

2nd year Medical Students – JU
Bacterial Structure and Morphology

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Bacterial structure and morphology

- Medical microbiology is science of studying micro-organisms that are associated with human disease
- Agents of infection include cellular organisms belonging to two of the three recently defined *domains of life*:

Bacteria (prokaryotes)

Eukarya: fungi and protozoa.

- *The subcellular entities viruses, viroids and prions also cause infection but depend on host cells and tissues for propagation.*

- It is important to understand the basic structural properties and physiology of micro-organisms to establish our approach to infections
- Our understanding of microbial cytology aided by developments in genetic manipulation combined with advances in fluorescence and electron microscopy

- Micro-organisms are microscopic in size and are usually unicellular.
- The diameter of the smallest body that can be resolved and seen clearly with the naked eye is about 100 μm .
- All medically relevant bacteria are smaller than this and a microscope is therefore necessary to see individual cells

Bacterial cell:

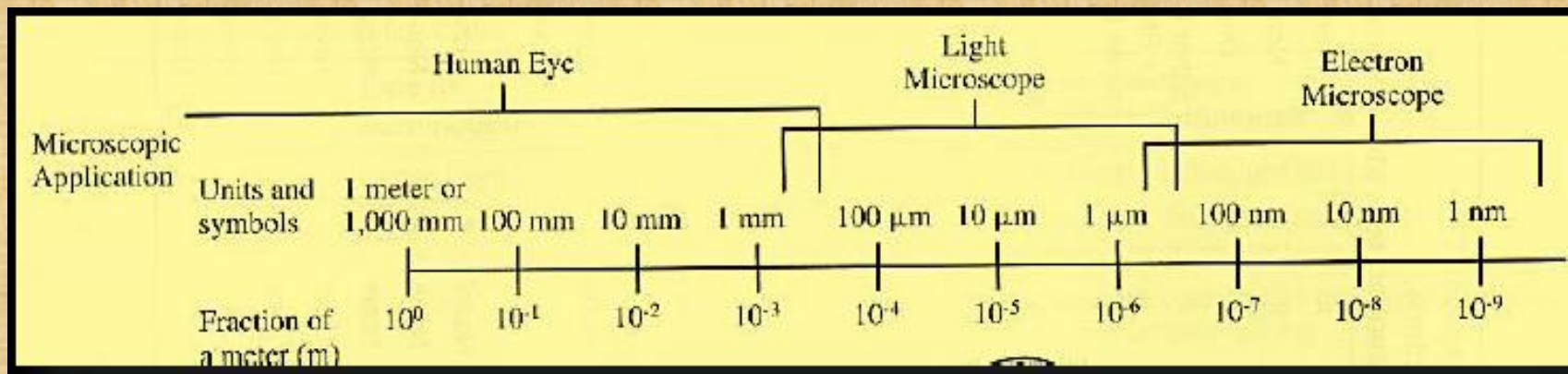
➤ Prokaryotes

- No true nucleus
- No organelles

➤ Divide-binary fission asexual

- Unit for measurement :Micron or micrometer, μm : $1\mu\text{m}=10^{-3} \text{ mm}$

- Size: Varies with kinds of bacteria, range from 0.2 to 6 μm .



