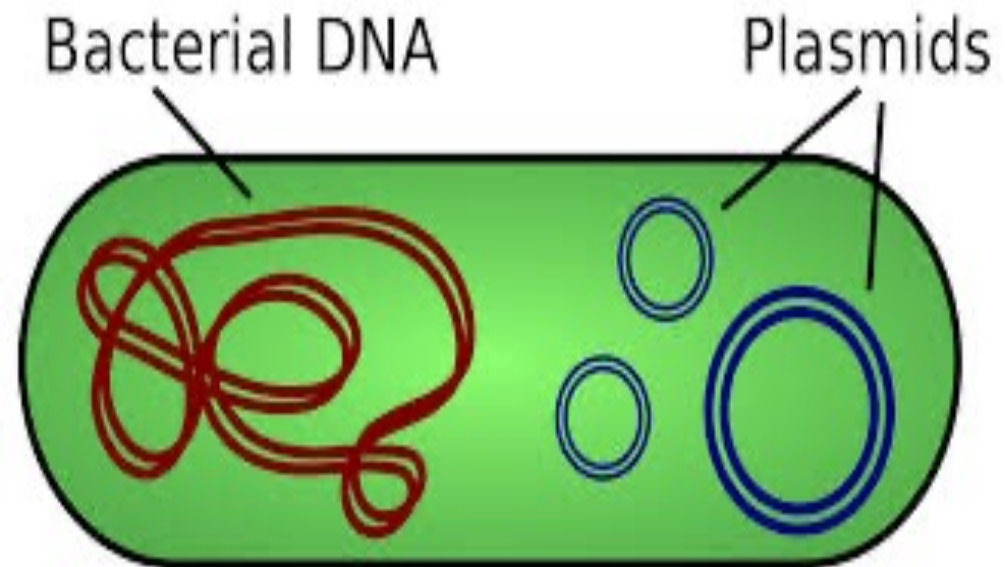
Plasmid

Glycocalyx

Granules

Plasmids

- Plasmids are extrachromosomal, double-stranded, circular DNA molecules that are capable of replicating independently of the bacterial chromosome.
- Although plasmids are usually extrachromosomal, they can be integrated into the bacterial chromosome.



Types of plasmid

Transmissible plasmids

- Can be transferred from cell to cell by conjugation.
- They are large
- They contain about a dozen genes responsible for synthesis of the sex pilus and for the enzymes required for transfer.
- They are usually present in a few (1–3) copies per cell.

Nontransmissible plasmids

- They are small
- They do not contain the transfer genes
- They are frequently present in many (10–60) copies per cell.

Functions

Plasmids carry the genes for :

- (1) Antibiotic resistance, which is mediated by a variety of enzymes, such as the beta-lactamase of *S. aureus*,
- (2) Exotoxins, such as the enterotoxins of *E. coli*.
- (3) Pili (fimbriae), which mediate the adherence of bacteria to epithelial cells.
- (4) Resistance to heavy metals, such as mercury, the active component of some antiseptics (e.g., merthiolate and mercurochrome).
- (5) Resistance to ultraviolet light, which is mediated by DNA repair enzymes.

Bacteriocins

Bacteriocins are toxic proteins produced by certain bacteria that are lethal for other bacteria.

Two common mechanisms of action of bacteriocins are

- (i) degradation of bacterial cell membranes by producing pores in the membrane and
- (ii) degradation of bacterial DNA by DNase.

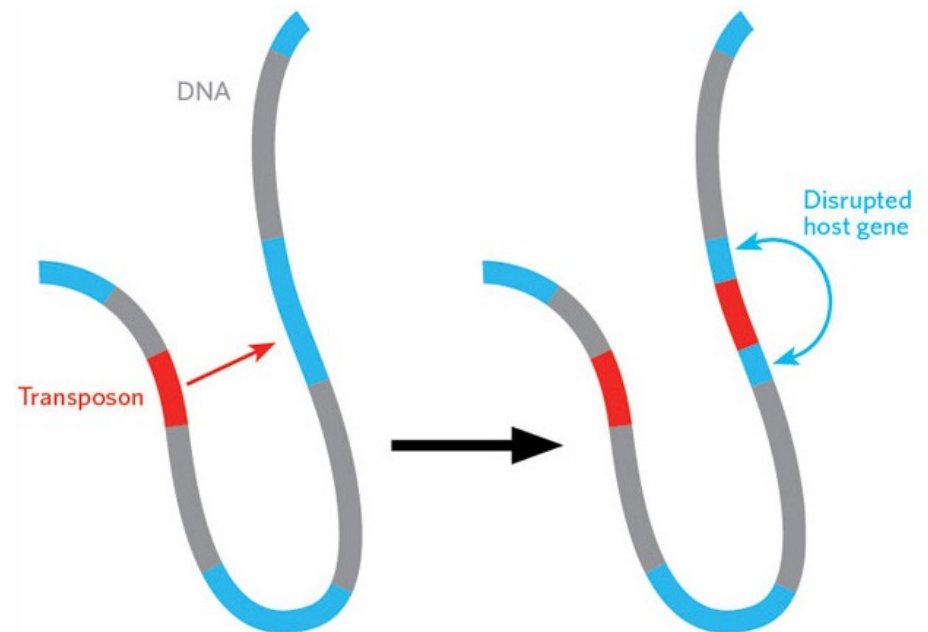
Examples of bacteriocins produced by medically important bacteria are **colicins** made by *E. coli* and **pyocins** made by *Pseudomonas aeruginosa*.

Function:

- Bacteria that produce bacteriocins have a selective advantage in the competition for food sources over those that do not.
- they may be useful in treating infections caused by antibiotic-resistant bacteria.

Transposons

- Transposons are pieces of DNA that move readily from one site to another either within or between the DNAs of bacteria, plasmids, and bacteriophages.
- Because of their unusual ability to move, they are nicknamed “jumping genes.”



Typs of transpositions

- **Direct transposition:**

Whereas others are excised from the site without replicating and then inserted into the new site.

- **Replicative transposition:**

Some transposons move by replicating their DNA and inserting the new copy into another site.

Two methods of transposition:

1. Cut-and-paste mechanism



2. Copy-and-paste mechanism

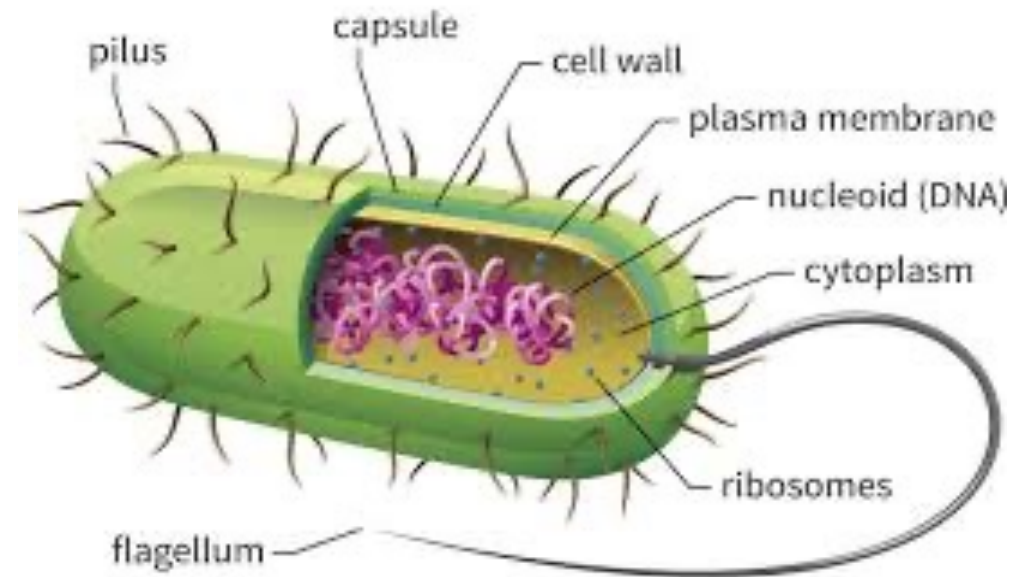


Functions

- Transposons can code for drug resistant enzymes, toxins, or a variety of metabolic enzymes and
- can either cause mutations in the gene into which they insert.
- (In contrast to plasmids or bacterial viruses, transposons are not capable of independent replication; they replicate as part of the DNA in which they are integrated)

Capsule

- The capsule is a gelatinous layer covering the entire bacterium.
- It is composed of polysaccharide, except in the anthrax bacillus, which has a capsule of polymerized d-glutamic acid.

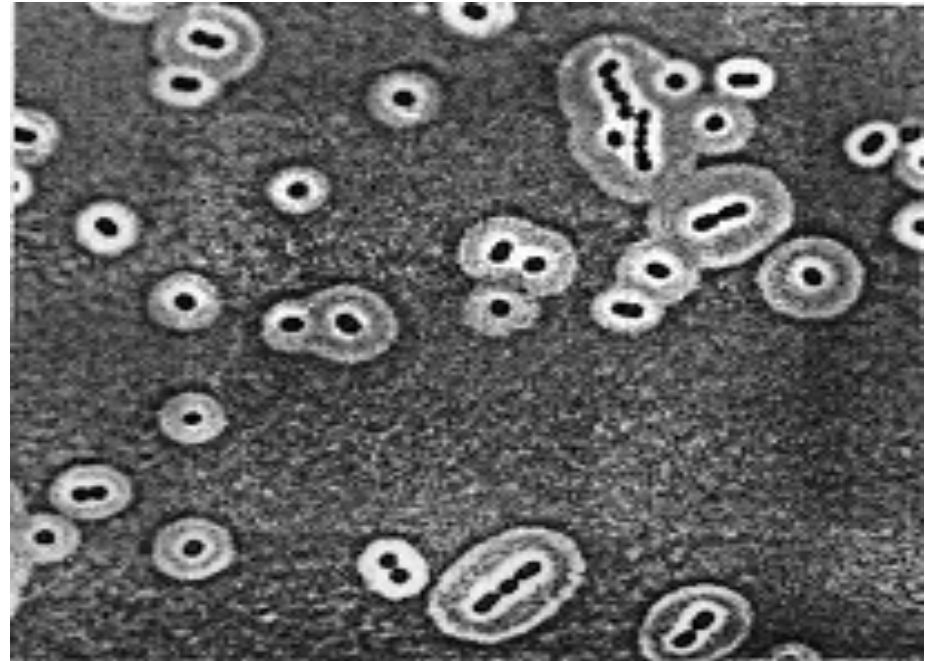


Functions of capsule

- (1) It **prevents phagocytosis**. Negative charges on the capsular polysaccharide repel the negatively charged cell membrane of the neutrophil and prevent it from ingesting the bacteria.
- (2) Specific identification of an organism can be made by using anti-serum against the capsular polysaccharide.
- (3) Capsular polysaccharides are used as the antigens in certain vaccines because they are capable of eliciting protective antibodies. For example, the purified capsular polysaccharides of 23 types of *S. pneumoniae* are present in the current vaccine.
- (4) The capsule may play a role in the adherence of bacteria to human tissues, which is an important initial step in causing infection.

Capsulated bacteria

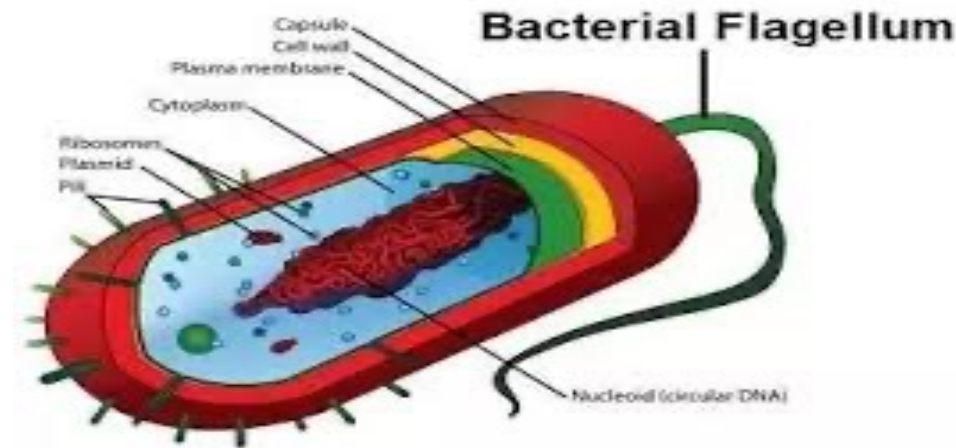
- *Streptococcus pneumoniae*
- *Klebsiella pneumoniae*
- *Bacillus anthracis*
- *Neisseria meningitidis*
- *Haemophilus influenza*



**Bacterial capsules outlined by India ink
viewed by light microscopy**

Flagella

- Flagella are long, whiplike appendages that move the bacteria toward nutrients and other attractants, a process called chemotaxis.
- composed of many subunits of a single protein, flagellin.



Types of flagella

- Monotrichous: Single polar flagellum e.g., *Vibrio cholera*.
- Amphitrichous: Single flagellum attached to each end, e.g., *Alkaligenes faecalis*
- Lophotrichous : Multiple polar flagella at one or both ends e.g., *Helicabacter pylori*,
- Peritrichous: Flagella distributed over the entire cell, e.g., *Salmonella*

ARRENGEMENT OF FLAGELLA



(a) Peritrichous

SEM 0.5 μ m



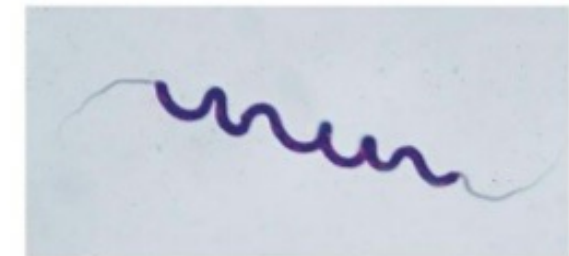
(b) Monotrichous and polar

SEM 0.5 μ m



(c) Lophotrichous and polar

SEM 0.5 μ m



(d) Amphitrichous and polar

SEM 5 μ m

Pili (Fimbriae)

- Pili are hair like filaments that extend from the cell surface.
- They are composed protein called pilin.
- They are shorter and thinner and flagella but more numerous than flagella.
- E.g., *Proteus*, *Shigella*, *Salmonella*

Type

- Common pili
 - Organ of adhesion
- Sex pili \ fertility pili
 - Transfer of genetic material from one to another.

