

GRAM POSITIVE ROD

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Medically Important Gram-Positive Bacilli

Three general groups:

1. Endospore-formers

Bacillus, Clostridium

2. Non-endospore-formers

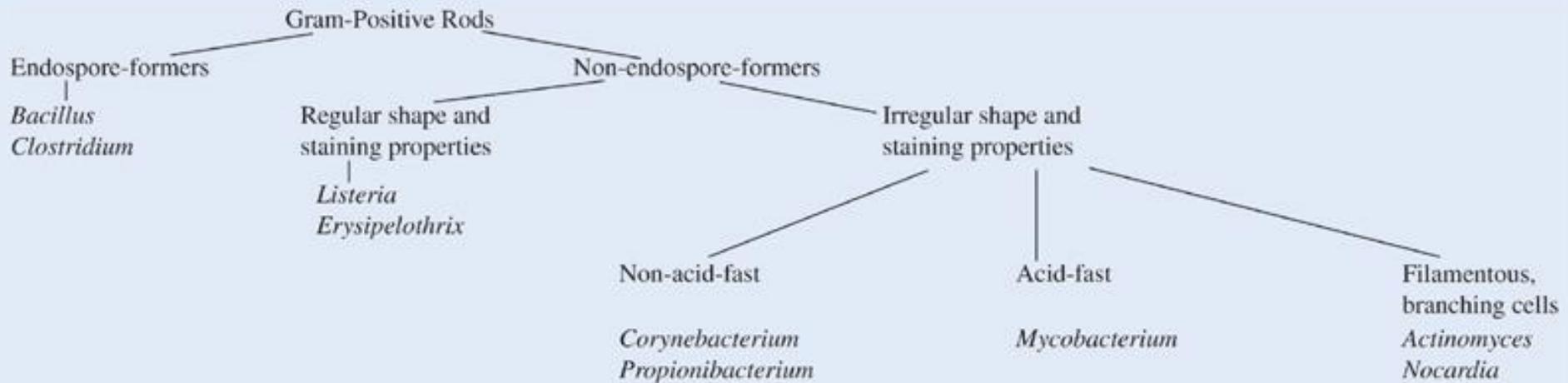
Listeria, Erysipelothrix

3. Irregular shaped and staining properties

*Corynebacterium, Propriobacterium,
Mycobacterium, Actinomyces, Nocardia*

TABLE 19.1

Gram-Positive Bacilli



Spore-forming Bacilli

Genus *Bacillus*

Genus *Clostridium*

General Characteristics of the Genus *Bacillus*

- Gram-positive, endospore-forming, rods
- Mostly saprobic
- Aerobic and catalase positive
- Primary habitat is soil
- 2 species of medical importance:
 - *Bacillus anthracis*
 - *Bacillus cereus*

***Bacillus* species are aerobes, the *Clostridium* species are anaerobes.**

Anthrax, is caused by *bacillus anthracis*.

Because of its potent toxins, *B anthracis* is a major potential agent of bioterrorism and biologic warfare.

***Bacillus cereus* and *Bacillus thuringiensis* cause food poisoning and occasionally eye or other localized infections.**