Bacterial structure

- **Essential components such as**

Cell wall

Cytoplasmic membrane

Ribosome

Nucleoid

- **Accessory components (not every bacteria has):**

Capsule, Pilus or fimbria, Flagella

Spores, Plasmid, Transposons

Bacterial structure

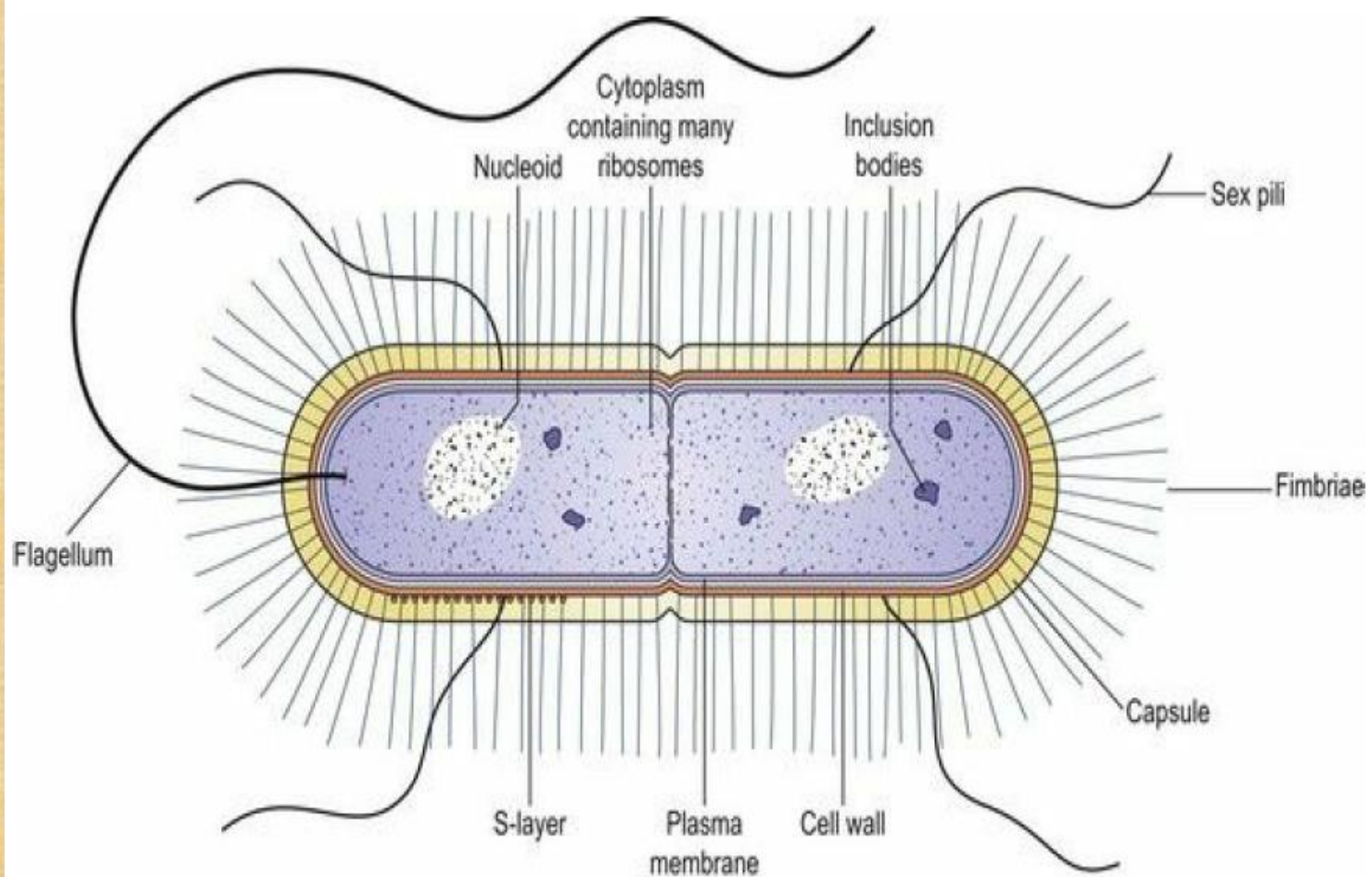


Fig. 2.2 Diagram illustrating the key features of bacterial cells. The S-layer is a variably demonstrated ordered protein layer.

- Cytoplasm is bounded peripherally by a very thin, elastic and semi-permeable cytoplasmic (or plasma) membrane (phospholipid bilayer).
- Outside, and closely covering this, lies the *cell wall, which is rigid, porous and relatively permeable*
- Cell wall and cytoplasmic membrane called collectively the cell envelope

Cytoplasm:

- *Is a predominantly aqueous environment*
- *Contains nucleoid, ribosomes and numerous other protein and nucleotide–protein complexes*
- Bacterial cytoplasm have cytoskeletal structures (filamentous proteins and filament systems)
- The importance of these cytoskeletal structures: determining cell shape, **division** and spore formation antimicrobials targeting.

Nucleoid:

- Area of cytoplasm where bacterial DNA is located
- Bacterial chromosome is double stranded circular and supercoiled
- No nuclear membrane as in eukaryotes

Ribosomes:

- Sites of protein synthesis
- They have a sedimentation coefficient of 70S, being composed of a 30S and a 50S subunit (80s in eukaryotes)

Inclusion bodies:

- Food and energy storage granules e.g glycogen and starch

Cytoplasmic (plasma membrane)

- Thin, permeable and elastic membrane
- Composed of phospholipids , **mesosomes** & proteins

• Functions:

- Synthesis of precursors of cell wall polymers and membrane lipids.
- Selective permeability and active transport of molecules into cells.
- **Energy generation** by oxidative phosphorylation.
- Excretion of enzymes and toxins.

Cell wall

Importance:

- Bacterial rigidity and shape
- protection against osmotic changes
- Porous to allow nutrients passage.
- Structure differs in gram positive & negative bacteria.

Cell wall

- In almost all bacteria except Mycoplasma..

- Many antibiotics (penicillins, and cephalosporins) stop bacterial infections by interfering with cell wall synthesis

- Has no effects on human cells (no cell wall only a cell membrane).

Cell wall components

Peptidoglycan (syn. mucopeptide or murein):

- *Is an important component of the cell wall of almost all bacteria.*

- Peptidoglycan is composed of:

N-acetylglucosamine and N-acetylmuramic acid molecules linked alternately in a chain (Fig).

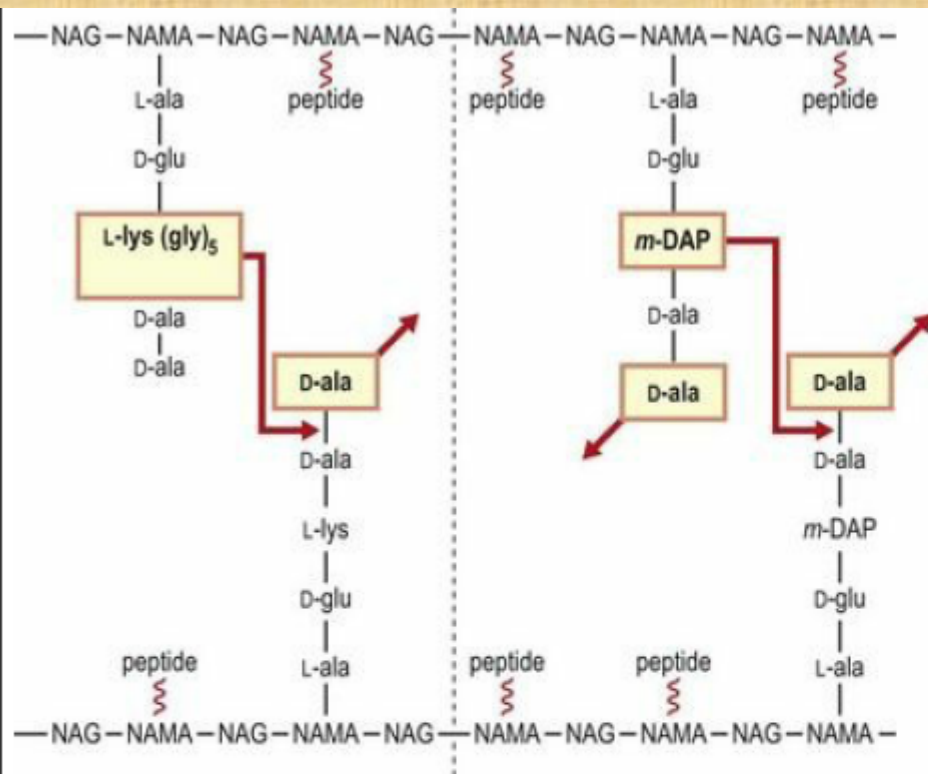
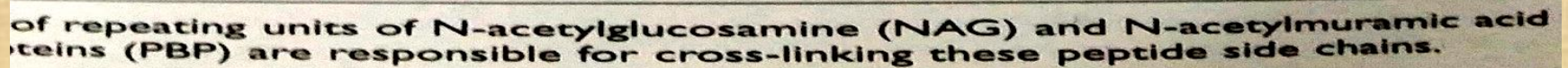