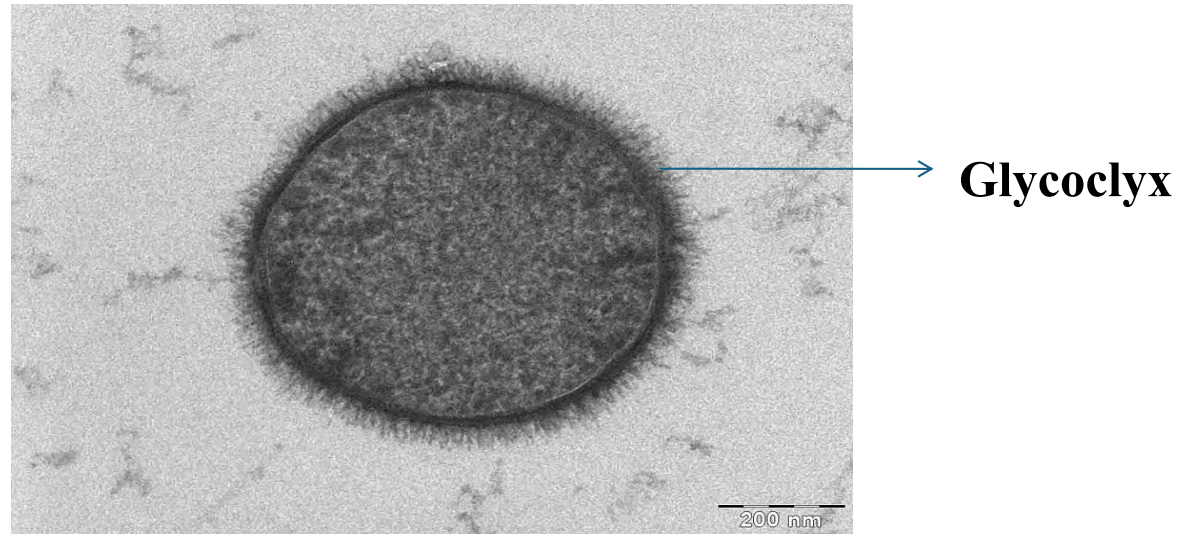


3D illustration of a pseudomonas aeruginosa bacteria.

It is antigenic and so helps in diagnosis of certain disease.

Glycocalyx (Slime Layer)

- The glycocalyx is a polysaccharide coating that is secreted by many bacteria.



The Bacterium *Bacillus subtilis* taken with a TEM.

Functions

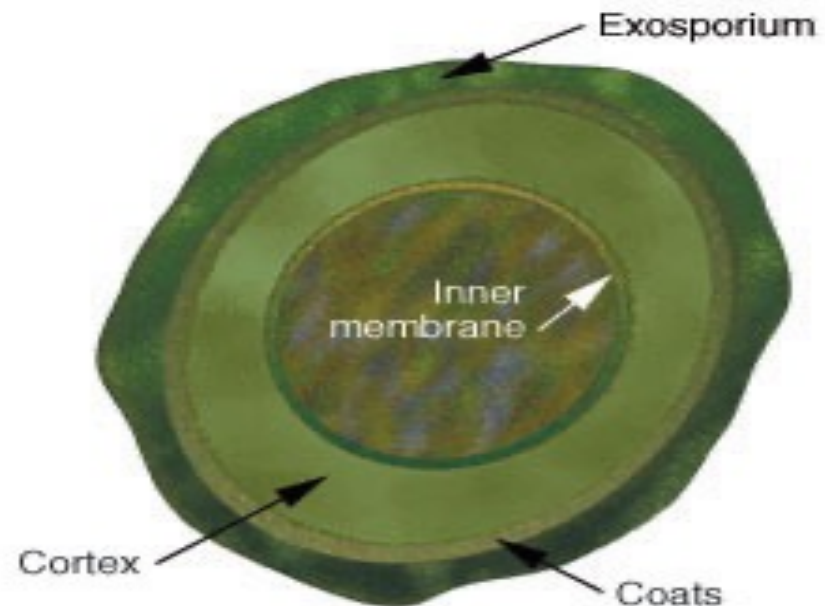
- It covers surfaces like a film and allows the bacteria to adhere firmly to various structures (e.g., skin, heart valves, prosthetic joints, and catheters). The glycocalyx is an important component of **biofilms**.
- The glycocalyx also mediates adherence of certain bacteria, such as *Streptococcus mutans*, to the surface of teeth. This plays an important role in the formation of **plaque**, the precursor of dental caries.

Bacterial Spores

- Spores are highly resistant dormant stage of bacteria formed in unfavourable environmental conditions such as starvation and desiccation.

Spores are resistance than vegetative form

- Presence of calcium dipicolinate in core
- Reduced metabolic activity
- Spore coat is impermeable to antibiotics and
- Due to their dehydrated state.



Functions of Spores

- Highly resistant to heating; spores are not killed by boiling (100°C), but are killed at 121°C.
- Highly resistant to many chemicals, including most disinfectants, due to the thick, keratinlike coat of the spore.
- They can survive for many years, especially in the soil.
- They exhibit no measurable metabolic activity.
- Spores form when nutrients are insufficient but then germinate to form bacteria when nutrients become available.
- Spores are produced by members of only two genera of bacteria of medical importance, *Bacillus* and *Clostridium*, both of which are gram positive rods.

Their Medical Implications

- Medical supplies must be heated to 121°C for at least 15 minutes to be sterilized.
- Only solutions designated as sporicidal will kill spores.
- Wounds contaminated with soil can be infected with spores and cause diseases such as **tetanus** (*C. tetani*) and **gas gangrene** (*C. perfringens*).
- Antibiotics are ineffective against spores because antibiotics act by inhibiting certain metabolic pathways of bacteria. Also, spore coat is impermeable to antibiotics.
- Spores are not often found at the site of infections because nutrients are not limiting. Bacteria rather than spores are usually seen in Gram-stained smears.
- Infections transmitted by spores are caused by species of either **Bacillus or Clostridium**.

TETANUS



tetanus (*C. tetani*)



gas gangrene (*C. perfringens*)

Structure of spore

- 1. Core—
It is the cytoplasm of bacteria.
It contains a complete nucleus , enzymes
- 2. Spore wall—
It contains normal peptidoglycan and becomes the cell wall of the germinating vegetative cell.
- 3. Cortex—thickest layer of the spore.
It contains an unusual type of peptidoglycan
- 4. Coat—The coat is composed of a keratin-like protein. Protects bacteria from antibiotics.
- 5. Exosporium—The exosporium is composed of proteins, lipids, and carbohydrates.

