Lec-1



Dr. Hanadi

Bacterial cell structure

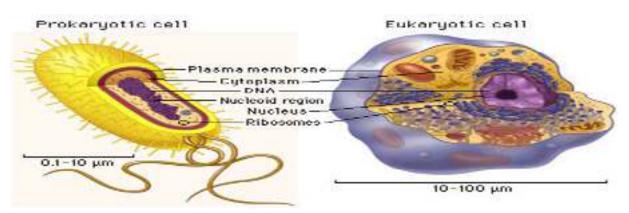
Objective: At the end of this lecture you will be able to:

- State and identify the basic structure and components that make up bacterial cells.
- Clarify and explain the functions of these structures.
- Compare and contrast the Gram-positive and the Gram-negative bacterial cells.
- Recognize the importance of these structures as virulence factors and in different lab diagnostic procedures.
- Q/ What is microbiology?
- Q/ What is the importance of microbiology?

Microbiology is the science (*logos*) of small (*micro*) life (*bios*), or the study of living things so small that they cannot be seen with the naked eye.

There are two basic and distinct forms of cells

- 1- **Prokaryotes:** unicellular organism that lack a membrane-bound nucleus (karyon), mitochondria, or any other membrane-bound organelle.
- **2- Eukaryotes**: are organisms, including humans, whose cells have a well-**defined** membrane-bound nucleus (containing chromosomal DNA) and organelles.



• The agent of human infectious diseases belong to five major groups of organisms: Bacteria, Fungi, Protozoa, Helminthes and Viruses

Q/ Compare between prokaryotes and eukaryotes?

Table-1

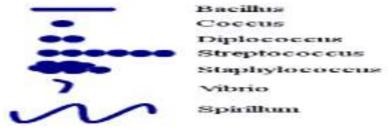
	PROKARYOTES	EUKARYOTES	
Organisms	Monera: Eubacteria and Archebacteria	Protists, Fungi, Plants and Animals single celled (protists mostly) or multicellular usually with tissues and organs	
Level of organization	single colled		
Typical cell size	small (1 -10 microns) large (10 - 100 microns)		
Cell wall	almost all have cell walls (murein)	st all have cell walls (mursin) fungi and plants (celluloss and chitin), none in animals	
Organelles	usually none many different ones with spec functions		
Me tabali sm	anaerobic and aerobic: diverse	mostly aerobic	
Senetic material	single circular double stranded DNA	complex chromosomes usually in pairs. each with a single double stranded DNA molecule and associated proteins contained in a nucleus	
Mode of division	binary fission mostly, budding	mitosis and meiosis using a spindle: followed by cytokinesis	

Bacteriology

It is a branch of microbiology dealing with the identification, study, and cultivation of bacteria and with their applications in medicine, agriculture, industry, and biotechnology.

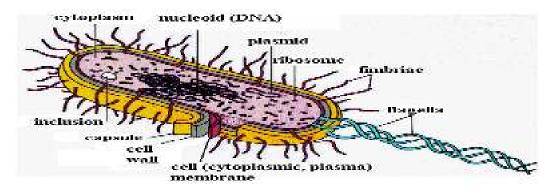
Bacterial Appearance:

- Size: The range from about $0.2 5 \mu m$ in diameter (RBC is 7um in diam.)
- **Shape:** Coccus (cocci); rod (bacillus, bacilli); spiral shapes (spirochetes; spirillum) and various shapes.
- Arrangement: Bacteria sometimes occur in groups, rather than singly.
 - Divide along a single axis, seen in pairs or chains.
 - Divide on one or more planes, producing cells in:
 - Pairs (diplococci), Chains (streptococci), Packets (sarcinae) and Clusters (staphylococci)



Bacterial structure

The typical bacterial cells have the following structures:



1- The cell envelope

The cell envelope is all the layers from the cell membrane outward, including the cell wall, the periplasmic space, the outer membrane, and the capsule.

