

CLASSIFICATION OF BACTERIA

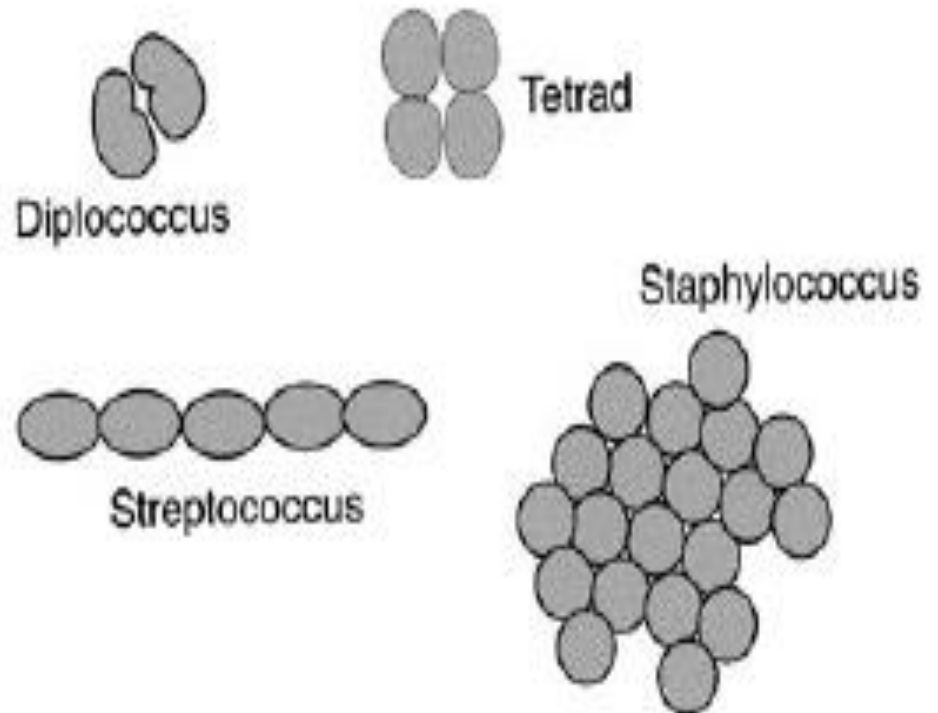


Classification of Bacteria

- Systemic & simplified classification & identification of bacteria is based on:
 1. Morphology
 2. Staining
 3. O₂ requirement
 4. Biochemical tests

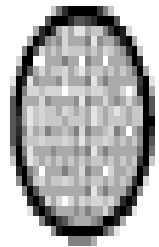
Morphology

1 - Cocci: streptococci (in chains), diplococci (pairs), staphylococci (clusters).

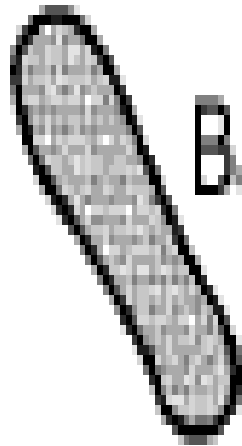


Morphology

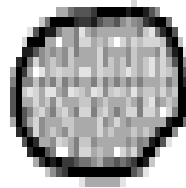
- 2- Bacilli: rod shaped
- 3- vibrio: comma shaped or curved
- 4- Filamentous: long filaments
- 5- Spirochete: many coils



Coccus



Bacillus



Coccobacillus

Fusiform
bacillus



Vibrio



Spirillum



Spirochete

Staining

- **Using gram stain**
- bacteria could be divided into two groups depending on their reaction to a particular staining procedure developed by microbiologist Hans Christian Gram, they are either:
 1. Gram positive.
 2. Gram negative.

