Subject: Bacterial External Structures

Lecture No.: 5

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In this sheet we'll talk about the third bacterial internal structure then we'll start with external structures, ending the chapter. (التشابترات بتخلص ونحنا قاعدين)

You see, in biology we have different rankings (taxonomic ranks) like kingdom and family etc.

- Last two are the genus (genera) and species.
- Genera include species that share many characteristics but with few differences.

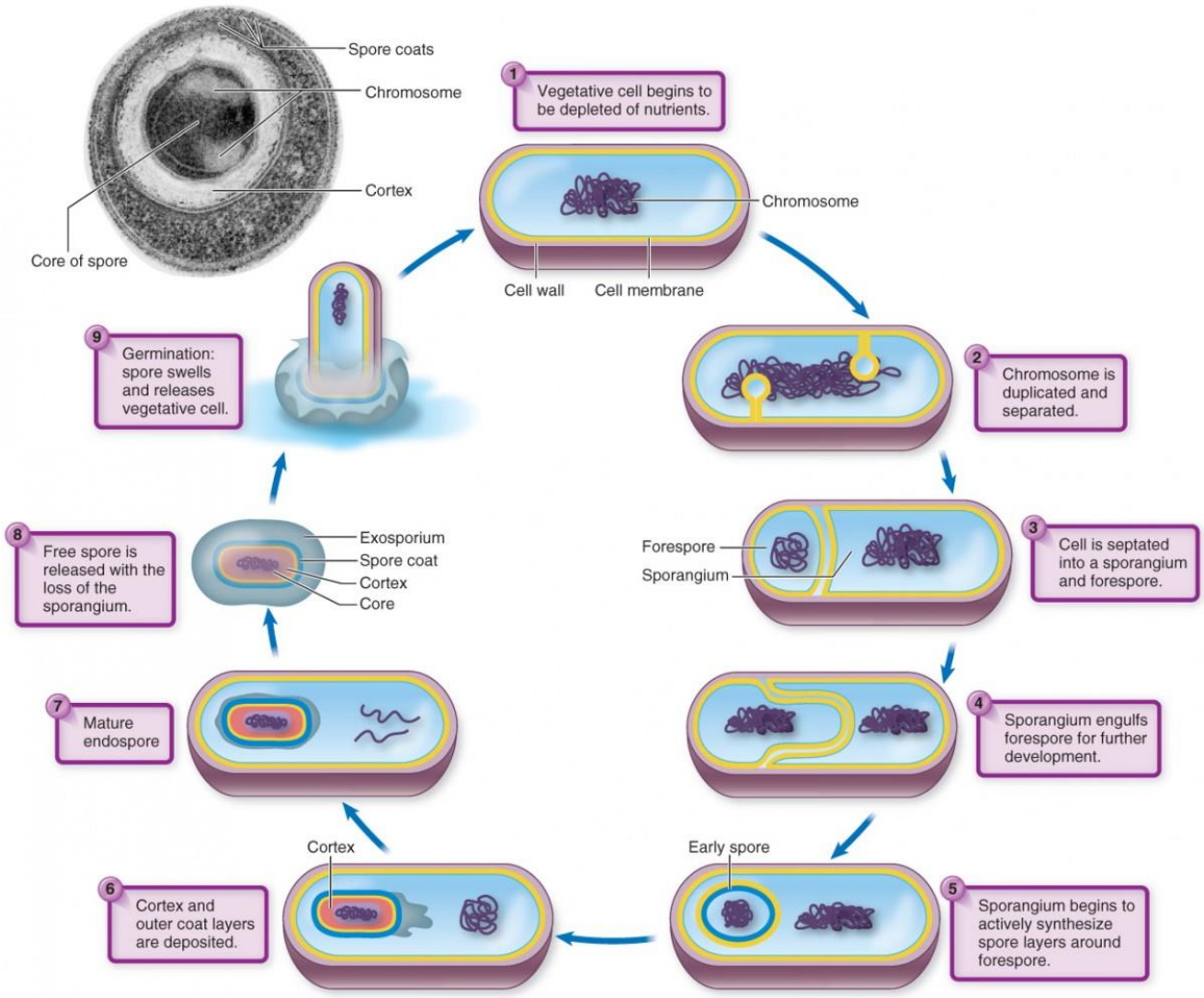
Some bacteria are known to form **spores** when exposed to harsh conditions such as nutrient depletion. We have two medically important genera, **Bacillus** and **Clostridium**, which are associated with many human diseases.

