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Spore-Forming Gram-Positive Bacilli

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Textbook of Diagnostic Microbiology (Mahon, Textbook of Diagnostic Microbiology), Connie R. Mahon MS, Donald C. Lehman EdD
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Mims' Medical Microbiology and Immunology, International Edition, Goering.

Spore-Forming Gram-Positive Bacilli:

Bacillus and Clostridium Species:

The gram-positive spore-forming bacilli are the *Bacillus* and *Clostridium* species. These bacilli are ubiquitous, and because they form spores, they can survive in the environment for many years. *Bacillus* species are aerobes and the *Clostridium* species are anaerobes.

BACILLUS SPECIES

Most members of this genus are saprophytic organisms prevalent in soil, water, and air, and on vegetation. This organism is also capable of causing disease in humans. *B. cereus* can grow in foods and cause food poisoning by producing either an enterotoxin (diarrhea) or an emetic toxin (vomiting). Both *B. cereus* and *B. thuringiensis* may occasionally produce disease in immunocompromised humans (eg, meningitis, endocarditis, endophthalmitis, conjunctivitis, or acute gastroenteritis). *B. anthracis*, which causes anthrax, is the principal pathogen of the genus.

BACILLUS ANTHRACIS

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**). A fourth category of the disease, injection anthrax, has caused outbreaks among persons who inject heroin that has been contaminated with anthrax spores. The spores germinate in the tissue at the site of entry, and growth of the vegetative organisms results in formation of a gelatinous edema and congestion. Bacilli spread via lymphatics to the bloodstream, and

they multiply freely in the blood and tissues shortly before and after the animal's death. *B. anthracis* isolates that do not produce a capsule are not virulent. The poly- γ -d-glutamic acid capsule is antiphagocytic.

Anthrax toxins are made up of three proteins,

- ☒ protective antigen (PA).
- ☒ edema factor (EF).
- ☒ and lethal factor (LF).

PA is a protein that binds to specific cell receptors, and after proteolytic activation, it forms a membrane channel that mediates entry of EF and LF into the cell. EF is an adenylate cyclase; with PA, it forms a toxin known as edema toxin. Edema toxin is responsible for cell and tissue edema. LF plus PA form lethal toxin, which is a major virulence factor and cause of death in infected animals and humans.

Antigenic structure:

- Capsule: Polypeptide of D-glutamic acid: Enables the organism to evade phagocytosis.
- Polysaccharide somatic antigen.
- Protein somatic antigen
- Anthrax toxin

Pathology

Acquired by the entry of spores through injured skin in cutaneous anthrax, or mucus membrane in intestinal anthrax, or inhalation of spores in the lung while handling skin and hides There are four forms of anthrax:

1. Cutaneous anthrax (Malignant pustule): 95 % of anthrax presentation Characterized by a black necrotic lesion with a definite edematous margin on hands, arms, face or neck with regional lymphadenitis associated systemic symptoms.

2. Pulmonary anthrax (Wool sorter's disease):5% of anthrax presentation Presents with substernal pain, cough with haemorrhagic mediastinitis; and fatal if not treated early.

3. Bacteremia anthrax: presents with clinical features of sepsis.

4. Intestinal anthrax: Presents with abdominal pain, vomiting, and bloody diarrhea, intestinal anthrax are rare to occur.

Epidemiology, Prevention, and Control

Soil is contaminated with anthrax spores from the carcasses of dead animals. These spores remain viable for decades. Perhaps, spores can germinate in soil at a pH of 6.5 at proper temperature. Grazing animals infected through injured mucous membranes serve to perpetuate the chain of infection. Contact with infected animals or with their hides, hair, and bristles is the source of infection in humans. Control measures include (1) disposal of animal carcasses by burning or by deep burial in lime pits, (2) decontamination (usually by autoclaving) of animal products, (3) protective clothing and gloves for handling potentially infected materials, and (4)

active immunization of domestic animals with live attenuated vaccines. Persons with high occupational risk should be immunized.