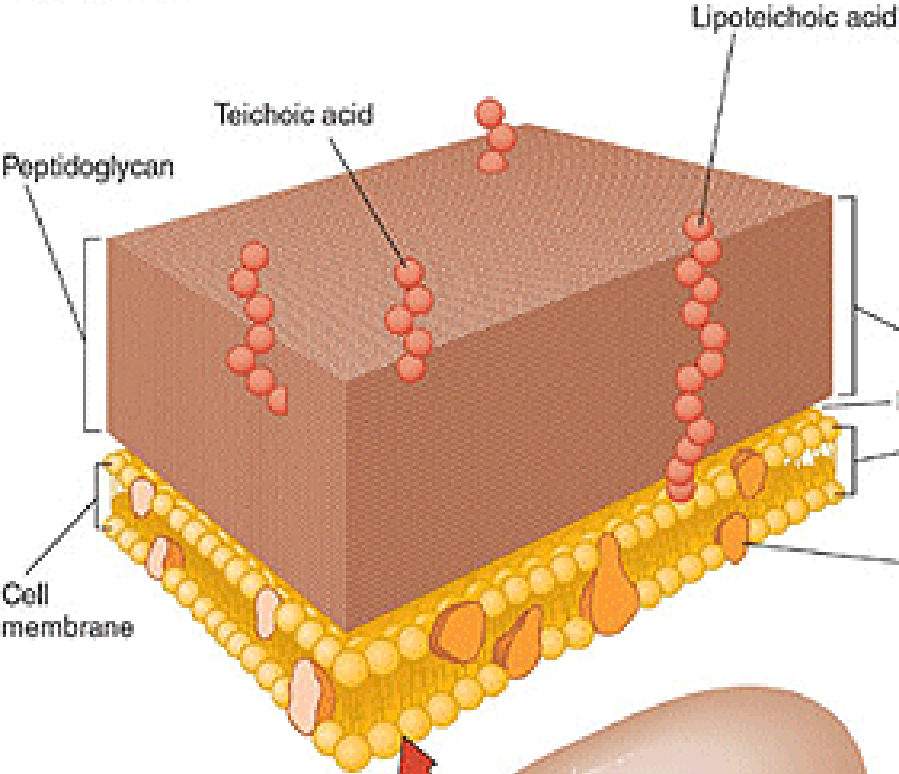
Staining

- All of the eubacteria with walls can be assigned a Gram response. However, there are few exceptions include:-
 1. Mycobacteria
 2. Most spirochetes, including *Treponema pallidum*

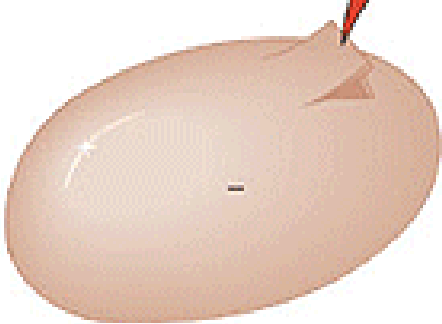
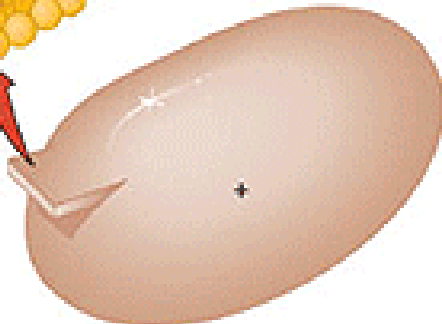
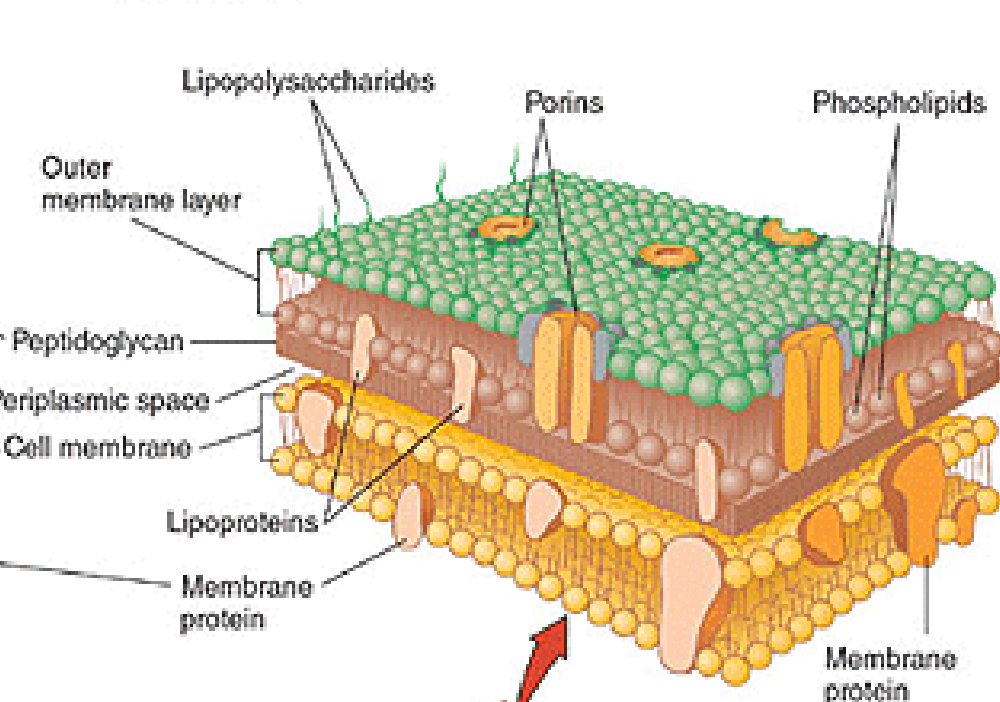
Structure of bacterial Cell Wall