2- The cell membrane

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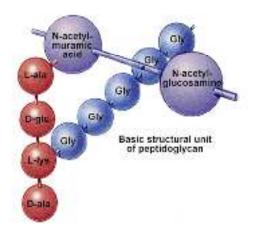
The cell membrane (often called the plasma membrane) is composed of 2 layers of phospholipids.

- Phospholipids have polar heads and non-polar tails.
 - \circ "Polar" implies that the heads are hydrophilic.
 - "Non-polar" means that the tails are hydrophobic.
- Proteins embedded in two layers of lipids (lipid bilayer)
- The membranes of prokaryotes are distinguished from those of eukaryotic cells by the absence of sterols, the only exception being mycoplasmas that incorporate sterols.
- **Function:** (1) Selective permeability and transport of solutes; (2) Electron transport and oxidative phosphorylation in aerobic species; (3) Secretion of enzymes and toxins; (4) Synthesis of precursors of DNA, cell wall polymers, and membrane lipids.

Mesosome: Invagination of the cytoplasmic membrane and it is important during cell division.

3- The cell wall

- Peptido-glycan Polymer (amino acids + sugars) : Long chains of polysaccharide crosslinked by short peptides (amino acid chains).
- Polysaccharide is composed of alternating "amino sugars" N-acetylglucosamine –NAG- and N-acetylmuramic acid- NAM-
- Unique to bacteria
- Provides structural support and maintains the shape of the cell.



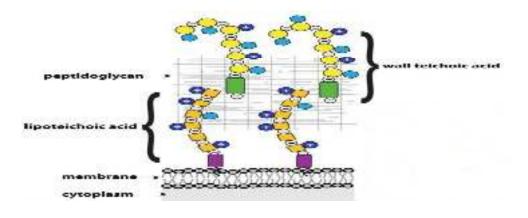
Teichoic Acids

- Bacterial copolymers of glycerol phosphate or ribitol phosphate and carbohydrates linked via phosphodiester bonds.
- Located in the outer layer of Gram +ve cell wall only, such as in the genera *Staphylococcus*, *Streptococcus*, *Bacillus*, *Clostridium*.
- There are two types of teichoic acids
 Lipoteichoic acids (LTAs): Teichoic acids that are anchored to the lipid membrane .
 Wall teichoic acids (WTA): Teichoic acids that are covalently bound to peptidoglycan

• Function

- Provide rigidity to the cell-wall.
- Mediate the attachment of some bacterial species (Staphylococci)
- It is an acidic polymer and contributes negative charge to the cell wall.

Note: In some cases the cell wall of Gram-positive bacteria may contain proteins of special significance, e.g.: **1-** The <u>M, T and R proteins</u> of the group A streptococci, 2-<u>Protein A</u> of *Staphylococcus aureus*.

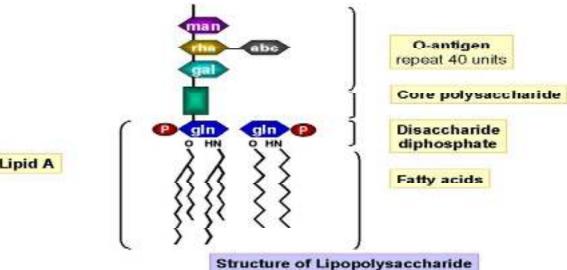


Lipopolysaccharide (LPS) (Somatic -o Antigen)

- Also known as **endotoxins**, (Fever causing) •
- Large molecules consisting of a lipid and a polysaccharide composed of O-antigen, outer core and inner core joined by a covalent bond;
- Found in the outer membrane of Gram-negative bacteria.
- Elicit strong immune responses in animals.
- Lipooligosaccharide (LOS): low-molecular-weight form of bacterial lipopolysaccharides.