Fig. 2.8 Schematic representation of the peptidoglycan of a representative Gram-positive organism (*Staphylococcus aureus*) and a representative Gram-negative organism (*Escherichia coli*). Note that in the Gram-positive bacterium cross-linking occurs through a peptide bridge (pentaglycine in *Staph. aureus*), whereas direct cross-linking occurs in *E. coli*. In both cases the terminal D-alanine is lost. Not all peptides are engaged in cross-linking in *E. coli*, and carboxypeptidases remove redundant D-alanine residues. NAG, *N*-acetylglucosamine; NAMA, *N*-acetylmuramic acid; *m*-DAP, *meso*-diaminopimelic acid.








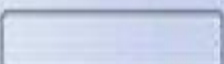




- The thickness of the peptidoglycan is of great practical importance in differentiating medically significant bacteria.
- A Danish physician, Christian Gram, devised a staining procedure that we now know distinguishes bacteria with a thick (Gram-positive) and a thin (Gram-negative) peptidoglycan layer (see Fig).
- The traditional classification of bacteria is basically relying on this method of staining

- Bacterial Shape (morphology) is determined by cell wall and cytoplasmic cytoskeleton
- Following staining; Bacteria is described by gram stain and shape e.g Gram positive cocci, Gram negative rods or bacilli
- Gram-positive species may sometimes appear Gram-negative under certain conditions of growth, especially in ageing cultures on nutrient agar or after exposure to antibiotics or over decolorization.

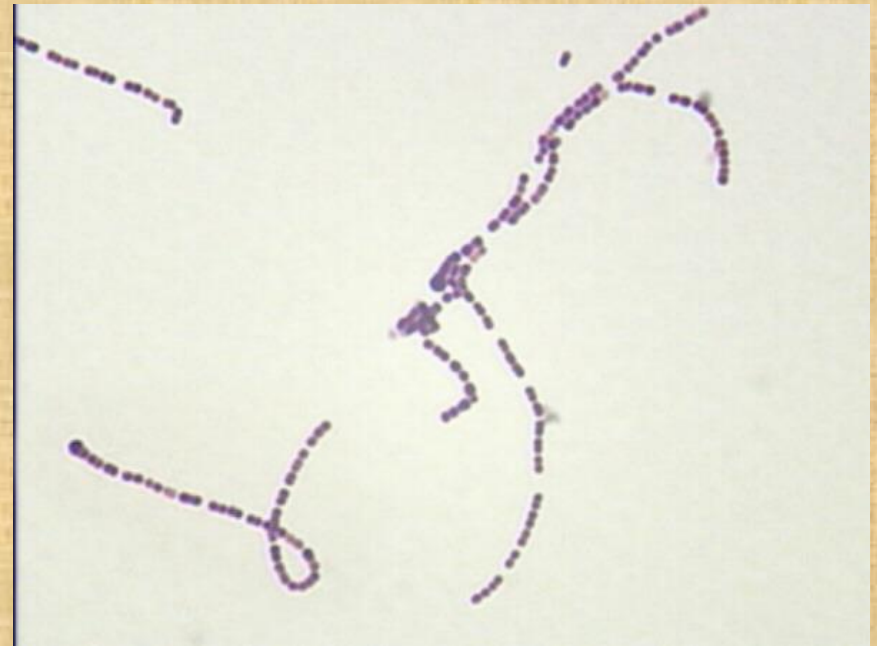
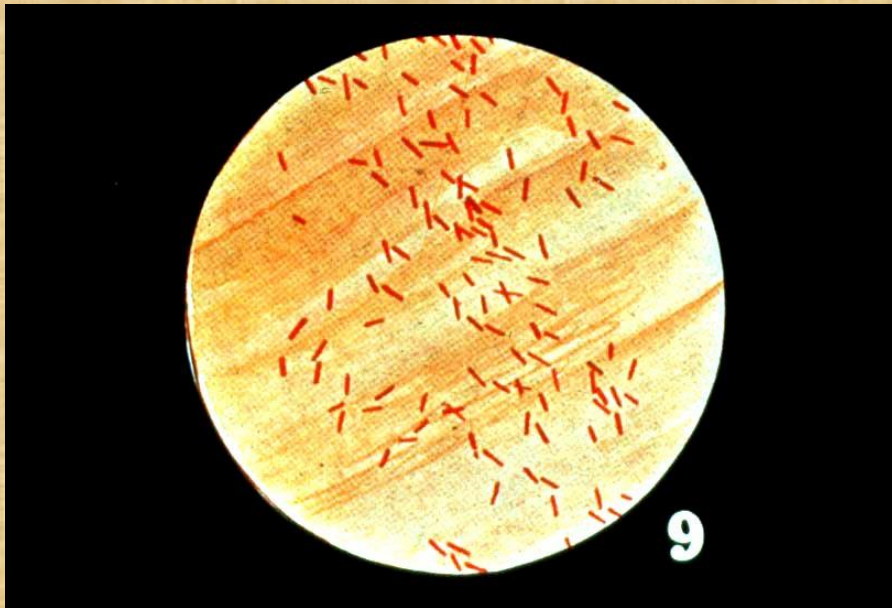
Gram stain: GO TO LAB