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Gram Positive



Gram Negative



Structure of bacterial Cell Wall

- Muerin:- matrix of cross linked sheet of peptidoglycans which is found nowhere except in prokaryotes.
- In Gram negative bacteria there is a unique outer membrane.
- Its inner layer consists of ordinary phospholipids
- The outer layer consists of **lipopolysaccharide (LPS)**, which is extremely toxic to humans, and is called **endotoxin**.

Gram stain procedure:

1. The crystal violet dye stains all cells in blue/purple
2. The iodine solution is added to form a crystal violet-iodine complex, all cells appear blue.
3. The organic solvent such as acetone or ethanol extract the blue dye from the lipid-rich, thin-walled gram (-)ve bacteria to a greater degree than from lipid poor gram (+)ve bacteria

Gram stain procedure:

- The gram (-)ve organism appear colourless & the gram (+)ve remain blue.
- 4. The red dye safranin stains the decolourized gram (-)ve cells red/pink, the gram (+)ve remains blue.

Gram-positive

Gram-negative

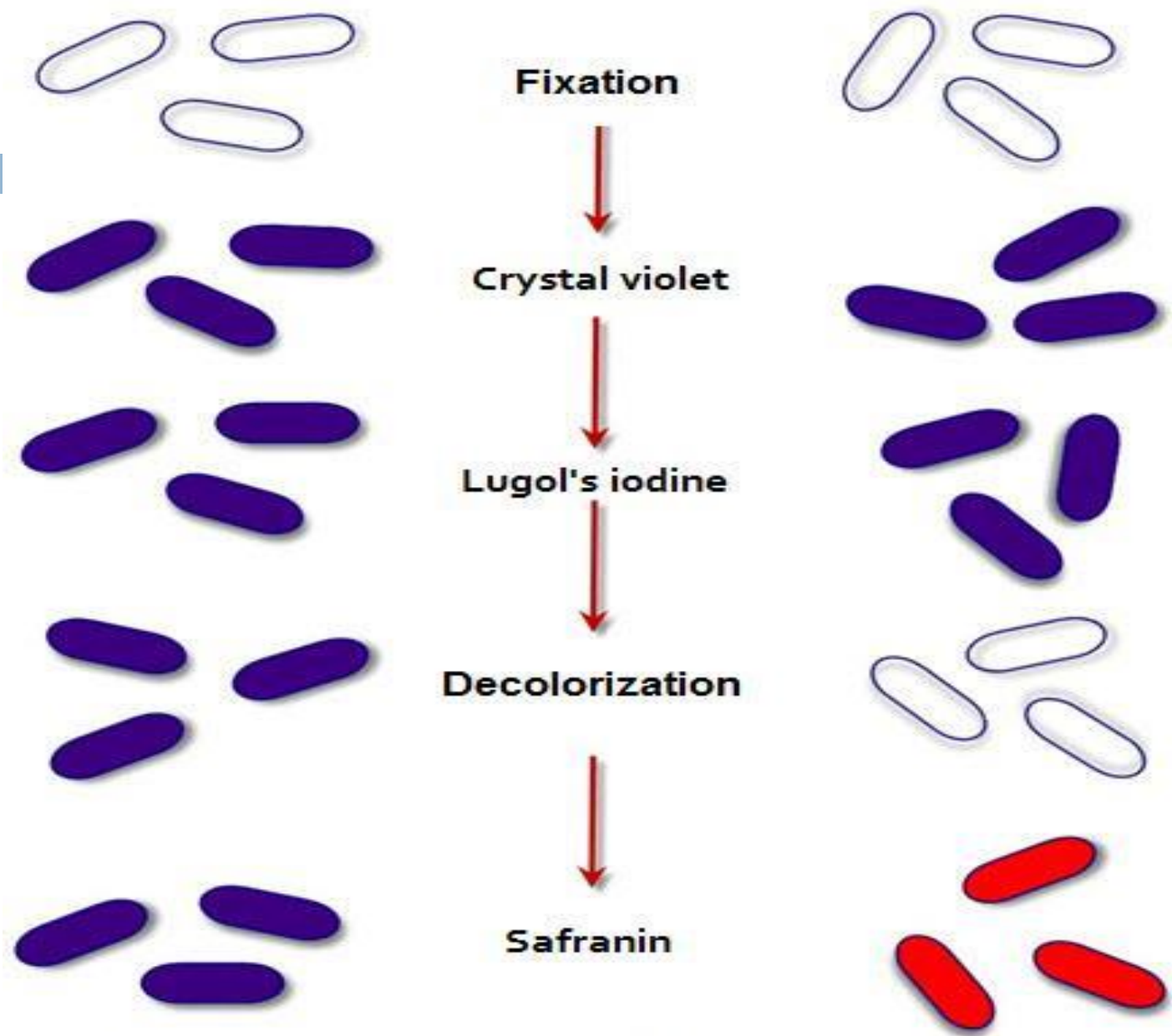


Diagram representation of the bacterial staining process

By gram stain we can know:

1. Morphology & arrangement of bacteria.