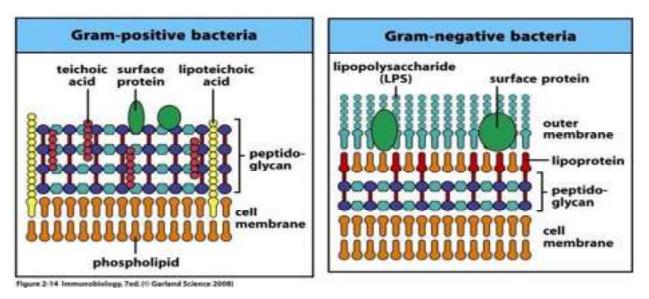
Structure

- Lipid A (phosphoplipid) Toxic component of endotoxin.
- A core polysaccharide of five sugars linked to lipid A.
- An outer Polysaccharide: Major surface Ag used in Lab diagnosis.
- Antigen of Salmonella and E. coli •
 - 2,000 different O Ag of Salmonella
 - 0 100 different O Ag of E. coli
- Ags differ in Sugars, not Lipid A.





Cell Wall of Gram-Positive & Gram-Negative bacteria



Q/What are the differences between Gram+ve and Gram -ve bacteria?

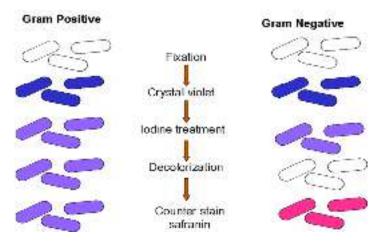
- The structure, chemical composition, and thickness of the cell wall differ in Gram positive and Gram –negative bacteria.
- Gram-positive: many layers of peptidoglycan, which is anchored to the cell membrane by teichoic acid.
- Gram-negative: 1-2 layers of peptidoglycan = thin
 - The periplasmic space is between the cell membrane and the cell wall.
 - Outside the peptidglycan layer is the "outer membrane". It is pierced by porins: protein channels, and its out surface is covered with lipopolysaccharides (sugars linked to membrane lipids), which are often antigenic and or toxic.

Q/Why are these differences in cell wall structure so important?

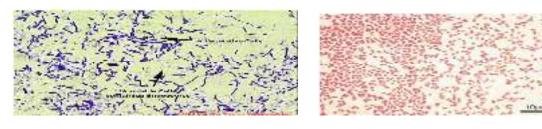
Q/ What are the applications of Gram staining?

Gram Stain (See Lab-2)

It is the most important diagnostic procedure in microbiology. It separates most bacteria into two major groups.



Stain is valuable in identification, Gram positives stain purple; Gram negatives stain pink.



Gram +ve bacteria

Gram -ve bacteria

Table-2

0	A comparison bet Gram ne
1	Item
	Peptidoglycan layer
	Teichoic acids
	Periplasmic space
	Lipopolysaccharide (LPS) content
	Lipid and lipoprotein content
	Resistance to physical disruption
l	Inhibition by basic dyes
	Susceptibility to anionic detergents
	Resistance to drying
	Gram reaction

ween Gram positive and gative cell wall

Item	Gram positive	Gram negative
Peptidoglycan layer	Thick (multilayered)	Thin (single-layered)
feichoic acids	Present	Absent
Periplasmic space	Absent	present
ipopolysaccharide (LPS) content	Virtually none	High
ipid and lipoprotein content	Low	High
Resistance to physical disruption	Low	High
nhibition by basic dyes	Low	High
Susceptibility to anionic detergents	Low	High
Resistance to drying	Low	High
Gram reaction	Retain crystal violet dye and stain dark violet	Can be decolorized to accept counter stain

4- Cytoplasm

- Has two distinct areas:
- 1- An amorphous matrix that contains ribosomes, nutrient granules, metabolites and plasmid.
- 2- An inner, nucleoid region composed of DNA.
- Site for cell's biochemical activities
- 70-80% water

Nucleoid

• The area in which DNA is located

DNA—bacterial chromosome

- Single long circular chromosome
- Double-stranded DNA
- No nuclear envelope
- May have small circular DNA plasmids outside chromosome
- **Plasmid**: This is a small (relative to the chromosome) a circular piece of DNA that often codes for virulence factors and antibiotic resistance.

Ribosome

- Site of protein synthesis.
- 70S in size with 50s and 30s subunits.
- rRNA + protein
- Inhibited by some antibiotics

Granules

- Serve as storage areas for nutrients.
- Volutin granules reserve high energy, it appear as metachromatic granules.

