

Microbiology Lecture No:....1.7..... Dr Name:..Asem.Shehabi Done by:.Raneem.Bader Sheet Slide

Mrym Ghuloom



Date: 27 /10/2014



وخل الهوينا للضعيف ولا تـكن *** نؤوماً فإن الحزم ليس بنائـم

همم الأحرار تحيي الرممـــا *** نفخة الأبرار تحيي الأممــــــا

Gram-Positive Bacilli

There are three major groups of Gram-Positive Bacilli (rod-shaped):

- 1) Aerobic Bacilli : e.g. Corynebacteria / bacilli
- 2) Anaerobic Bacilli : e.g. Clostridium
- **3)** Facultative Anaerobic Bacilli : that can live under aerobic and anaerobic conditions. E.g. Corynebacteria

If you look at a gram stain cultured bacteria (gram positive bacilli), you can notice some empty spaces that indicate the formation of spores. So they are spore-forming bacilli.





This picture shows us the Pleomorphic bacilli, meaning they have different morphological structures. Therefore, in this case it is difficult to differentiate between bacilli and cocci bacteria.

Note: according to Wikipedia:

Corynebacteria in general are aerobic or facultatively anaerobic. (in the slides the dr has them as only aerobic but in the recording he says both) Corynebacterium: diphtheriae facultative anaerobic (the dr never mentions this). Please refer to the correction of this part in future posts by the correction team.

Written by

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Raneem Bader





Corynebacteria Group -Pleomorphic bacilli-

Corynebacteria are gram positive bacilli bacteria. They are usually found between the pharynx and the tonsils. They are considered part of the normal flora of the upper respiratory tract. Corynebacteria are rarely associated with clinical infections, but if associated with infections (under some special conditions) they are then considered as opportunistic pathogens.

Corynebacteria can be opportunistic pathogens in some immunodeficiency diseases or during the manipulation of the mucosa of the respiratory tract during surgical treatment. However they can cause only mild infections that can be treated using antimicrobial drugs.

There are two types of Corynebacteria:

- **1)** Non-pathogenic Corynebacteria: found in the upper respiratory-tract as part of the normal flora.
- **2)** Pathogenic Corynebacteria: can cause some diseases, e.g. the species *Corynebacterium diphtheria* that causes Diphtheria.

Corynebacterium diphtheria:

It is a highly pathogenic bacterium. It produces some dangerous and potent toxins like: diphtheria toxins.



Sheet #17

Introduction to Microbiology Dr. Asem Shehabi Date: 27 /10/2014



Corynebacterium diphtheria can be classified into a <u>toxigenic group</u> and a <u>non-toxigenic</u> group. These two strains can be found in the upper respiratory tract of any person.

Children who are not immunized with DPT vaccine are highly susceptible to Diphtheria.

Once *Corynebacterium diphtheria* is attached to the mucosa -especially to that of the pharynx-, it adheres and produces inflammatory reactions, toxins the and enzymes released by the bacteria produce severe inflammation in the pharynx and cause sore throat. The inflammation can transfer from the pharynx to the tonsils or from the tonsils to the pharynx.

Moreover, the inflammation can cause pseudomembranous inflammation reactions (severe damage of the mucosa of tonsils and pharynx). Then the Toxins produced from the bacteria will travel to the blood stream reaching the heart and affecting its muscles and some respiratory muscles which will end up in diphtheria.



What is DTP vaccine? It is a triple vaccine against <u>d</u>iphtheria, <u>t</u>etanus, and bordetella pertussisus –the causative agent of <u>p</u>ertussisus or whooping cough. It's composed of Corynebacteria diphtheria toxoids. What are Toxoids? Toxoids are inactivated toxins produced by bacteria.

** Diphtheria was eradicated in 1980



Sheet #17



How can Diphtheria be characterized?

Diphtheria is characterized by a large swelling in the neck due to the severe inflammatory reaction and the accumulation of fluid. After that, respiratory failure will occur causing some difficulties in breathing which will eventually cause death.

The potent toxin -Diphtheria Toxin-, which is produced by the <u>lysogenic strains of Corynebacterium</u> (lysogenic= a strain of bacterium that is infected with a bacteriophage, the bacteriophage integrates the toxin-encoding genetic elements into the bacteria).

So the main two causes of inflammation development are the adherence of the bacteria and the release of toxins.

Few days are enough to produce a sufficient amount of toxins to cause death.

Prevention of Diphtheria can be acquired by taking the triple vaccine (DTP). This vaccine should be taken in three doses within the 1st year (2-4-6 Months Children).

Another point to mention is that Corynebacteria diphtheria are not easily demonstrated by gram stain since they have some morphological similarities with other types of non pathogenic Corynebacteria.

Corynebacteria bacilli have at their endings metachromatic granules. These granules can be demonstrated using stains other than gram stain, e.g. Albert stain.

Within the first 24-48 hour we won't be able to differentiate between *Corynebacterium diphtheria* and *Haemophilus influenza cocci*. Therefore we culture it using tellurite blood agar which is only selective for *Corynebacterium*.





** Corynebacterium is susceptible to penicillin and other antibiotics, but the issue is to detect that this is a case of Corynebacterium diphtheria and not a case of sore throat caused by group A streptococci for example, and this needs experience.

Gram+ve Spore-forming Bacilli

Some types can live under aerobic conditions (Bacillus cereus) or under anaerobic conditions (Clostridium).

Gram-positive spore-forming bacilli (both aerobic and anaerobic) are widely distributed in nature in large numbers.

Spore-forming bacteria can contaminate food and can survive in dryness. Since they can contaminate food, they can easily reach our stomach. The number of bacilli is important to indicate the level of hygiene and the methods that keep food safe for consumption.