2. Gram reaction of bacteria.
3. Number of bacteria.
4. If the bacteria is spore forming or not.

Ziehl–Neelsen staining

- **Mycobacteria**, e.g., *Mycobacterium tuberculosis*; too much lipids (called **mycolic acids**) in cell wall results in their inability to be Gram-stained
- However, they can be stained by application of more concentrated dyes, with heat treatment by **Ziehl–Neelsen staining**

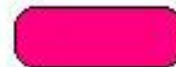
Acid Fast

Non Acid Fast

Carbol fuchsin



**Acid alcohol
decolorizer**



**Counterstain
Malchite Green**



**or methylene
Blue**

O₂ requirement

- Bacteria can be classified according to atmospheric condition to:-
 1. **Strict aerobes:** cannot survive in the absence of oxygen.
 2. **Strict anaerobic:** are killed in the presence of oxygen.
 3. **Facultative anaerobes:** can grow in the absence of oxygen but grow better in its presence.
 4. **Microaerophiles:** prefer a reduced oxygen.

Biochemical Tests

- This is useful for identification of bacteria because bacteria differ in enzymes they have.

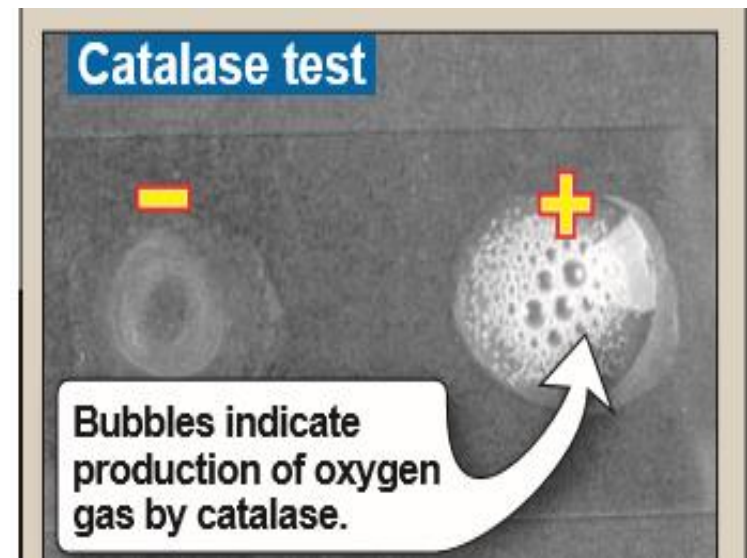
Catalase enzyme

- The **enzyme catalase** catalyzes the degradation of hydrogen peroxide to water and molecular oxygen