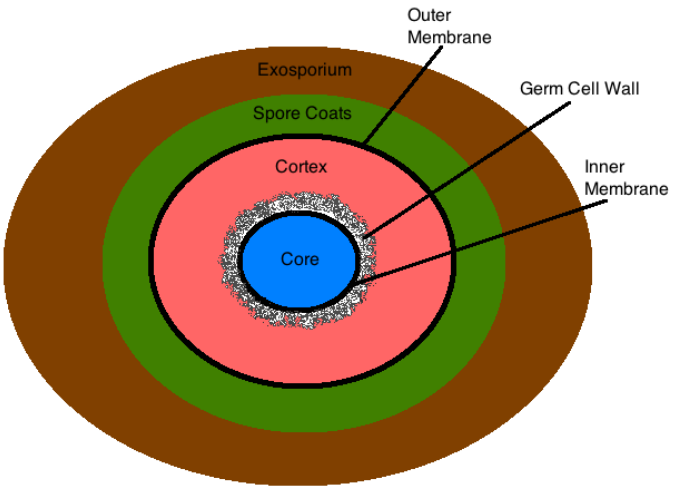
A **bacterial endospore** or an **endospore** is a dormant, tough, and non-reproductive structure produced by certain bacteria as a way of adaptation. It is a highly resistant form that preserves the cell's genetic material in times of extreme stress. Sporulation is the formation of spores (what else would it be).
Note: spores are merely a method of **survival** for bacteria, that's why they don't reproduce themselves unlike fungal spores (survival + reproduction).

How do spores form?

Note: *please watch an animation I posted on the group page, it'll help you imagine/understand what's going on here*
https://youtu.be/NAcowliknPs
https://www.youtube.com/watch?v=7zCQLITFEb0

It's a tiny-bit similar to the replication process; the cell will replicate its DNA then a septum forms. It divides the cell asymmetrically. This results in the creation of two unequal compartments, the larger mother cell and the smaller "forespore". Note that these two cells have different developmental fates. Next, the forespore is engulfed by the mother cell, **forming a cell within a cell. **Calcium dipicolinate (the calcium salt of dipicolinic acid) is incorporated into the forespore**. **The peptidoglycan cortex forms and the bacterium adds a spore coat to the outside of the forespore.** This is followed by the final dehydration and maturation of the endospore. Finally, the endospore is released into the environment. **The endospore will remain dormant until it senses the return of more favourable conditions.**





The spore coat is composed of a highly compact layer of strong structural proteins.
Both the coat and the cortex work as a permeability barrier, preventing the entry of toxic materials into the spore. They also prevent water from entering (especially the coat).
The core contains the bacterial DNA, RNA, ribosomes, calcium salts of dipicolinic acid (which are water insoluble) and most essential enzymes (like DNA polymerase). Its dehydrated state contributes to the enzymatic dormancy (metabolic inactivity) and heat resistance of the spore.
Endospores contain dipicolinic acid and a large quantity of calcium ions (Ca++). These materials appear to contribute to the heat resistance of endospores, as does their very low water content.
No water (even when enzymes are available), no metabolic activity.

Endospores have a mechanism to destroy the cortex (using enzymes called autolysins), but have no mechanism to destroy the coat. Even if an endospore is located in a normal growth medium, it cannot return to the vegetative state easily. Endospores can stay dormant for a very long time because they cannot destroy their own coat and thus, water cannot enter. That's why we need an activation step (an external factor or force that breaks the coat), and if the conditions are appropriate, water and nutrients will enter the spore because peptidoglycans are not a strict permeability barrier, thus, gene expression begins, new components are synthesized, and it returns to be a vegetative cell again.
Medically important gram negative bacteria are not known to form spores.
The bacteria that form spores are of clinical importance and are usually **gram positive** (only a limited number of gram positive bacteria), such as **bacillus** and **clostridium**.