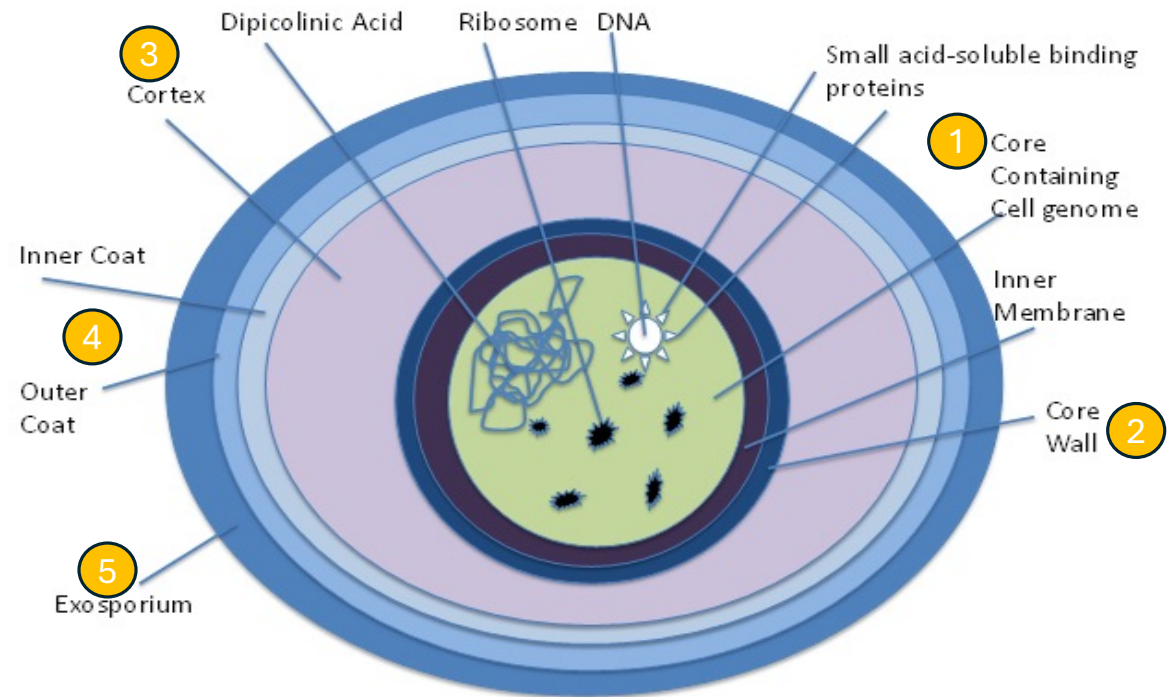


Fig 6 Bacterial spore

Spore forming bacteria

Gram positive rods:

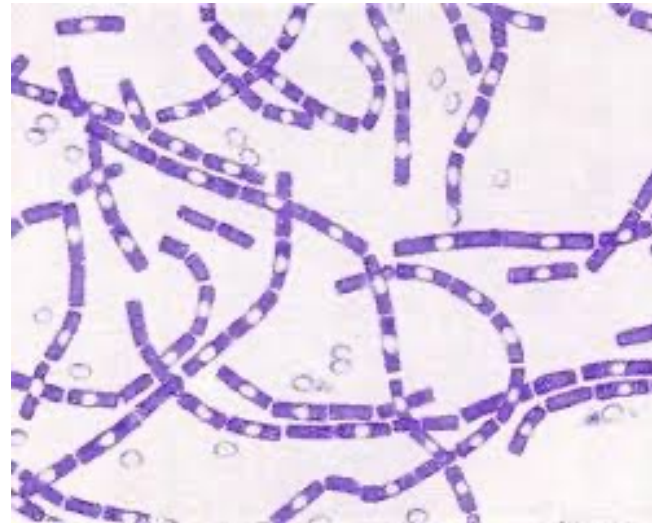
- *Clostridium*
- *Bacillus*

Gram positive cocci:

- *Sporosarcina*

Gram negative rod:

- *Coxiella burnetii*



Bacillus sp.



Coxiella burnetii

A microscopic image of various bacteria, primarily rod-shaped, with some showing flagella. The bacteria are stained and appear in shades of blue and grey against a lighter background. The image is used as a background for the slide.

Classification of bacteria

Basis of classification

- **Phenotypic classification**
 - ❖ **Morphological**
 - ❖ **Anatomical**
 - ❖ **Staining**
 - ❖ **Cultural characteristics**
 - ❖ **Nutrition**
 - ❖ **Environmental factors**
 - ❖ **Biochemical reactions**
 - ❖ **Antigenic structure**
- **Genotypic classification**
 - ❖ **DNA-DNA hybridization**
 - ❖ **G+C content**

• Morphological classification

Bacteria can be classified into **five** major groups on morphological basis.

1. **True Bacteria**

- Cocci- These are spherical or oval cells. On the basis of arrangement of individual organisms they can be described as Monococci(*Monococcus spp.*), Diplococci(*Streptococcus pneumoniae*), Staphylococci(*Staphylococcus aureus*).
- Bacilli- these are the rod shaped bacteria .On the basis of arrangement of organisms they can be described as Diplobacilli, Streptobacilli and Coccobacilli.

2. **Actinomycetes**(actin-ray, mykes-fungus)

These are the rigid organisms like true bacteria but they resemble like fungi in that they exhibit branching and tend to form filaments. They are termed such because of their resemblances to the sunrays when in tissue sections.

3. **Mycoplasmas**

These bacteria are lack in rigid cell wall and are highly pleomorphic having indefinite shape and size.

4. **Spirochaetes**

These are relatively longer, slender, non-branched microorganisms of spiral shape having several coils.

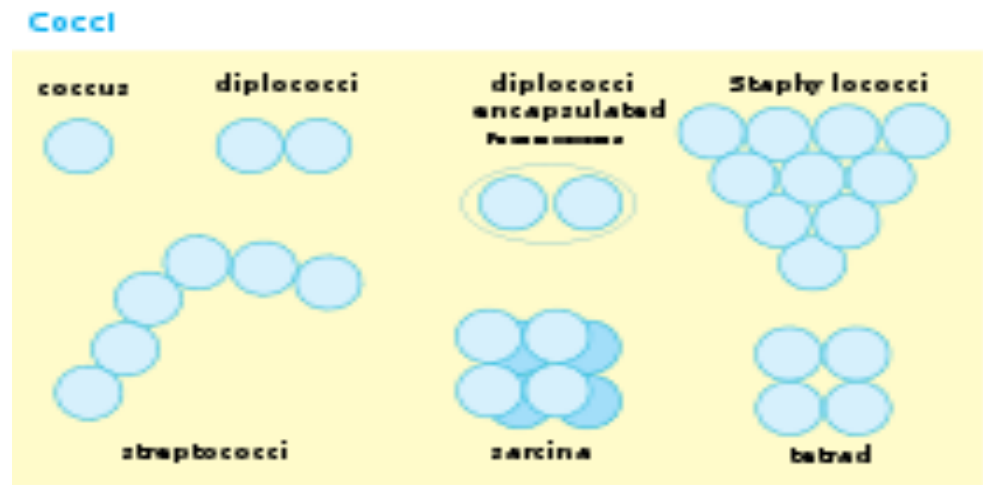
5. **Rickettsiae and Chlamydiae**

These are very small, obligate parasites and at onetime were considered closely related to the viruses. So now these are regarded as bacteria.