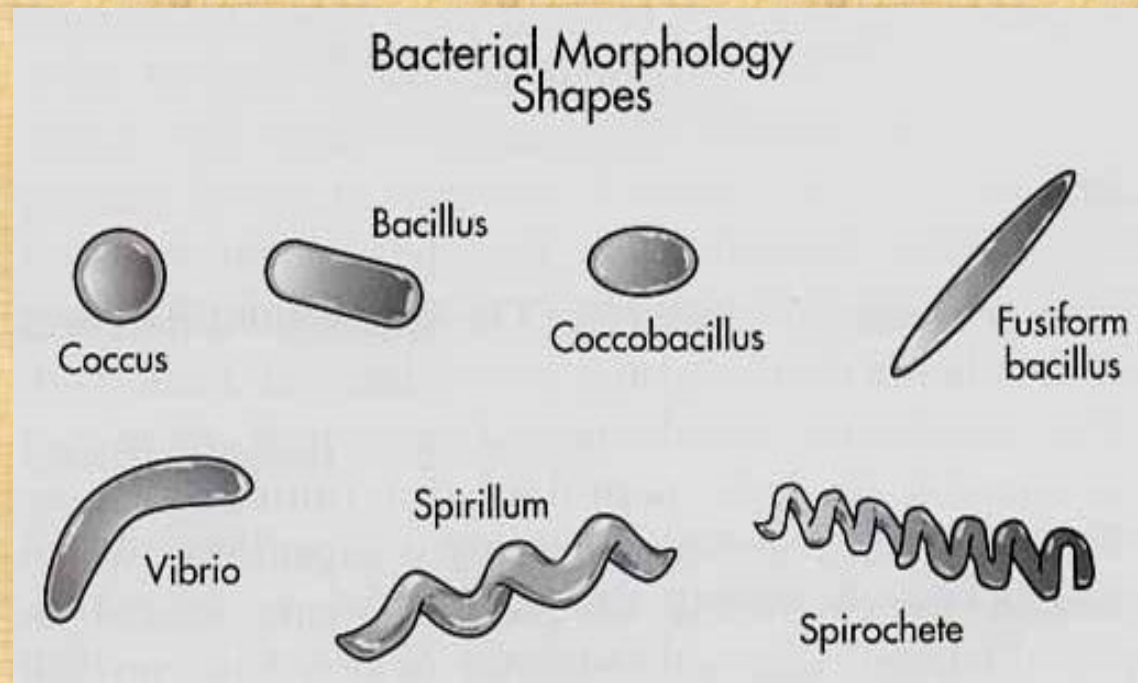
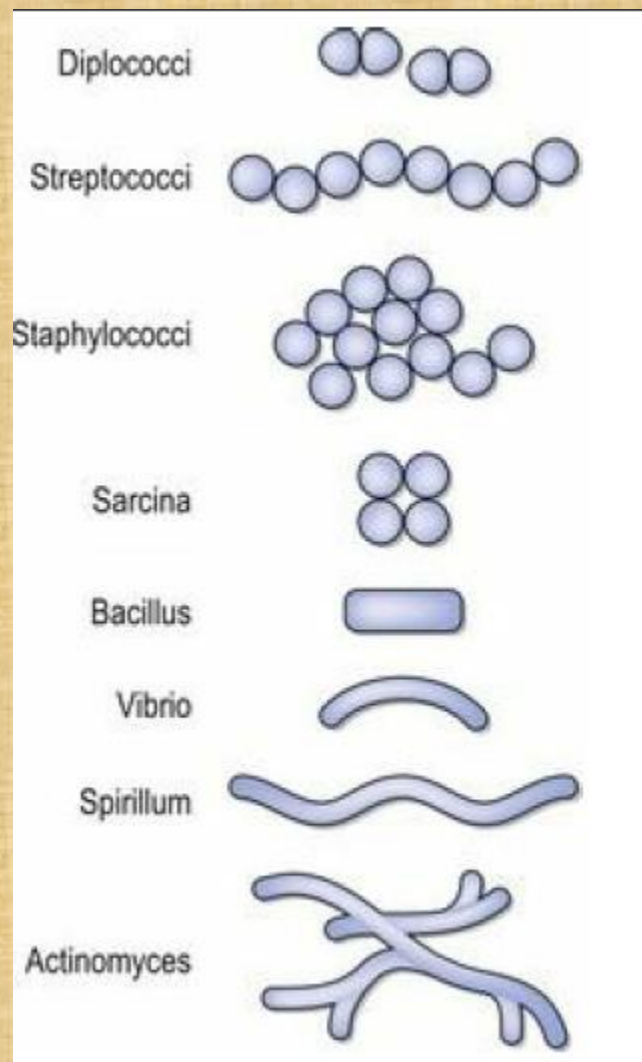
Outline of Gram stain

	Gram-positive	Gram-negative	
1. Unstained			
2. Crystal violet			<p>The dye is non-covalently bound to negatively charged molecules (particularly nucleic acids) in the cell</p> <p>This forms macromolecular complexes with Crystal violet</p>
3. Iodine			
4. Decolorize			
5. Red/pink dye			<p>The complexes are extracted through the Gram negative wall by solvents such as acetone but retained by Gram positives</p> <p>A further dye is needed to colourise the unstained Gram negatives</p>