Actin cytoskeleton

- Found in rod-shaped and spiral bacteria just under plasma membrane
- Influences cell shape

Endospores

- The spore is a resting cell, highly resistant to desiccation, heat, and chemical agents; when returned to favorable nutritional conditions and activated the spore **germinates** to produce a single vegetative cell.
- The two most common bacteria are Gram-positive rods:
 - Obligately aerobic genus *Bacillus*
 - Obligately anaerobic genus *Clostridium*.
- Location important in classification (Central, Subterminal, Terminal)

- <u>Sporulation</u> is spore formation.
- <u>Germination</u> is return to vegetative (growing) cell.
- Spore stain can be used for demonstration of bacterial spore. (See lab)

5- Capsule (K-antigen) and Glycocalyx

- Some bacteria (often pathogens) are surrounded by a thick polysaccharide capsule.
- The **capsule** is a major virulence factor.
- This is a loose jelly-like or mucus-like layer. It helps prevent immune system cells from reaching the bacteria (prevent phagocytosis), and it forms part of biofilms.
- It composed of polysaccharide except in the anthrax bacilli (D-glutamic acid).

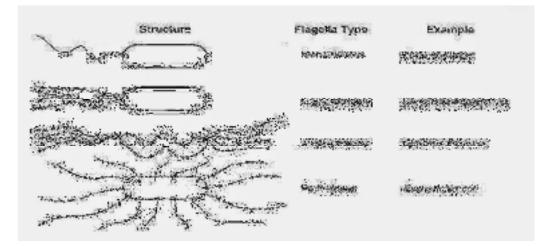
Q/ Give an example of capsulated bacteria?

All of the principal pathogens which cause pneumonia and meningitis, including *Haemophilus influenzae*, *Neisseria meningitidis*, *Escherichia coli*, *Streptococcus pneumoniae*, *Klebsiella pneumoniae* and group B streptococci have polysaccharide capsules on their surface.

- Some bacteria have an additional layer outside of the cell wall called the glycocalyx.
- It is a polysaccharide coating.
- Slime layer causes bacteria to adhere to (skin, heart valve, and catheter) and helps prevent the cell from drying out.
- The slime layer of **Gram**+ve *Streptococcus mutans* allows it to accumulate on tooth enamel; this plays an important role in the formation of plaque and dental caries.

6- Flagella (H-antigen)

- Flagella are long hairs used to propel the cells.
- They are composed of flagellin protein.
- At the base of each flagellum is a motor embedded in the membrane and cell wall.
- Flagella are the source of the **H-antigen** used in serotyping many motile species of bacteria



7- Pili (Fimbriae)

- Pili (singular = pilus) are hairs projecting from the surface.(found mainly on Gram ve)
- 4 Shorter and straighter than flagella
- **↓** They are composed of pilin protein.
- **4** Adhere bacteria to surfaces, (*E. coli* has numerous types).
- **F**-pilus; used in conjugation, (Exchange of genetic information).

Atypical cell walls

Mycobacteria (TB, leprosy) and Nocardia.
 -Very thick outer lipid layer - acid fast
 -Slow growth

4 Mycoplasma

- -Lack cell wall
- -Sterol in plasma membrane
- 📕 Archaea
 - -Wall-less or wall of varying polysaccharide and protein.

-No peptidoglycan in cell wall.

Questions

- Q1/ Does Gram stain procedure demonstrate all bacterial species?
- Q2/ Mention the medically important bacteria that cannot be seen in Gram stain?
- Q3/ Mention the importance of some bacterial cell structure as a virulence factors?
- *Mycoplasma* species lack which of the following components?
 - (A) Ribosomes
 - (B) Plasma membrane
 - (C) Both DNA and RNA
 - (D) Lipids
 - (E) Peptidoglycan

References

- Jawetz in medical microbiology
- Baily and Scott, Diagnostic microbiology
- Review of medical microbiology
- Web sites---
- Note: All figures and pictures are available freely in web during preparation of this lecture. Good luck.