Morphological classification

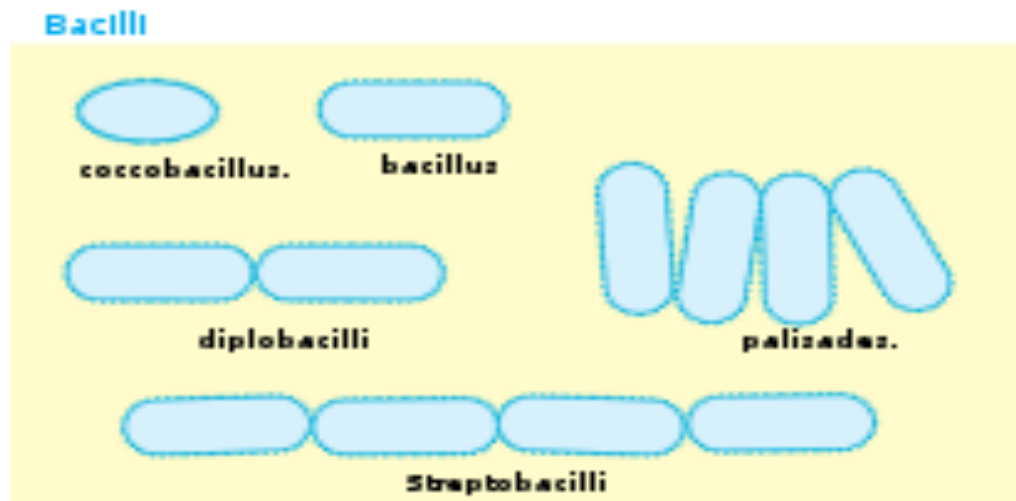
1. TRUE BACTERIA

- **Cocci** – These are spherical or oval cells. On the basis of arrangement of individual organisms they can be described as
 - **Monococci** (Cocci in singles) – *Monococcus* spp.
 - **Diplococci** (Cocci in pairs) – *Streptococcus pneumoniae*
 - **Staphylococci** (Cocci in grape-like clusters) – *Staphylococcus aureus*
 - **Streptococci** (Cocci in chains) – *Streptococcus pyogenes*
 - **Tetrad** (Cocci in group of four) - *Micrococcus* spp.
 - **Sarcina** (Cocci in group of eight)



1. TRUE BACTERIA

- **Bacilli** – These are rod-shaped bacteria. On the basis of arrangement of organisms, they can be described as
 - **Diplobacilli**
 - **Streptobacilli**
 - **Palisades**
 - **Coccobacilli**
 - **Comma-shaped**

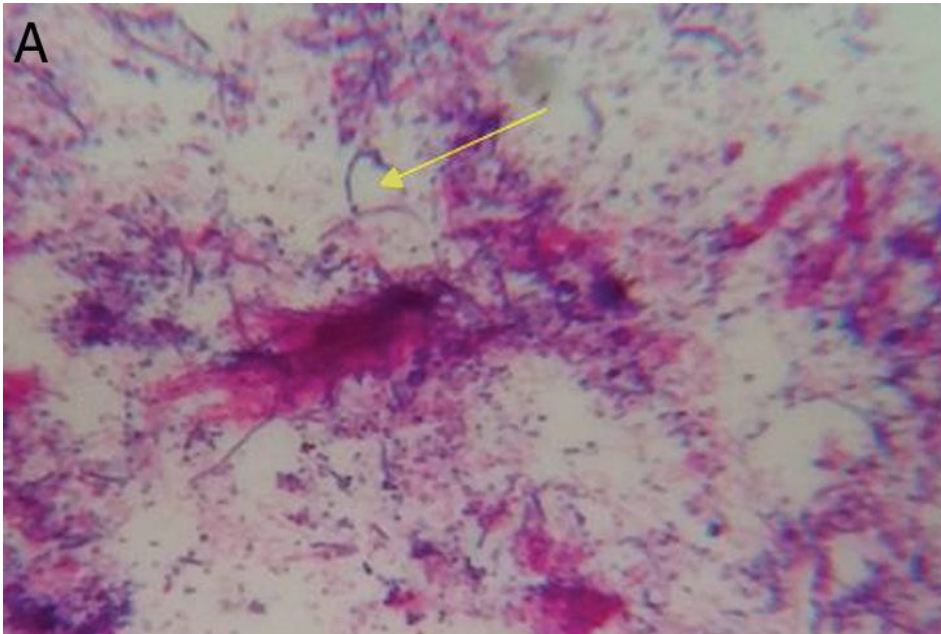


Morphological classification

2. ACTINOMYCETES (actin- ray, mykes-fungus)

These are rigid organisms like true bacteria but they resemble fungi in that they exhibit branching and tend to form filaments.

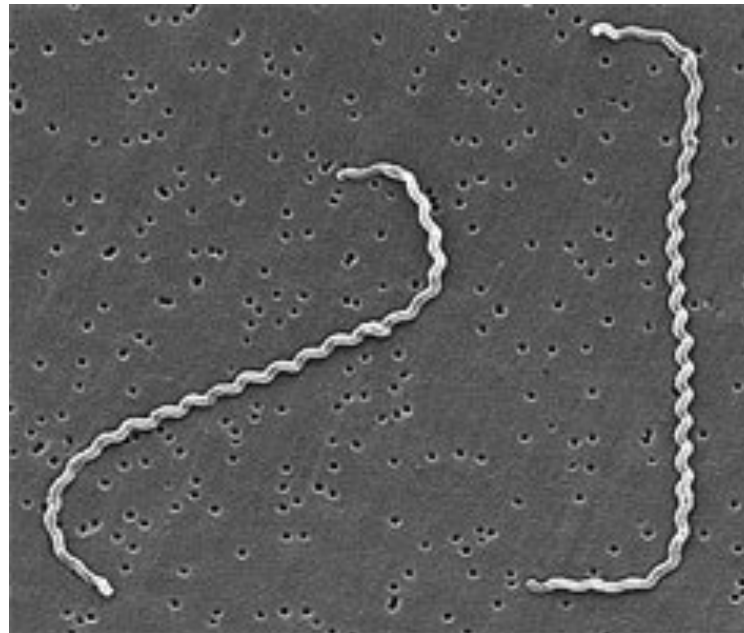
They are termed such because of their resemblance to sun rays when seen in tissue sections.



Morphological classification

3. Spirochaetes

These are relatively longer, slender, non-branched microorganisms of spiral shape having several coils.

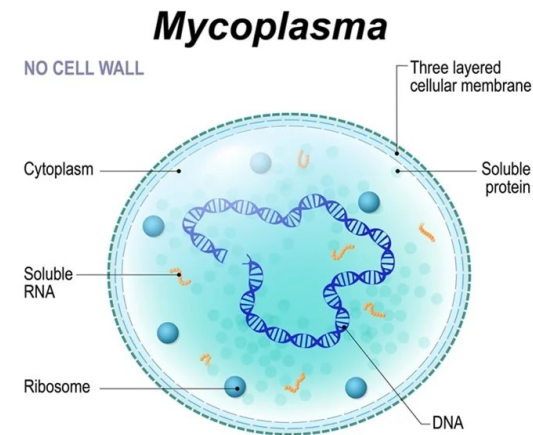


Morphological classification

4. Mycoplasmas

These bacteria lack in rigid cell wall (cell wall lacking) and are highly pleomorphic and of indefinite shape.