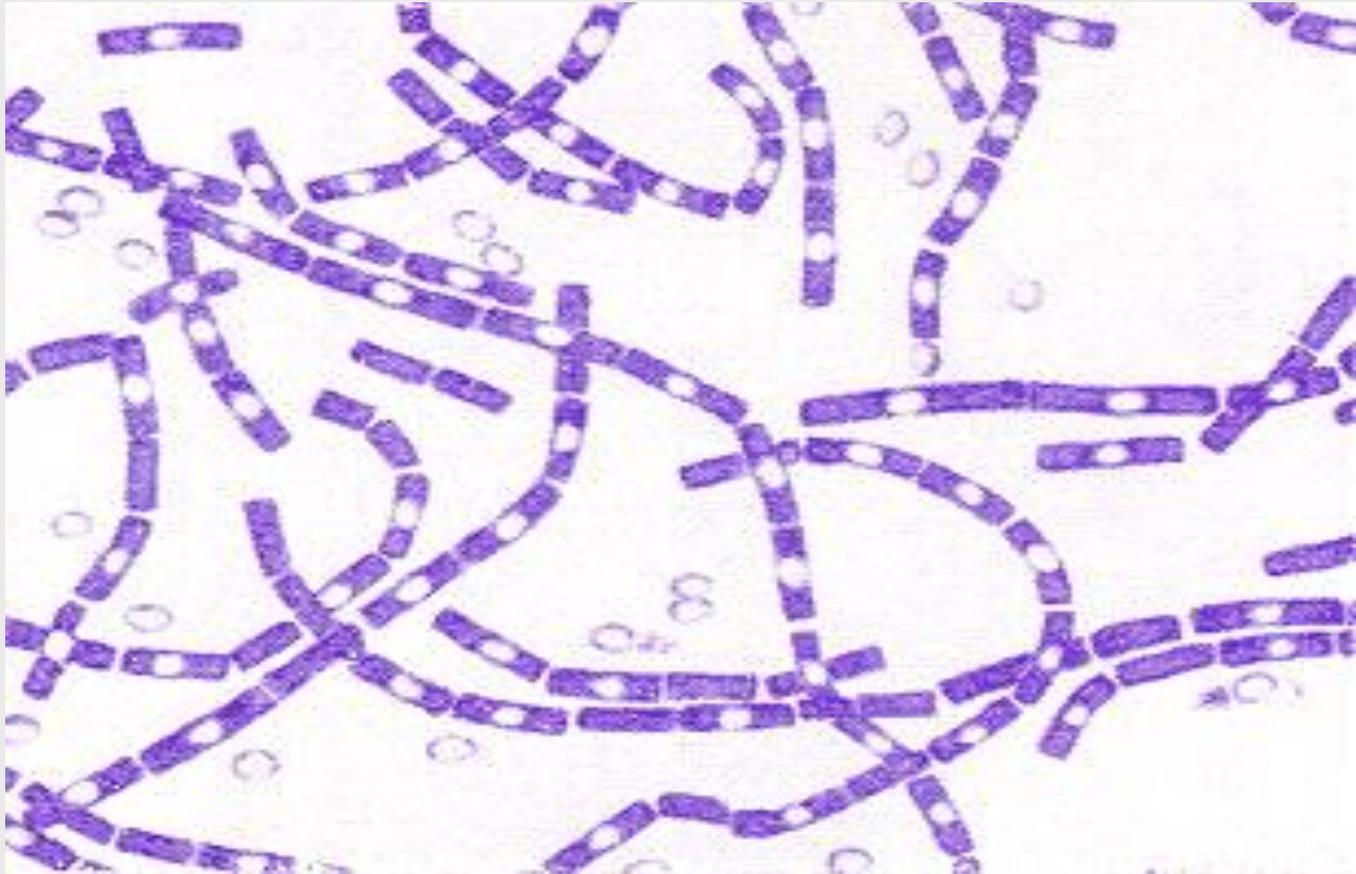
***Clostridium tetani*, tetanus**

***Clostridium botulinum*, botulism**

***Clostridium perfringens*, gas gangrene**

***Clostridium difficile*, pseudomembranous colitis.**

Colonies of *B anthracis* are round and have a “cut glass” appearance.



Bacillus anthracis

- Large, block-shaped rods
- Central spores that develop under bad conditions except in the living body
- Virulence factors – polypeptide capsule and exotoxins
- 3 types of anthrax:
 - **Cutaneous** – spores enter through skin
 - **Pulmonary** –inhalation of spores
 - **Gastrointestinal** – ingested spores

- In inhalation anthrax (**Woolsorters' disease**), the spores from the dust of wool, hair, or hides are inhaled; phagocytosed in the lungs; and transported to lymph node where germination occurs.
- This is followed by toxin production and the development of hemorrhagic and sepsis, which are rapidly fatal.

➤ **B. anthracis produce**

1. Capsule

2. Anthrax toxins that are made up of three proteins

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

PA binds to specific cell receptors, and it forms a channel that mediates entry of EF and LF into the cell.

EF is an adenylate cyclase (edema toxin).

LF plus PA is the major virulence factors that cause of death in infected animals and humans.

Bacillus cereus

- Common airborne and dustborne; usual methods of disinfection and antiseptics are ineffective
- Grows in foods, spores survive cooking and reheating
- Ingestion of toxin-containing food causes nausea, vomiting, abdominal cramps and diarrhea; 24 hour duration

Food poisoning caused by *B. cereus* has two distinct forms

- 1. Emetic type, which is associated with fried rice**
- 2. Diarrheal type, which is associated with meat dishes and sauces.**