BACILLUS CEREUS

B. cereus causes food poisoning, especially when reheated, contaminated rice is eaten.

General characteristics:

- Exhibit motility by swarming in semisolid media
- Produce β lactamase, so not sensitive to penicillin
- Produce exotoxin

Pathogenicity

1. Food poisoning:

a. Emetic type food poisoning: which is associated with rice, milk, and pasta. *B. cereus* produces toxins that cause disease. IP is 1-5 h after ingestion of preformed toxin contaminating rice and pasta dishes characterized by nausea, vomiting, abdominal cramps, and self-limited within 24 h.

b. Diarrheal type food poisoning: which is associated with meat dishes and sauces. IP is 1-24 h after ingestion of contaminated meat dishes with sporulation or preformed toxin. Characterized by profuse diarrhea and abdominal cramps. Fever and vomiting is uncommon.

2. Ocular infection: Ocular disease following trauma from non-surgical penetrating objects Manifests with keratitis, endophthalmitis, and panophthalmitis

In this syndrome, ingested spores that develop into vegetative cells of *B. cereus* secrete one of three possible enterotoxins which induce fluid accumulation and other physiological responses in the small intestine.

CLOSTRIDIUM SPECIES

Genus: *Clostridium*

Characteristics:

- Clostridia are anaerobic, spore-forming motile, gram-positive rods.
- Most species are soil saprophytes but a few are pathogens to human.
- They inhabit human and animal intestine, soil, water, decaying animal and plant matter.
- Spores of clostridia are wider than the diameter of organism and located centrally, subterminally and terminally
- Species of medical importance: *C. perfringens*, *C. tetani*, *C. botulinum*, *C. difficile*.

Clostridium perfringens

- Capsulated, non-motile, short gram-positive rods.
- There are five toxigenic groups : A-E.
- Human disease is caused by *C. perfringens* type A and C.

- Spores are found in the soil, and vegetative cells are normal flora of the colon and vagina. This bacterium causes two discrete diseases, due to either exogenous or endogenous infection.

Virulence factors

1. Enzymes: Digest collagen of subcutaneous tissue and muscle.

- Collagenase
- Proteinase
- Hyaluronidase
- DNase

Toxins

- Phospholipase C (α toxin) It has lethal, necrotizing and hemolytic effect on tissue. It causes cell lysis due to lecithinase action on the lecithin which is found in mammalian cell membrane.
- Theta toxin: It has hemolytic and necrotic effect on tissue.
- Enterotoxin

Pathogenicity

1. Clostridial myonecrosis (Gas gangrene): Incubation period 1-3 days colonization of traumatized wound by *C. perfringens* spores, and organism germination and release of toxins. Muscle and subcutaneous tissue necrosis

and foul smelling wound discharge fever, toxemia, hemolytic anemia and shock.

2. Clostridial food poisoning: It causes secretory diarrhea due to release of enterotoxin in the intestine Self-limiting diarrhea similar to that produced by *B. cereus*.

Clostridium tetani

General characteristics:

- Longer and thinner gram-positive rods with round terminal spores giving characteristic “drum-stick” appearance

There are ten antigenic types of *C. tetani* but all produce the same neurotoxin. The toxin has two components:

1. Tetanospasmin: Neurotoxic property
2. Tetanolysin: Hemolytic property

Pathogenicity

The agent of tetanus (lockjaw), which is a typical toxin mediated disease. The powerful, heat-labile neurotoxin (tetanospasmin) is produced at the wound site and released during cell lysis . It is retrogradely carried via the peripheral nerves to the central nervous system where it blocks inhibitory mediators at spinal synapses. This causes sustained muscle spasm and the characteristic signs of spasm of jaw muscles (lockjaw) and facial muscles (Risus sardonicus). *C. tetani* also produces an oxygen-labile haemolysin (tetanolysin).

Clostridium botulinum

General characteristics:

- Spores of *C. botulinum* are widely distributed in soil, they often contaminate vegetables, fruits and other materials.
- Produce a neurotoxin which is the most active known poison, and considered to be the major agent of bioterrorism and biologic warfare
- There are seven serotypes (A-G) of which A,B and E are the principal causes of human illness.
- The agent of botulism, a form of food poisoning, has powerful toxins that can be used in bioterrorism and warfare.

All the serotypes interfere with neural transmission by blocking the release of acetylcholine, which is the principal neurotransmitter at the neuromuscular junction.