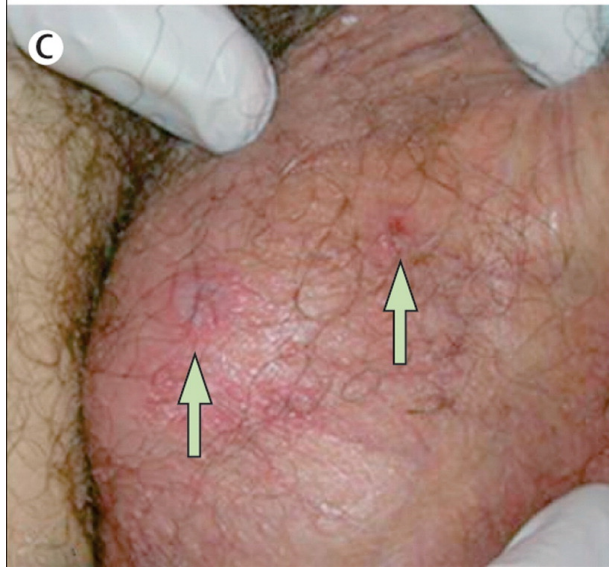
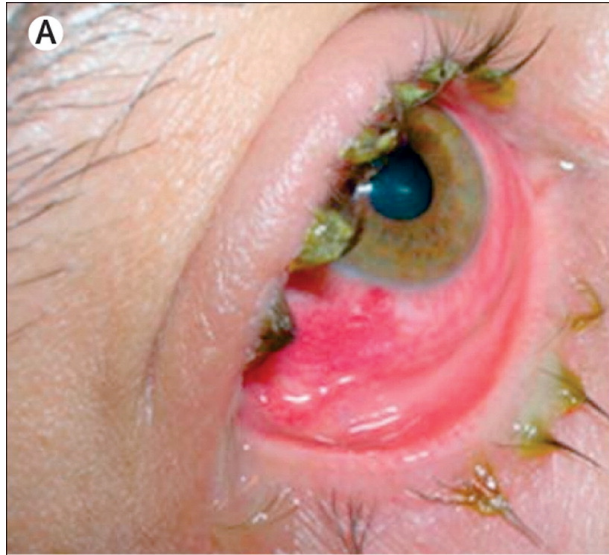
They occur in round or oval bodies and in interlacing filaments.



Diseases Caused by Mycoplasma

<u>Organism</u>	<u>Disease</u>
<i>M. pneumoniae</i>	Upper respiratory tract disease, tracheobronchitis, atypical pneumonia, (chronic asthma??)
<i>M. hominis</i>	Pyleonephritis, pelvic inflammatory disease, postpartum fever
<i>M. genitalium</i>	Nongonococcal urethritis
<i>U. urealyticum</i>	Nongonococcal urethritis, (pneumonia and chronic lung disease in premature infants??)

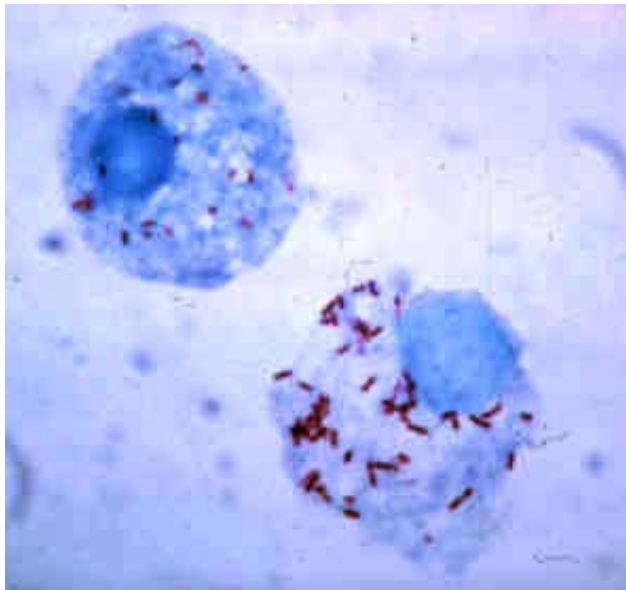
N.B. Other organisms infect humans but their disease association is not known.



Morphological classification

5. Rickettsiae and Chlamydiae

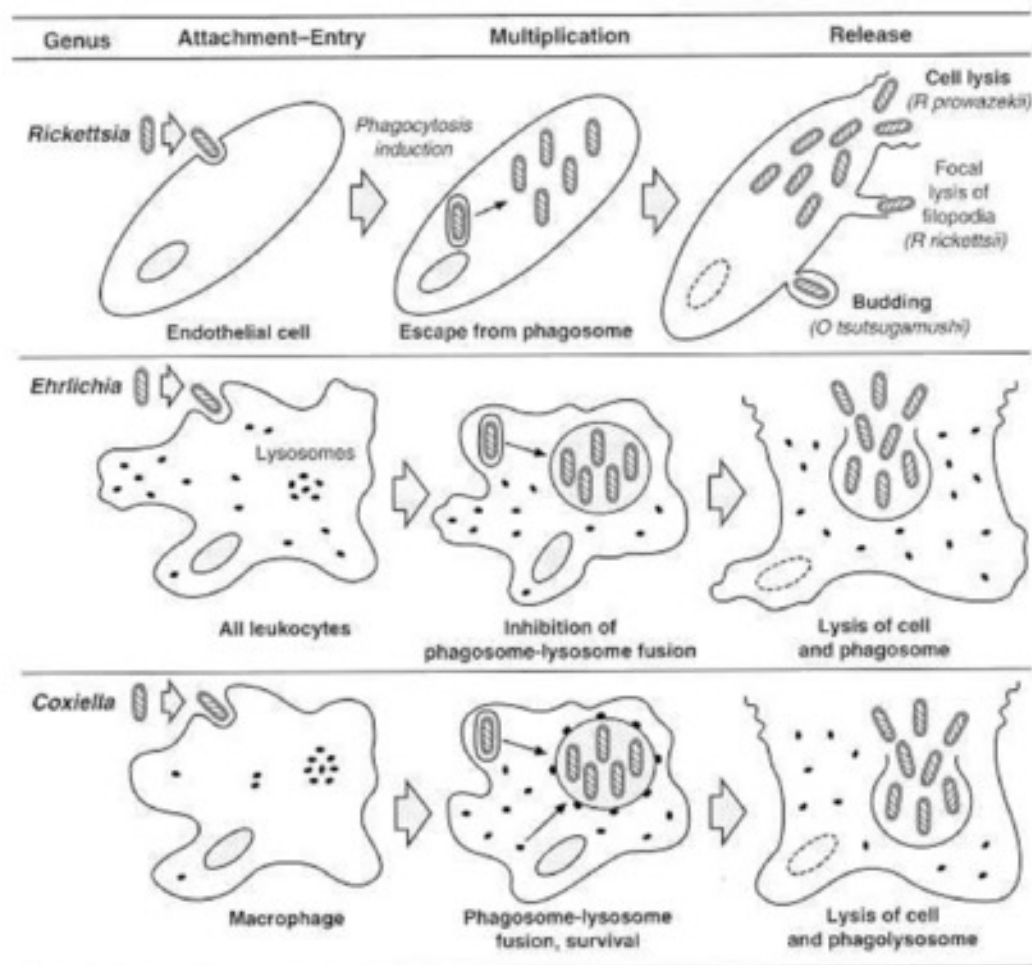
These are very small, obligate parasites, and at one time were considered closely related to the viruses. Now, these are regarded as bacteria.