- **Anthrax** (الجمرة الخبيثة) is caused by *bacillus anthracis.* It is infectious and spread by contact with the spores of the bacteria. Disinfectants usually don't affect them.
- **Wet gangrene** is caused by a type of *clostridium.*
- Another type is *clostridium botulinum*, It's anaerobic and lives in soil and contaminated canned food. It is one of the most toxic organisms on Earth (one gram can kill millions), since it paralyses skeletal muscles (the most important skeletal muscle is the diaphragm; if paralysed it would cause respiratory arrest). However, it can be used as a drug (botox) which is used for wrinkles. **hello, future plastic surgeons!**

External bacterial structures

1. Flagella

- Flagella are cellular projections that protrude out of the cell wall of some bacteria.
- Structure: they're made of flagellin (a protein).
- Function: movement (locomotion).
    - Bacteria that have flagella are motile and vice versa.
    - Cocci/spherical bacteria *rarely* have flagella, but in bacilli and spiral bacteria they are common.
- Presence/absence of flagella is important in identifying pathogenic bacteria.
- We also classify bacteria depending on place and number of flagella. That's why bacteria can be:
    - Atrichous: without flagella.
    - Monotrichous: with a single polar flagellum (located at one end or pole).
    - Amphitrichous: 2 flagella one at each end (both are polar).
    - Lophotrichous: bacteria with 2 or more flagella at one or both ends.
    - Peritrichous: flagella all over the surface.

- Let's get back to the function of flagella. We said that it's motility, but why should bacteria move? Easy. So that they can search for a better environment, nutrients and escape toxic materials.
- Movement of bacteria is done by a process called "chemotaxis" which is *movement of an organism in response to chemical stimulus, it has two kinds:*
    - Positive: movement toward an attractant.
    - Negative: movement away from a repellent.
- Movement can also be triggered by *light stimuli.* In this case we call it "phototaxis" which can also be positive and negative.
- In some bacterial types (i.e. spirochetes) there is a special type of flagella that is tightly bound around the cell (not extending beyond cell wall) which is called axial filament or endoflagellum. It helps in keeping the characteristic shape of these bacteria (Locomotion happens by twitching, not by swimming). (See the figure in slide 12).

2. Pili

- **Tiny** (very short) **hollow** projections composed of a protein called ‘pilin’ and used for bacterial attachment to other surfaces or other cells (unlike flagella, they are not for movement). We classify them into two groups depending on the function:
    - Attachment pili (fimbriae):
      Short type that attach bacteria to surfaces or air-water interface.
      They contribute to the pathogenicity (ability to produce disease) of certain bacteria by enhancing colonization on surface of cells of other organisms.

    - Conjugation pili (sex pili or f pili)
        - A relatively long pili found in some groups of bacteria,
          Attach 2 bacterial cells where DNA is transferred through them in a process called conjugation* (like sexual reproduction).
          Conjugation may result in the transfer of antibiotic resistance genes between bacteria horizontally (will be discussed later.)
          *Gene transfer has two major types:
          Vertical: from parents to offsprings.
          Horizontal: from one organism to another unrelated organism. It has three mechanisms: conjugation, transformation, and transduction.
          These pili are too thin to transfer normal DNA, they transfer plasmids.
          Extrachromosomal DNA: small circular DNA that we call “plasmid” that doesn’t contain essential genes, so even if the cell loses it, it’s no problem. Plasmid is medically important and carries accessory genes such as antibiotic resistance genes. Bacteria can have multiple copies of a plasmid. If a bacterium transfers a plasmid to another cell (whether it is from the same species or from another one), it transfers its resistance to antibiotics too.
          This is how vancomycin resistance was transferred from Enterococcus bacteria to staphylococcus bacteria.
          Staphylococcus Aureus bacteria are a nightmare in medical practice. Even though they are normal flora found in us without causing any disease, they are opportunistic bacteria. When there is any compromise in the skin they can cause infections and some of them are serious.