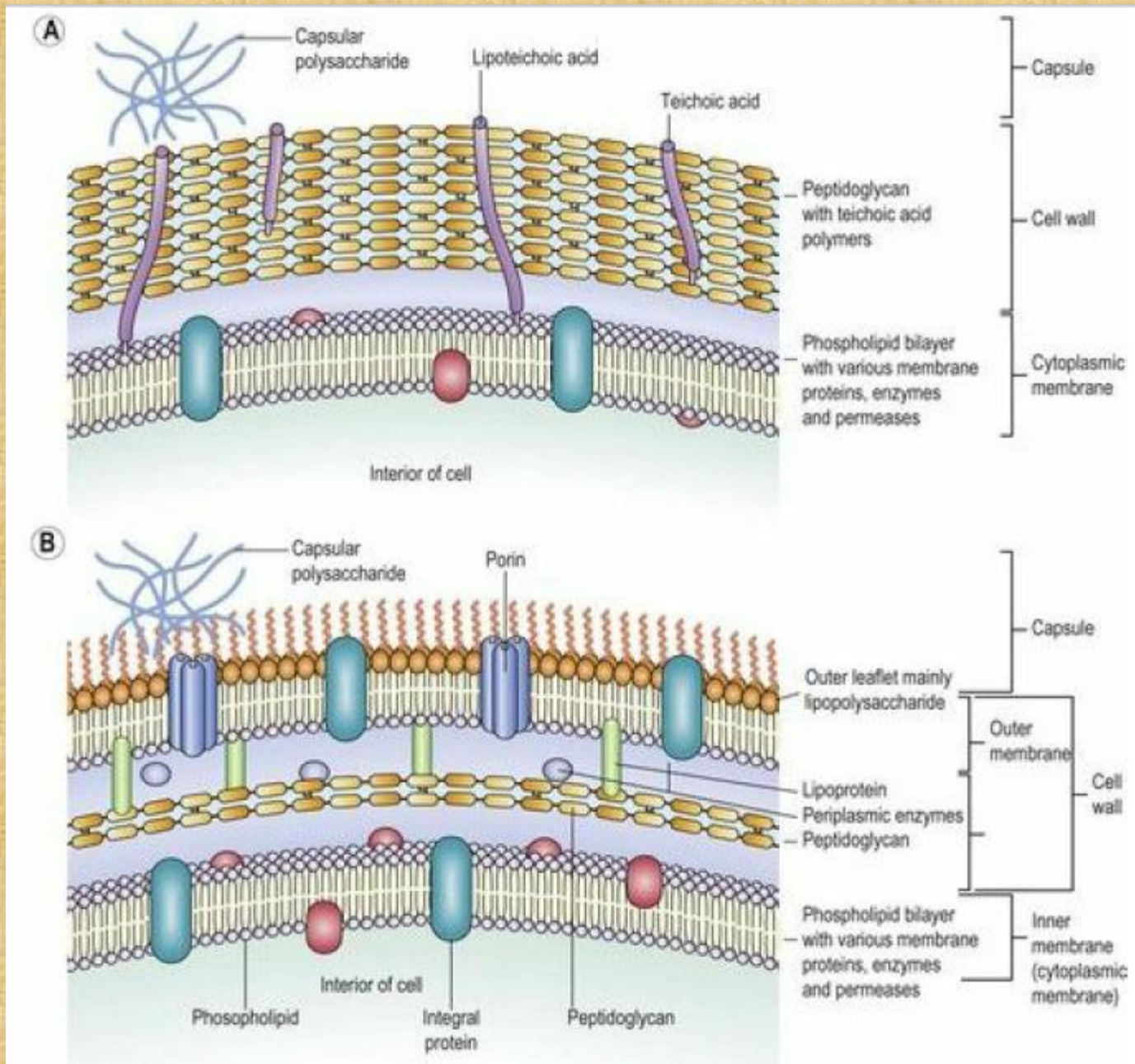
Medical Microbiology

- Gram positive appear violet/blue while gram negative appear pink.
- Importance? Guide your choice of antibiotics.





cell envelop of gram positive (A) and negative bacteria (B)



The gram positive cell wall

- Thick, and the peptidoglycan layer constitutes almost 95% of the cell wall.
- Many Gram-positive bacteria contain relatively large amounts of *teichoic acid* (a polymer of ribitol or glycerol phosphate complexed with sugar residues) interspersed with the peptidoglycan.
- Some of this material (*lipoteichoic acid*) is linked to lipids buried in the cell membrane.
- Functions of the Teichoic acids include attachment and antigenic function.

The gram negative cell wall

➤ The peptidoglycan layer is thin constitutes as little as 5-10% of the cell wall

➤ Outer membrane

2 layers of lipids

-Inner layer-phospholipids

-Outer layer-Lipopolysaccharide (endotoxins causing endotoxic shock) consists of 3 regions

1.Lipid A (toxic effect)

2.Core polysaccharide

3.O antigen (antigenicproperties)

The gram negative cell wall

Advantages of outer membrane:

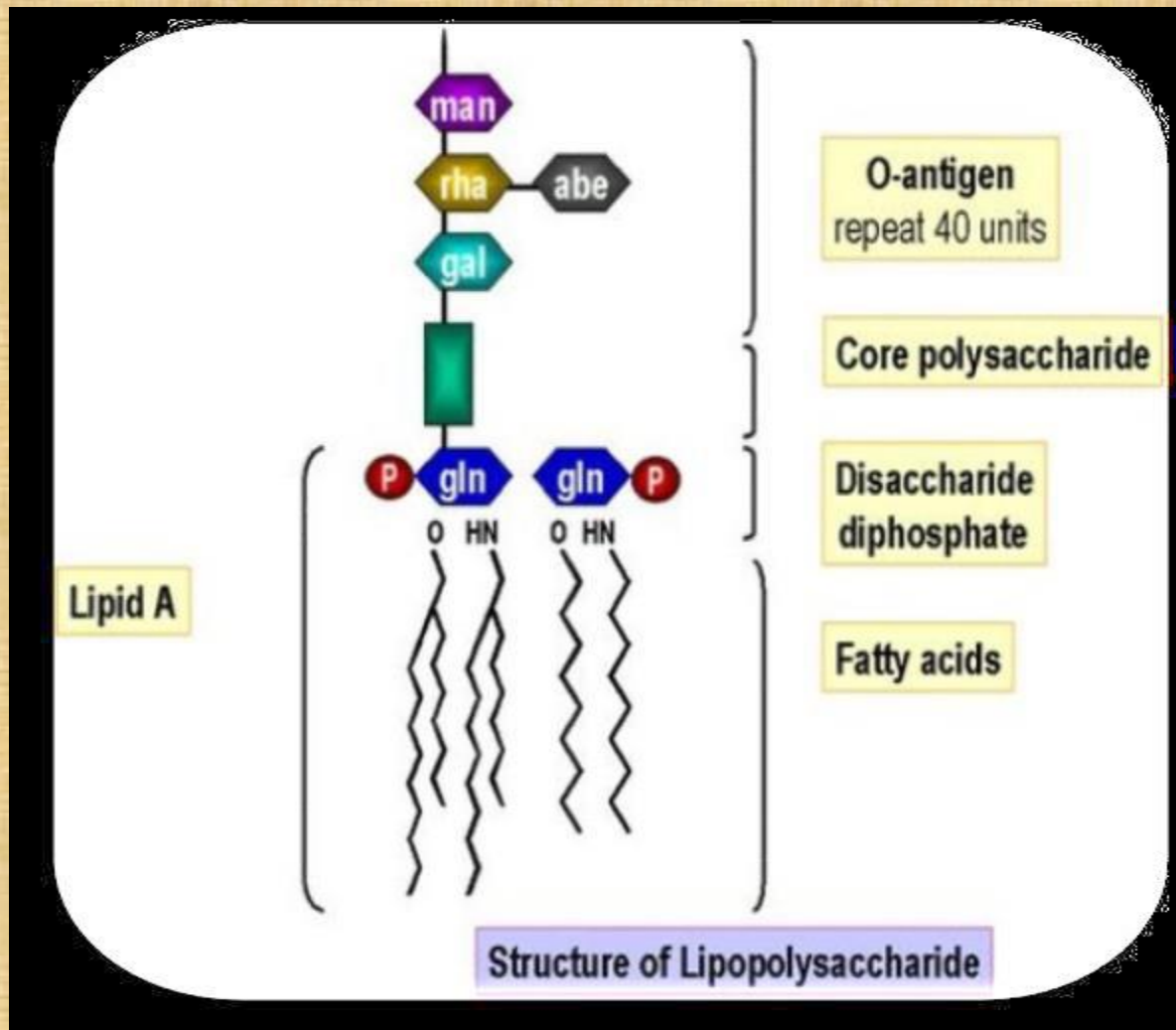
- It protects the peptidoglycan from the effects of lysozyme (a natural body defence substance that cleaves the link between *N-acetylglucosamine* and *N-acetylmuramic acid*).