Rickettsia and Chlamydia

Order	Family	Genus	
Rickettsiales	Rickettsiaceae	<i>Rickettsia</i> <i>Orientia</i>	<i>Vascular endothelium</i>
	Anaplasmataceae	<i>Anaplasma</i>	<i>Erythrocytes, phagocytes, platelets</i>
		<i>Ehrlichia</i>	<i>Phagocytic cells</i>
		<i>Neorickettsia</i>	<i>Macrophages and enterocytes</i>
Chlamydiales	Chlamydiaceae	<i>Chlamydia</i>	<i>Epithelial cells</i>
Orphan	Coxiellaceae	<i>Coxiella</i>	<i>Epithelial cells</i>

Don't really know where they go



Based on Anatomical features

- **Capsule**
 - **Capsulate**– *Streptococcus pneumoniae*
 - **Non-capsulate** – Viridans streptococci
- **Flagella**
 - **Flagellate** –
 - **Monotrichous**
 - **Lophotrichous**
 - **Amphitrichous**
 - **Peritrichous**
 - **Aflagellate** – *Shigella* spp.
- **Spore**
 - **Spore-forming** – *Bacillus* spp.
 - **Non-sporing** – *Escherichia coli*

Based on Staining reaction

- **GRAM'S STAIN**

- **Gram-positive cocci** – *Staphylococcus aureus*
- **Gram-negative cocci** – *Neisseria gonorrhoeae*
- **Gram-positive rods** – *Clostridium* spp.
- **Gram-negative rods** – *E. coli*

- **ACID FAST STAIN**

- **Acid-fast bacilli** – *Mycobacterium tuberculosis*
- **Non-acid-fast bacilli** – *Staphylococcus aureus*

Based on Cultural characteristics

- Extra growth factors requirements
 - **Fastidious** – *Hemophilus influenzae*
 - **Non-fastidious** – *Escherichia coli*
- Hemolysis on Sheep Blood Agar
 - **Alpha-hemolysis** – *Streptococcus pneumoniae*
 - **Beta-hemolysis** – *Streptococcus pyogenes*
- Utilization of carbohydrates
 - **Oxidative** - *Micrococcus*
 - **Fermentative** – *Escherichia coli*

Based on Cultural characteristics

- **Growth rate**
 - **Rapid growers**– *Vibrio cholerae*
 - **Slow growers** – *Mycobacterium tuberculosis*
- **Pigment production**
 - **Pigment producer** – *Staphylococcus aureus*
 - **Pigment non-producer** – *Escherichia coli*

Based on Nutrition

- **Autotrophs**
- **Heterotrophs**

Based on environmental factors

- **Temperature**
- **Oxygen dependence**
- **pH**
- **Salt concentration**
- **Atmospheric pressure**

Temperature

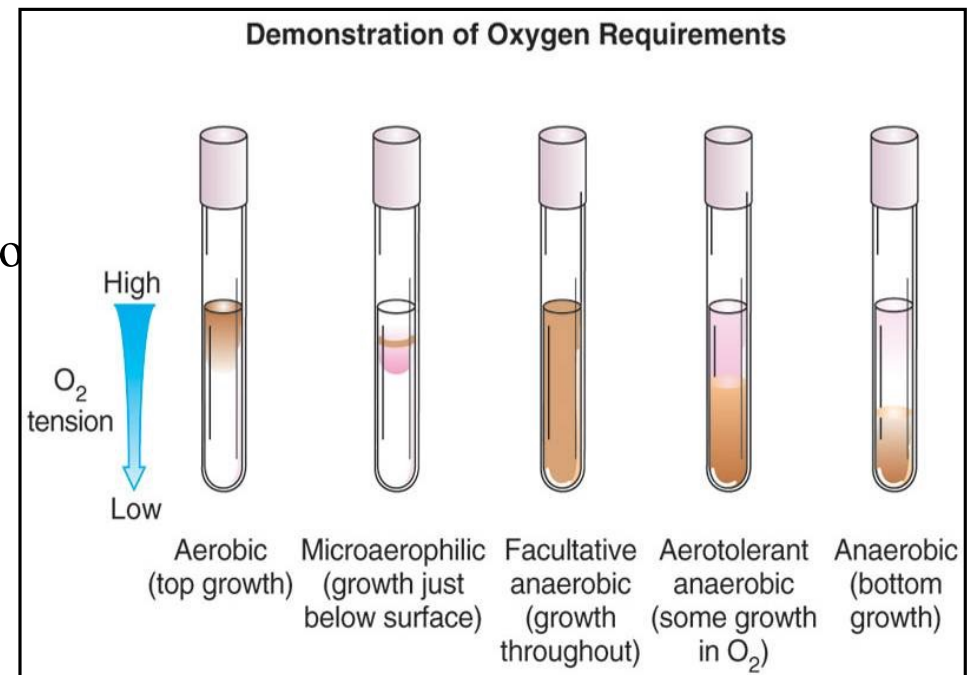
- **Psychrophiles** (15-20⁰C) – *Pseudomonas fluorescens*
- **Mesophiles** (20-40⁰C) – *Escherichia coli*, *Salmonella enterica*, *Staphylococcus aureus*
- **Thermophiles** (50-60⁰C)- *Bacillus stearothermophilus*
- **Extremely thermophiles** (as high as 250⁰C)

Oxygen dependence

- **Aerobe** (grow in ambient temperature, which contains 21% O₂ and a small amount of CO₂, 0.03%)
- **Obligate aerobes** – Strictly require O₂ for their growth (*Pseudomonas aeruginosa*)
- **Microaerophilic** (grow under reduced O₂, 5-10% and increased CO₂, 8-10%)-
Campylobacter jejuni, *Helicobacter pylori*

Oxygen dependence

- **Facultative anaerobe** (capable of growing either in presence or absence of O_2)- *E. coli*
- **Obligate anaerobe** – *Clostridium* spp.
- **Capnophilic** (require increased concentration of CO_2 , i.e., 5-10%) –
H. influenzae,
N. gonorrhoeae
- **Aerotolerant**



pH

- **Acidophiles** (*Lactobacillus acidophilus*)
- **Alkaliphiles** (*Vibrio*)
- **Neutralophiles** (pH 6-8)

Majority of the medically important bacteria grow best at neutral or slightly alkaline reaction (pH 7.2-7.6)