** Aerobic spore-forming bacilli:

1) <u>Bacillus cereus</u>

Bacillus cereus is a spore-forming bacterium.

Bacillus cereus can produce toxins. However, in order to prove that the food **intoxication** is due to this type of bacteria we have to keep in mind that this bacteria needs at least 24 hours to make a deadly amount of toxins and to contaminate food .

Bacillus cereus is rarely associated with a fatal outcome. Its toxicity is mild. In this case the patient may vomit, have muscle- spasm, dizziness and some weakness for few hours. And all these symptoms





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are not fatal in contrast with other symptoms of other types of bacteria which can be fatal.

Bacillus cereus is found in any food in large numbers. It is not dangerous unless it produces toxins.

Bacillus cereus can contaminate food especially rice when it is not well boiled (Chinese method). And that's because during rice harvesting, some spore-forming bacteria will attach to it. Also cooking rice in very large amounts and in large containers may convert spore forming bacteria to vegetative bacteria and thus it can produce toxins in 24 hours. Rice is not dangerous to eat in the first day; however, it is dangerous to eat it on the second or third day especially if it wasn't refrigerated.

Food intoxication due to the toxins released by bacillus ceres is manifested in the form of vomiting and diarrhea (in the 2nd day) but will not produce any fever. (indicators of food contamination).

** Keep in mind:

Bacillus cereus is rarely associated with death, and produces the toxins outside the body, doesn't produce enough toxins while in the intestines.

2) <u>B. subtilis:</u>

B. subtilis is another type of spore-forming bacteria. It is found in the intestines of adults and children as part of the normal flora. It is non-toxigenic to humans. However it can enter the blood stream



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during surgery and contaminate it. Also it can produce some infections in immunodeficient infants or elderly.

Therefore, *B. subtilis* is considered not to be a highly pathogenic bacterium and it has no potential for sepsis like other organism.

3) <u>B. anthracis:</u>

The name originated from the blackness of infected tissues.

B. anthracis is related to animals more than humans.

It is considered a very dangerous type of spore-forming bacteria. And it can produce a variety of toxins and extracellular products.

B. anthracis produce these extracellular products in humans especially in farmers and people who work with animals. These products can cause chronic skin lesions but not systemic infection.

B. anthracis can contaminate grass and thus reach the intestine of animals. If there is an injury of the animal's intestine, the bacteria will reach the blood stream then the liver, causing liver cirrhosis, and then the animal will die. *POOR ONE ©*

Furthermore, *B. anthracis* can be used in biological warfare. In wars they prepare a toxic form of the bacteria (small particles to be dispersed in the atmosphere and inhaled). Once inhaled, they reach the lungs and cause Pneumonia, Septicemia, severe damage to lungs and cause death (High mortality), since the damage is not easily treated with antibiotics.





Keep in mind: under normal conditions *B*. anthracis cause only skin lesions to humans and rarely reach the blood stream and cause sepsis.

Skin lesions are usually treated using surgical methods, or antibiotics. This type of bacteria is isolated using a culture medium \ agar.

The isolation and identification of any type of bacilli is not difficult; however, it is difficult to detect the toxins produced by these bacilli.

**** Anaerobic spore-forming bacteria:**

<u>Clostridia</u>

Clostridia are obligate anaerobic bacilli and they are found widely in nature. They are associated with soil and can easily contaminate our bodies and food.

Clostridia can't be dangerous under normal conditions. However, during car accidents, while using knifes or any instrument that can injure our skin or tissues, clostridia may contaminate the wound and become dangerous.

Clostridia can produce two types of diseases:

- 1) Tetanus by *Clostridium tetani*.
- 2) Gas gangrene by *Clostridium perfringens*.





<u>Clostridium tetani</u>

Clostridium tetani is the cause of tetanus. Tetanus means the spasm of the muscles.

Once *Clostridium tetani* reach the subcutaneous tissues, which have a lack of oxygen, the bacteria will be able to produce toxins within a short period. These toxins will reach the CNS and other body organs and cause tetanospasmin.

Tetanospasmin: is a complex clinical feature related to the effect of toxins –Tetanus toxin- on the CNS and on the ending of nerves reaching the muscles that cause muscle spasm.

Tetanus will cause a respiratory failure and if not treated with antibiotics and antiserum, we have to use surgical clearance of the infected part to induce aerobic conditions to prevent the increase of the bacteria and the production if toxins.

DTP vaccine reduces the severity of tetanus but sometimes the vaccine isn't enough especially when it is associated with contamination. In this case the wound should be cleared and cleaned and the damaged tissues should be removed. Also antibiotics should be given and some tetanus toxoids (inactivated toxins) so to enhance the immune response of the body.

Tetanus is not very common now but it still occurs among soldiers or those who have had car accidents.

Lab diagnosis of *Clostridium tetani*

It is not easy to prove that *Clostridium tetani* is the cause of the infection and this is because of two main reasons:





- 1. The physicians, in the Emergency department, have no time to collect proper specimens and send them to the lab.
- 2. The number of bacterial cells –clostredia- in clinical specimens is low so they're difficult to culture under anaerobic conditions.

Therefore in order to diagnose a patient with tetanus we have to focus on the clinical features and on the history of the case (car accident, contamination...etc.), or by the presence of toxins in the serum of a patient -using PCR-. But not by waiting for the bacteria to be cultured, since it is difficult to culture especially if it was with other organisms and bacteria.

** the 1st stage of treating tetanus is much easier than the 2nd stage.

We will survive...Just let the fire inside you burn brighter than the fire around you...

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