- It impedes the entry of many antibiotics.

➤ Transmembrane proteins:

- Porins proteins for selective permeability
- Integral proteins that help in attachment

The gram negative cell wall LPS

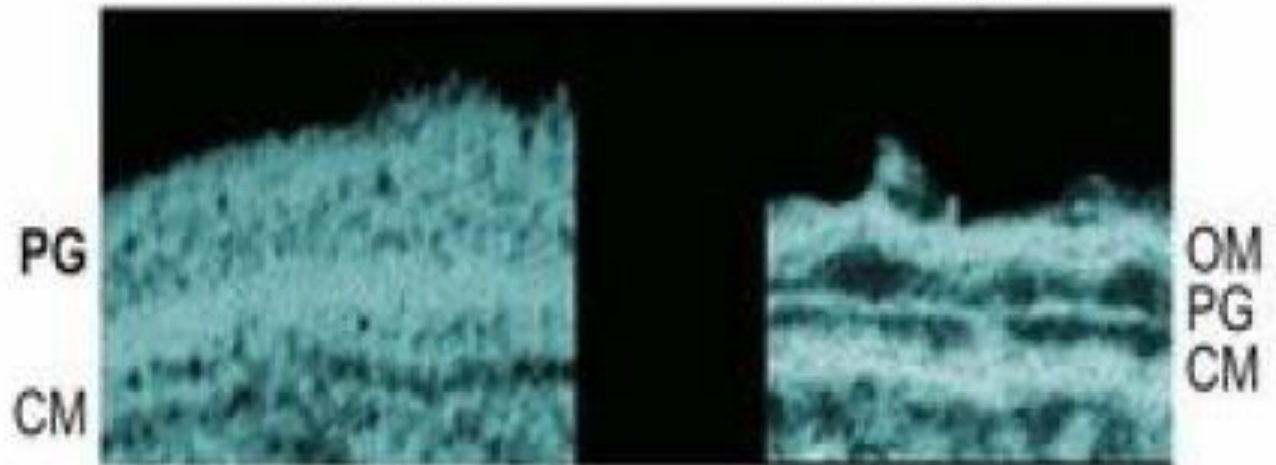


Transmission electron micrograph

(Colourized negative image)

Gram-positive

Gram-negative



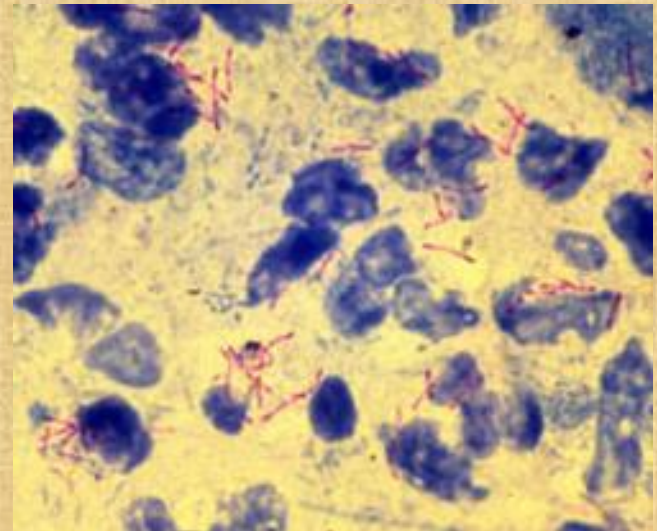
Diagram



- Some bacteria are classified as Gram positive but stain poorly because they have a cell envelop that is rich in hydrophobic lipid mycolic acid

- Examples: mycobacterium and corynebacterium

Staining method used: Acid fast stain (Ziehl Neelsen stain)



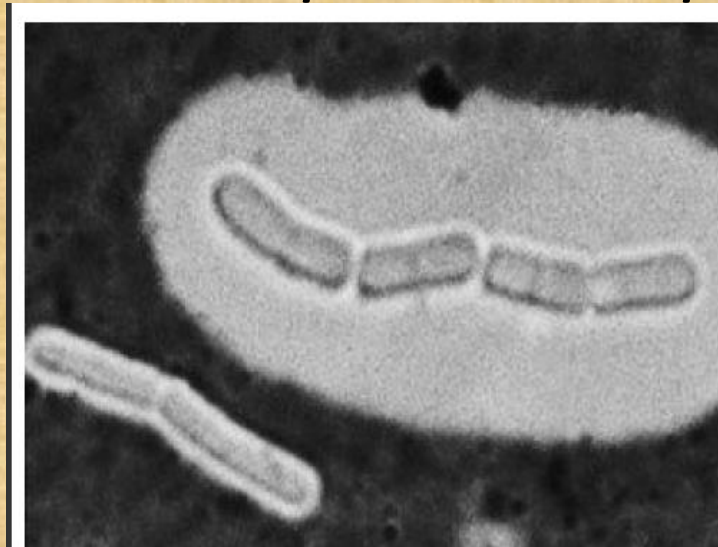
- Some bacteria can not be stained because they are intracellular as *Chlamydiae* & *Rickettsiae*.