Salt concentration

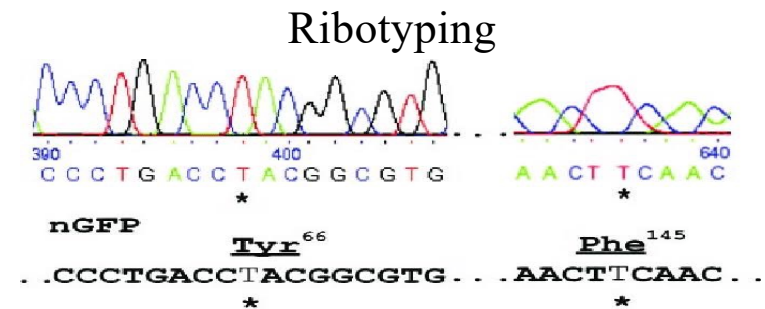
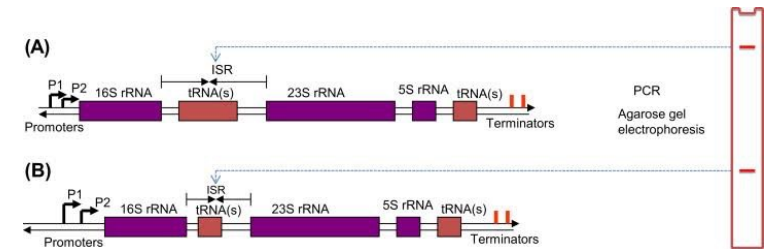
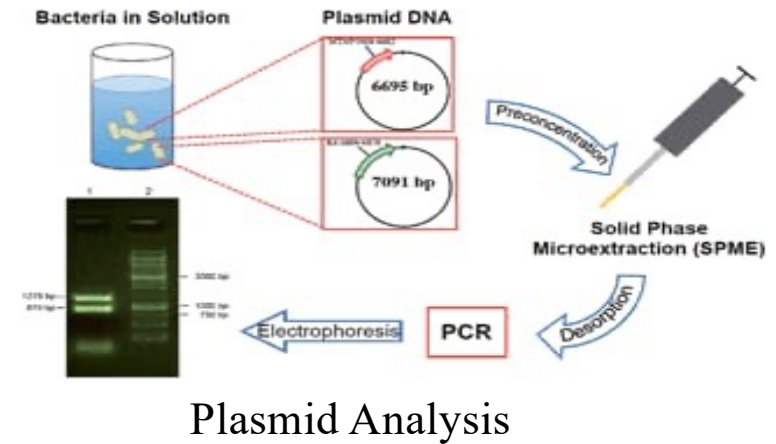
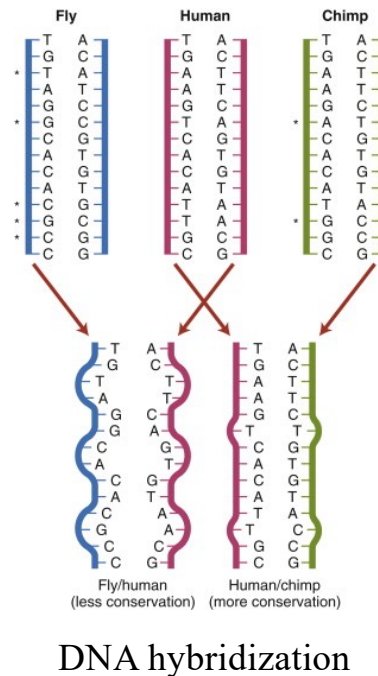
- **Halophiles** (*Halobacterium*)
- **Non-halophiles**

Other ways of classification

- **Motile/Non-motile**
- **Pathogenic/Non-pathogenic**
- **Sensitive/Resistant (to particular antibiotic/ chemicals)**
- **Lactose fermenter/Lactose non-fermenter**
- **Bergey's Manual of Determinative Bacteriology**
 - **Gram-negative eubacteria that have cell walls**
 - **Gram-positive eubacteria that have cell walls**
 - **Cell wall-less eubacteria: Mycoplasma**
 - **Archaeobacteria**

Genotypic classification of Bacteria

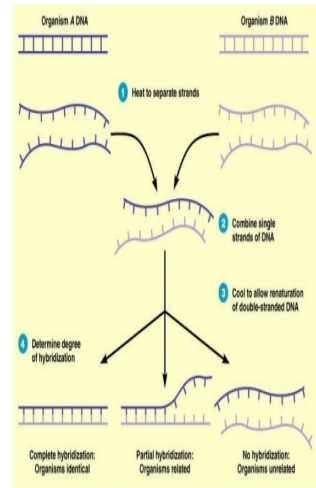
- Organisms are classified on the **basis of genetic similarities** so that the genomes can be compared.
- Genotypic classification is based on –
 1. DNA hybridization
 2. Plasmid analysis
 3. Ribotyping
 4. DNA sequences analysis



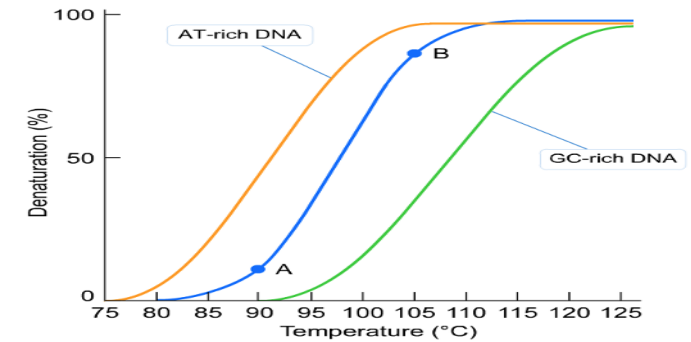
Chromosomal DNA sequence analysis

Phylogenetic classification of Bacteria

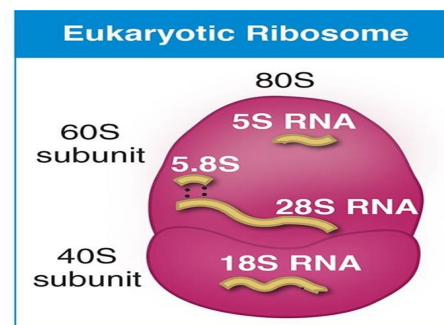
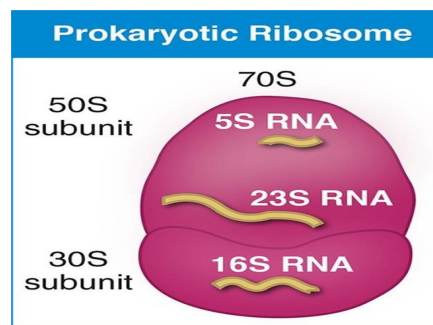
- Organisms are classified on the **basis of the evolutionary relationships** between microbes having similarities and dissimilarities between genes sequences.
- Phylogenetic classification is based on –
 1. Base composition (G:C Ratio).
 2. Nucleotide hybridization.
 3. Ribosomal sequences analysis
 4. Protein profiling



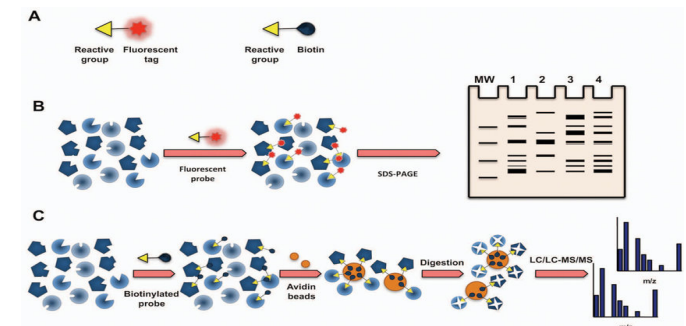
Nucleotide hybridization.



Base composition (G:C Ratio)



Ribosomal sequences analysis



Protein profiling and amino acid sequencing.

Thank you