We call this type of bacteria superbugs because it has a high ability to form resistance to many antibiotics (multidrug resistance).
The first antibiotic that was discovered was Penicillin G, and it was effective against these bacteria, but in a few years, they developed resistance. Every time a new antibiotic was discovered to treat them, they developed resistance within a few years. This doesn't apply to all strains, only to certain ones, such as MRSA (Methicillin-resistant *Staphylococcus aureus),* which is resistant to most common antibiotics.
What is the ultimate solution? An antibiotic discovered in the 70s called vancomycin. By definition, if we had MRSA, without even thinking about it, doctors would use vancomycin to treat it. Until now, MRSA is not able to form resistance against vancomycin. But, where is the problem?
During the last decade, some reports (very rare ones) have shown that staphylococcus aureus became resistant to vancomycin.
These bacteria cannot in principle form resistance against vancomycin, but they got it from bacteria called Enterococcus.
That's why conjugation is important.

3. <u>Glycocalyx</u>

- It's a polysaccharide produced then secreted to the outside.
- Has two kinds:
    - Capsule
        - It is a structure outside the cell for protection and found only in certain types of bacteria. Example: Bacillus anthracis has no capsule when outside an organism, but does have it (mostly protein) when it infects animals, to **protect** itself from host defense (the immune system cannot recognize it).
        - Some cells of the immune system can now recognize the structure of the capsule, but it is still much harder to be attacked compared to normal situations.
        - It contains polysaccharide molecules arranged in loose gel with unique composition according to the strain.
        - It doesn't work as a permeability barrier.

    - Biofilm (slime)
      It is a self-attached bacterial community embedded in a self-produced polysaccharide matrix.

It makes bacteria much more tolerant to the immune system.
Capsules are defined for every cell, but a biofilm is common between many cells.
If there is a matrix, then it is a biofilm. If there is no matrix, then it is not a biofilm.
In a colony in an agar plate, there are millions of cells near each other but they don't have a slime layer so we don't call them a biofilm, but planktonic bacteria.
Planktonic: individually living.
Biofilm: bacteria are present in a community, interact and communicate with each other via certain chemicals.
Stages of biofilm formation:
It begins with the adsorption or attachment of few cells on a solid surface. These cells begin to grow and multiply. Attachment happens usually via the attachment pili, which are considered to be one of the virulence factors (they increase the pathogenicity). Multiplication begins and when a certain number of cells is reached on this surface, they switch to the second mode of growth (the biofilm) by producing the matrix (polysaccharides like a gel or glue).
These polysaccharides:
- hold the cells together and mechanical removal becomes difficult.
- help trapping nutrients.
-prevent dehydration of these cells even if we put them in harsh conditions.
-complicate the entry of antibiotics and disinfectants. The same bacteria are 100 to 1000 times more tolerant or resistant to antibiotics and disinfectants when found in biofilms than when planktonic.

To our surprise, science has revealed that more than 80% of chronic bacterial infections are associated with biofilms. That's why they are chronic and treatment is usually ineffective with them or with recurrent infections.

Dental plaque is an example on biofilms, extremely tightly bound to the tooth surface & can cause dental caries. Diabetic foot infections are other examples that require the use of higher than normal antibiotic durations.

- Less tightly bound to cell wall than a capsule
  – When present, it protects the cell against drying, helps trap nutrients & binds cells together.
  – It also allows bacteria to adhere to objects & surfaces (environment,humans).
  – Microbial biofilms are surface-associated, organised, multicellular communities held together by a self-produced extracellular matrix forming architecturally complex structures.
  – Biofilms are estimated to be implicated in around 80% of all chronic human infections and are important mediators of hospital-acquired infections.
  – Biofilms constitute a protected mode of growth and bacteria within biofilms typically exhibit significantly enhanced tolerance/resistance to antimicrobial challenge and host defences.

☺ SORRY FOR MISTAKES ☺