Capsule

- Consists largely of water and has only a small content (e.g. 2%) of solids.
- In most bacteria, the solid material is a complex polysaccharide,
- In some bacteria its main constituent is polypeptide e.g. *Bacillus*.

Capsule

- Antiphagocytic and protect against lytic action of complement(virulence).
- Adherence (initial step of infection)
- Antigenic (vaccines *Streptococcus pneumoniae*)



- Useful for diagnosis using antiserum against capsular polysaccharide (quellung reaction)

Free slime / Glycocalyx

- Polysaccharide coat similar to capsule but secreted extracellularly
- Cover the surfaces like a mucoid film.
- Allow firm adherence to structures e.g.: heart valves, skin, catheters, surface of the teeth (*S. mutans* in dental caries)

Flagella

- A long, filamentous whip-like locomotor appendages
- Originate from cytoplasm and cytoplasmic membrane and protrude via the cell envelope to the surrounding environment
- Some bacteria have for movement towards food, cells and other attractants in a process called chemotaxis.
- Flagella consist of many subunits of protein called flagellin

➤ flagella are important in:

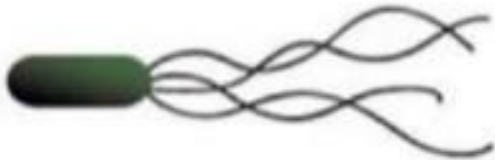
- 1. Identification of Bacteria: specific antibodies against flagellar protein
- 2. Pathogenesis (*E. coli* in urinary tract infection)
- 3. Motility of bacteria

fig

Types of flagellar arrangement



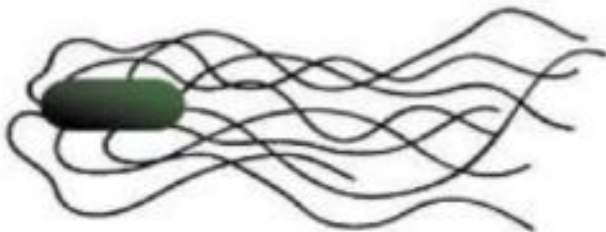
Polar/ Monotrichous – single flagellum at one pole



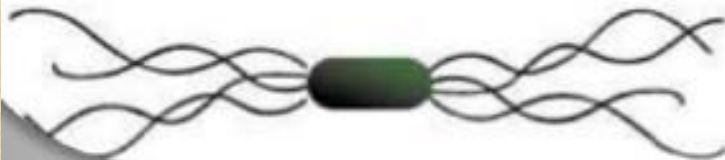
Lophotrichous – tuft of flagella at one pole



Amphitrichous – flagella at both poles



Peritrichous – flagella all over



Amphilophotrichous – tuft of flagella at both ends



→ Pili

→ Flagella

Pilli and Fimbriae

- Filamentous appendages made of pilin protein subunit
- More numerous and straight than flagella

➤Fimbriae:

important in mediating adhesion between the bacterium and host cells

➤Pilli:

- Attach specifically to other bacteria that lack these appendages to initiate the process of conjugation (genetic material transfer)
- also act as receptor sites for certain bacteriophages described as being 'donor specific'

Spores

- **Dormant cell**, Highly resistant structures are formed to adverse conditions such as shortage or lack of nutrients.
- John Tyndal

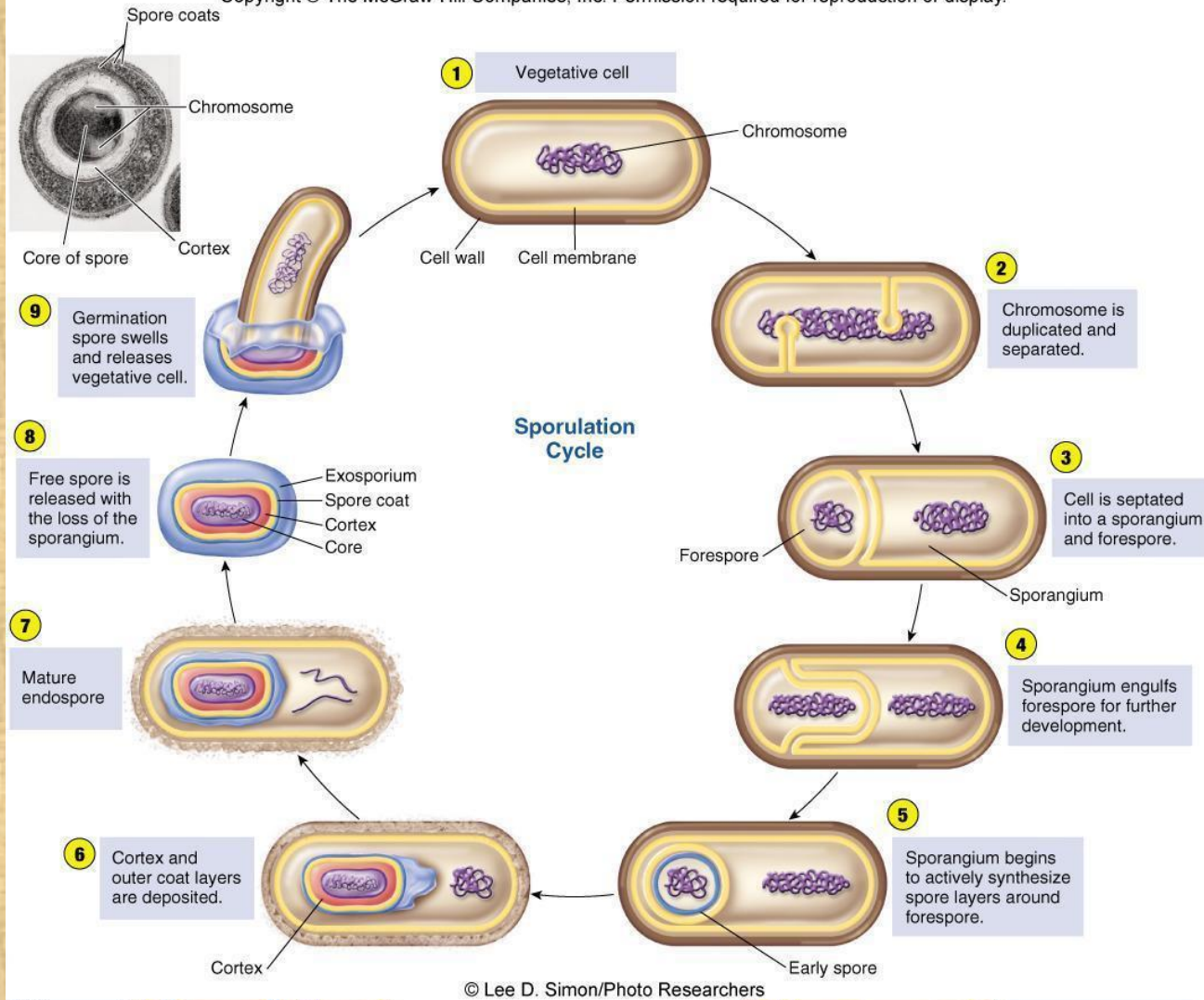
- Resistant to:
 - ✓ Heat, drying irradiation, cold
 - ✓ Boiling >1 hrs till viable