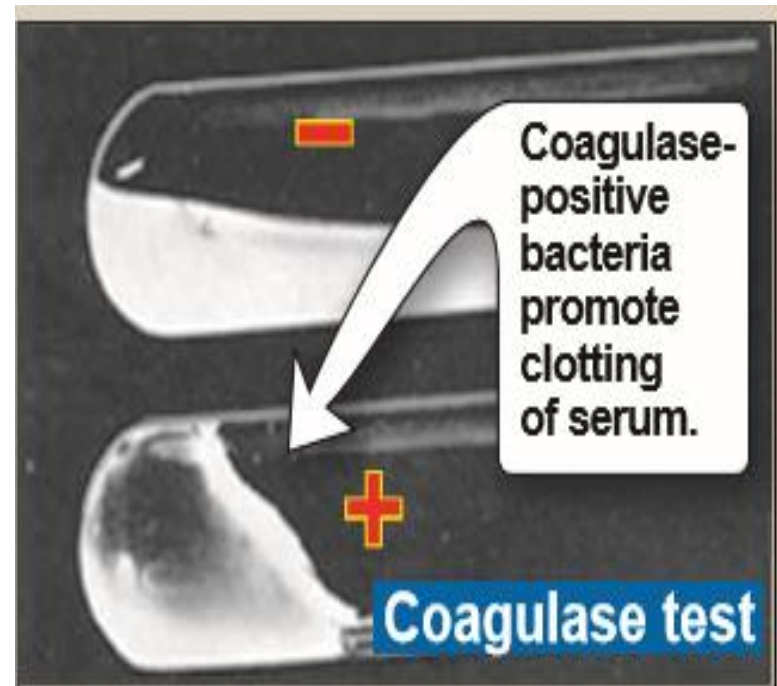
- The catalase test is key in differentiating between many gram-positive organisms

- staphylococci are catalase positive, whereas streptococci are catalase negative



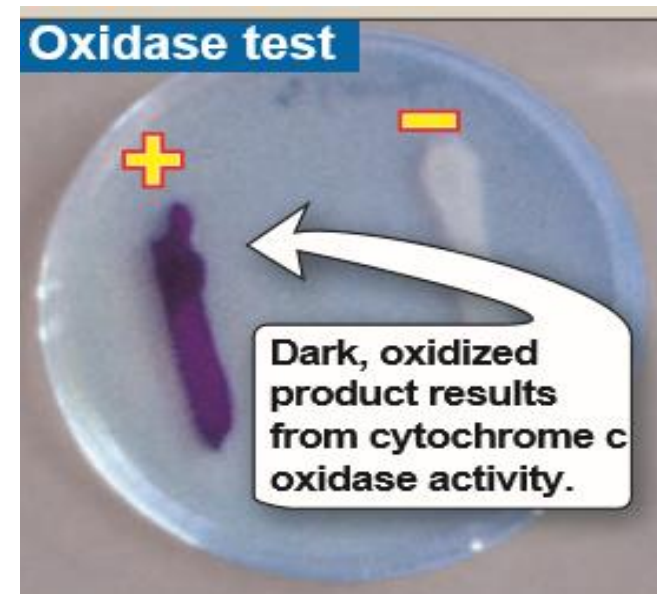
Coagulase enzyme

- Coagulase is an enzyme that causes a clot to form when bacteria are incubated with plasma.
- The test is used to differentiate *Staphylococcus aureus* (coagulase positive) from coagulase-negative staphylococci



Oxidase enzyme

- The enzyme can accept electrons from artificial substrates, producing a dark, oxidized product
- This test assists in differentiating between groups of gram-negative bacteria. *Pseudomonas aeruginosa*, for example, is oxidase positive



Urease enzyme

- The enzyme urease hydrolyzes urea to ammonia and carbon dioxide



- The ammonia produced can be detected with pH indicators
- The test helps to identify certain species of Enterobacteriaceae, *Corynebacterium urealyticum* and *Helicobacter pylori*.



The only way to do great work
is to love what you do.



~ Steve Jobs

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