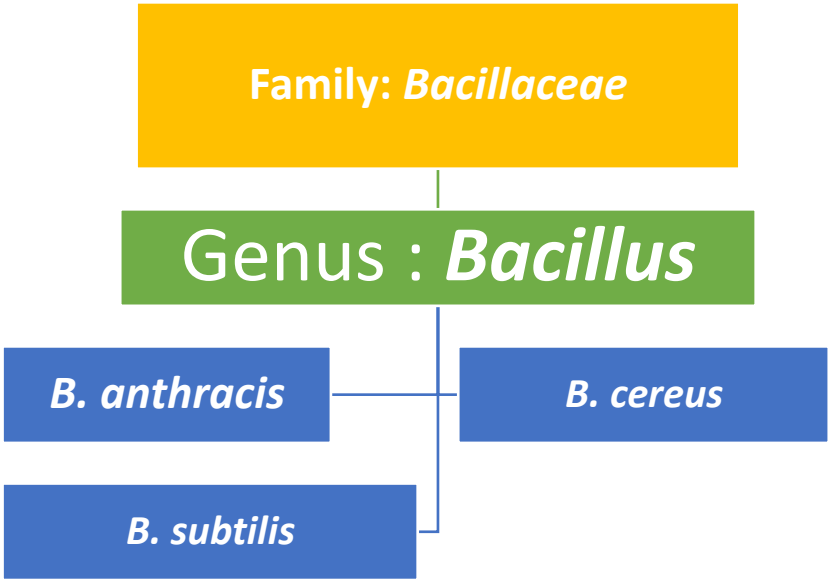


General Characteristics :

1- gram-positive rods have square ends and are arranged in long chains.

2- aerobic .

3- **spores** are located in the center of the nonmotile bacilli.

4- Most members of this genus are saprophytic organisms prevalent in soil, water, and air and on vegetation, such as *Bacillus cereus* and *Bacillus subtilis*.

Culture and Growth Characteristics:

When grown on blood agar plates, the organisms produce non hemolytic gray to white round colonies with a rough texture and have a "cut glass" appearance in transmitted light. Comma-shaped outgrowths (Medusa head) may project from the colony. Hemolysis is uncommon with *B. anthracis* but common with the saprophytic bacilli.

Pathogenesis:

- ***B. anthracis* causes anthrax:** In humans, the infection is usually acquired by the entry of spores through injured skin (cutaneous anthrax) or rarely the mucous membranes (gastrointestinal anthrax), or by inhalation of spores into the lung (inhalation anthrax).
- ***B. cereus*** cause **Food poisoning**, eye infections, severe keratitis, panophthalmitis, localized infections and systemic infections, including endocarditis, meningitis, osteomyelitis, and pneumonia.

Specimens: Specimens to be examined are fluid or pus from a local lesion, blood, and sputum.

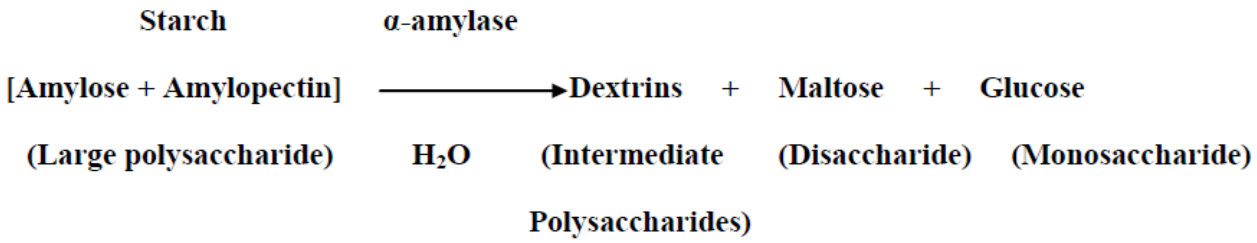
Laboratory diagnostic tests:

1. Gram stains (chains of large gram-positive rods).

2. Blood agar

3. Starch Hydrolysis: (*B. subtilis* is α-amylase positive)

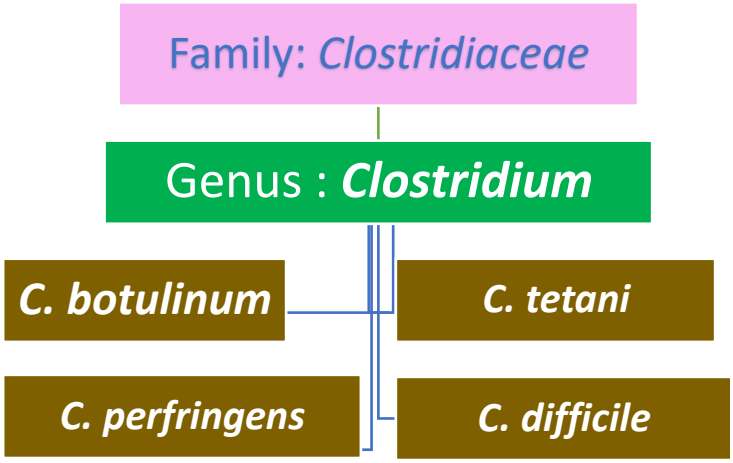
The starch molecule consists of two constituents: amylose, an unbranched glucose polymer and amylopectin, a large branched polymer. Both amylopectin and amylose are rapidly hydrolyzed by certain bacteria, using their α-amylases, to yield dextrins, maltose, and glucose, as follows:



Gram's iodine can be used to indicate the presence of starch. When it contacts starch, it forms a blue to brown complex. Hydrolyzed starch does not produce a color change. If a clear area appears after adding Gram's iodine to a medium containing starch and bacterial growth, α-amylase has been produced by the bacteria. If there is no clearing, starch has not been hydrolyzed.

Colonies of on blood agar *B. cereus*

***Bacillus* chains of large gram-positive**



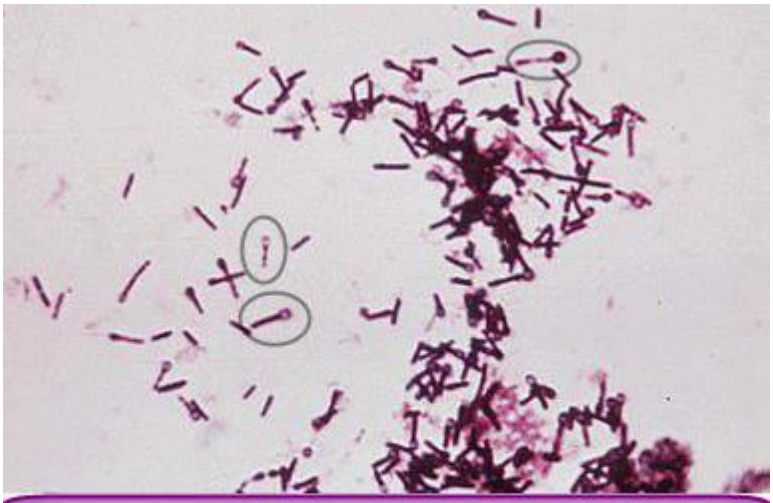
General Characteristics:

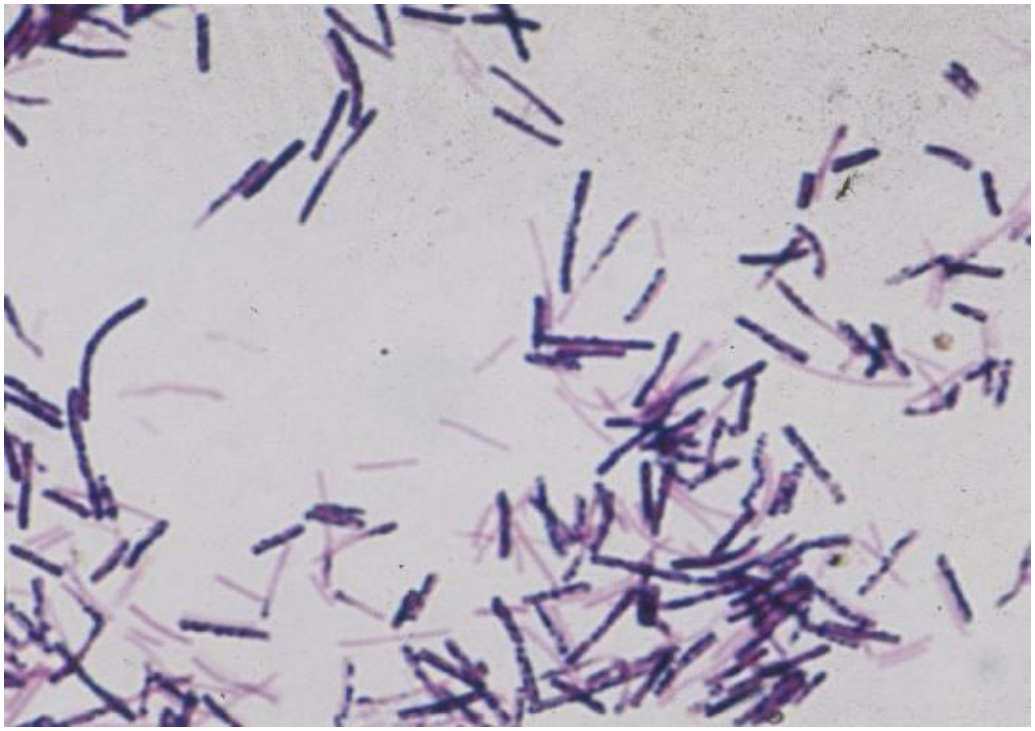
1- The clostridia are large anaerobic.
2- gram-positive.
3- motile rods. and possess peritrichous flagella.
4- Many decompose proteins or form toxins, and some do both. Their natural habitat is the soil or the intestinal tract of animals and humans, where they live as saprophytes.
5- Spores forming.

Culture and Growth Characteristics: the clostridia grow well on the blood-enriched media used to grow anaerobes and on other media used to culture anaerobes as well. Some clostridia produce large raised colonies (eg, *C. perfringens*); others produce smaller colonies (e.g, *C. tetani*). Clostridia can ferment a variety of sugars; many can digest proteins. Milk is turned acid by some and digested by others and undergoes "**stormy fermentation**" (ie, clot torn by gas) (e.g, *C. perfringens*).

Pathogenesis:

- ***C. botulinum*** causes **botulism.**
- ***C. tetani*** causes **tetanus.**
- ***C. perfringens*** can produce invasive infection (including **myonecrosis** and **gas gangrene**) if introduced into damaged tissue.
- ***C. difficile*** causes **Pseudomembranous Colitis**





***Clostridium ssp.* gram-positive rods**

Laboratory diagnostic tests:

1. ***C. botulinum:*** Toxin can often be demonstrated in serum from the patient, and toxin may be found in leftover food. Mice injected intraperitoneally die rapidly. The antigenic type of toxin is identified by neutralization with specific antitoxin in mice. *C botulinum* may be grown from food remains and tested for toxin production, but this is rarely done and is of questionable significance. Toxin may be demonstrated by passive hemagglutination or radioimmunoassay.

2. *C. tetani* :

 Specimens: wounds swb, exudates in tissue from wound, gram staining shows gram positive bacilli with drum-stick appearance.

 Culture: Specimens are inoculated on the blood agar or on cooked meat medium under anaerobic conduction, *C. tetani* produce swarming growth after 1-2 days of incubation.

3. *C. perfringens*:

Specimens: Specimens consist of material from wounds, pus, and tissue. The presence of large gram-positive rods in Gram-stained smears suggests gas gangrene clostridia; spores are not regularly present.

 Culture: Material is inoculated into **chopped meat-glucose** medium and **thioglycolate** medium and onto **blood agar** plates incubated anaerobically. *C perfringens* rarely produces spores when cultured on agar in the laboratory.

 Haemolysis on blood agar

 Litmus milk reaction: The growth from one of the media is transferred into milk. A clot torn by gas in 24 hours is suggestive of *C. perfringens.*

 biochemical reactions (various sugars in thioglycolate)

 Lecithinase activity is evaluated by the precipitate formed around colonies on egg yolk media.
 Final identification **rests on toxin production and neutralization by specific antitoxin.**