Only moist heat e.g 120 C for 20 minutes will kill them

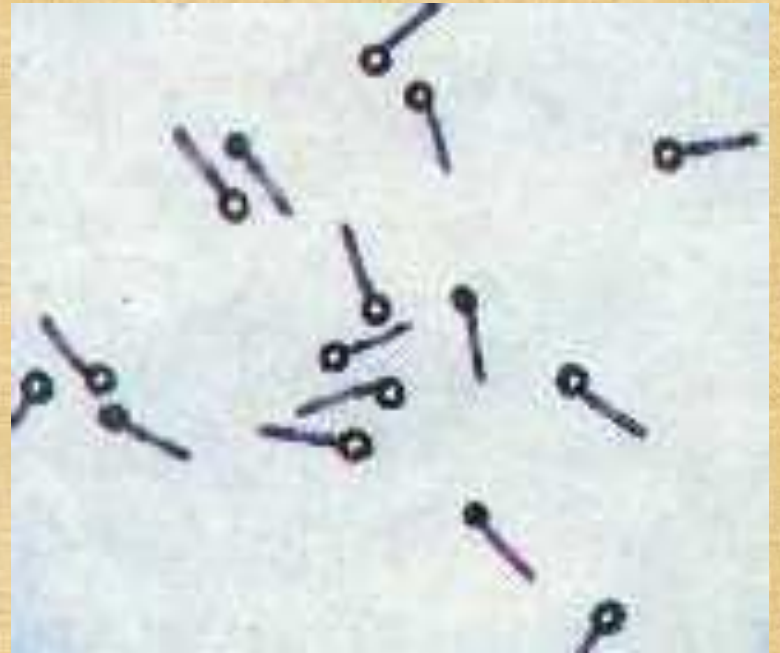
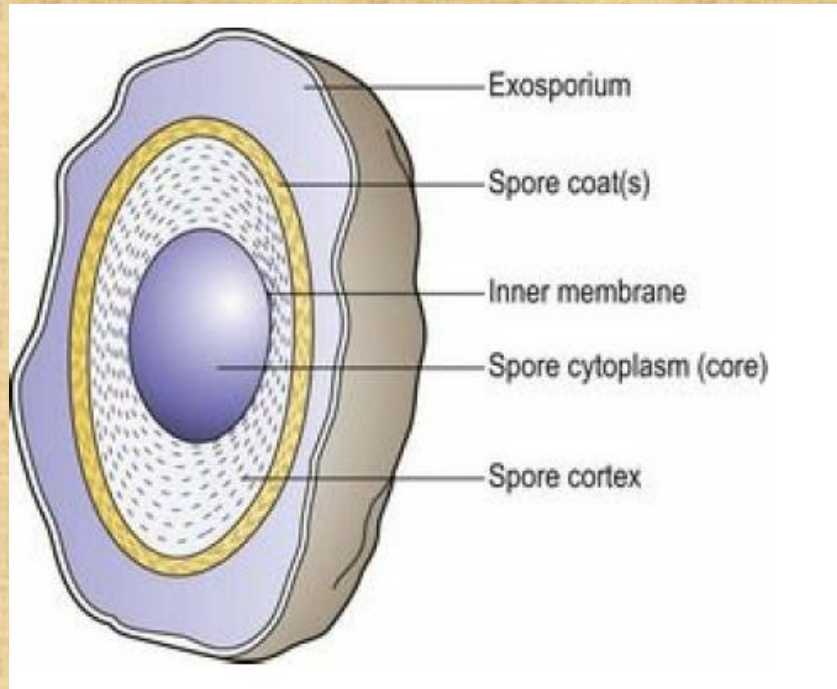
- Produces by two genera of medical importance: *Bacillus and Clostridium*
- Contain calcium dipicolinate and keratin layer
- Stained by different stains e.g **ZN stain, malachite green**
- **Location important in classification**
- Central, Sub-terminal, Terminal
- Sporulation vs germination

Spores

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Spores



➤ Plasmid:

- Extra-chromosomal, circular DNA, double-stranded molecule.
- Replicate independent of bacterial chromosome
- Transmissible or non-transmissible plasmids
- contain genes that confer some properties such as antibiotic resistance, virulence factors (exotoxin), genes for pili.
- Plasmids are not essential for cellular survival.

➤ Transposons:

- Pieces of DNA that moves from one site to another either within or between the DNAs of bacteria, plasmids and bacteriophages “Jumping genes”.

The end