***B. cereus* produces toxins that cause disease that is more an **Intoxication** than a **Foodborne infection**.**

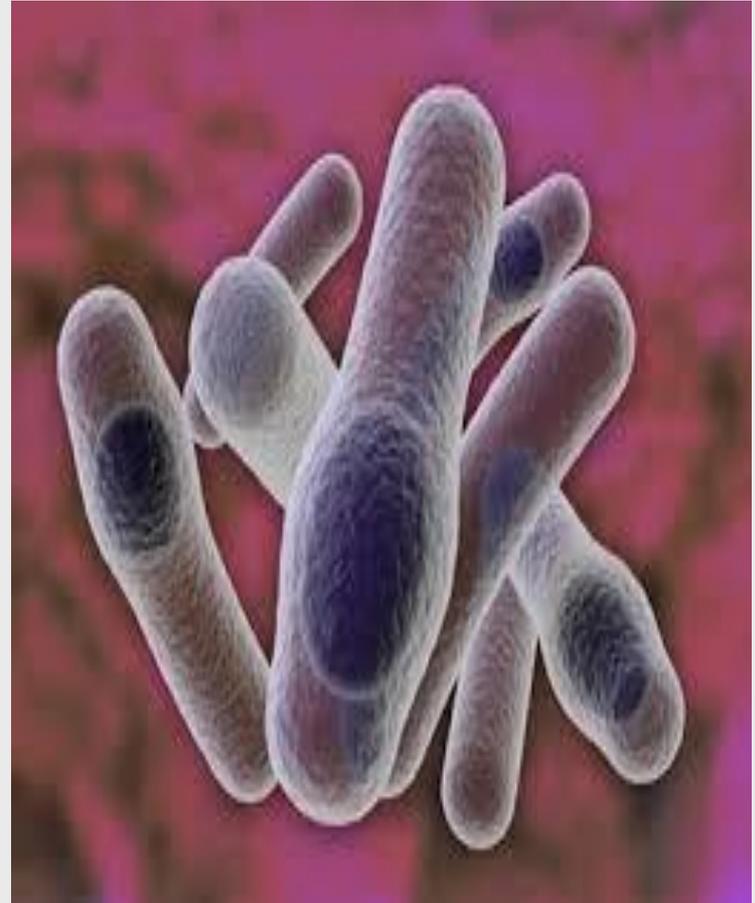
The Genus *Clostridium*

- Gram-positive, spore-forming rods
- Anaerobic and catalase negative
- 120 species
- Oval or spherical spores produced only under anaerobic conditions
- Synthesize organic acids, alcohols, and exotoxins
- Cause wound infections, tissue infections, and food intoxications

- *Clostridium perfringens*

Most frequent clostridia involved in soft tissue and wound infections - **myonecrosis**

- Spores found in soil (sub terminal)
- Predisposing factors – surgical incisions, compound fractures, diabetic ulcers, septic abortions, puncture wounds, gunshot wounds
- Virulence factors
 - toxins : alpha toxin – causes RBC rupture, a necrotizing that cause edema and tissue destruction
 - collagenase
 - hyaluronidase
 - DNase



Clostridium perfringens

(1) gas gangrene.

- All types of *C perfringens* produce the alpha toxin.
- Gas gangrene occurs when a soft tissue wound is contaminated by *C perfringens*.
- Once infection is initiated, the organisms elaborate necrotizing toxins; CO₂ and H₂ accumulate in tissue, edema occurs
- Therapy involves surgical removal of the infection and administration of penicillin G.



(2) Food poisoning

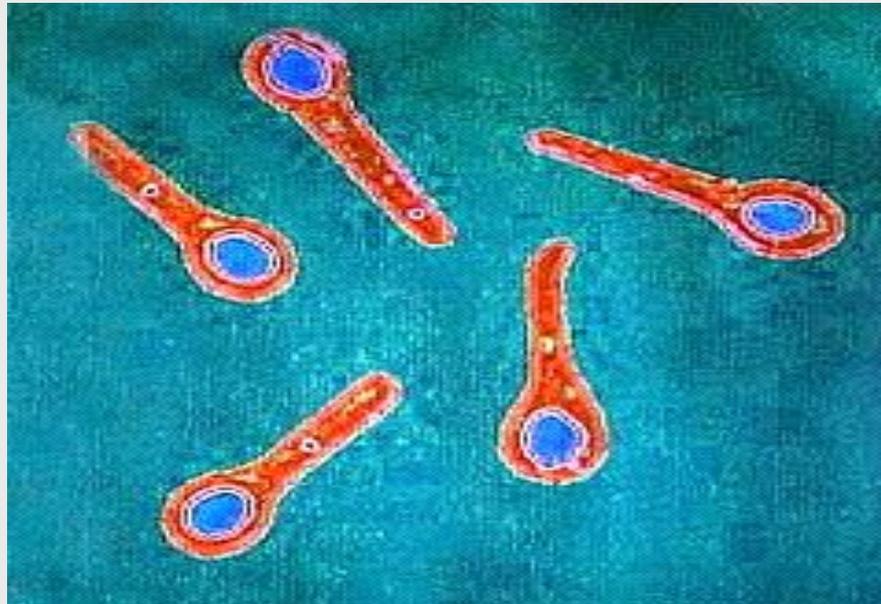
- **Enterotoxin produced and released during sporulation. The incubation period for the abdominal pain, nausea, and acute diarrhea is 8–24 hours.**

Clostridium difficile

- **Caused Pseudomembranous colitis.**
- **It is part of the normal gastrointestinal flora in 2–10% of humans.**
- The organisms are relatively resistant to most commonly used antibiotics.
- Associated with or following antibiotic use, the normal gastrointestinal flora is suppressed and *C difficile* proliferates, producing cytopathic toxin and enterotoxin.
- Symptoms of the disease vary from diarrhea alone to necrosis of mucosa with accumulation of inflammatory cells and fibrin, which forms the pseudomembrane.



- *Clostridium tetani*
- Common resident of soil and GI tracts of animals
- Causes tetanus or **lockjaw**, a neuromuscular disease
- Most commonly among geriatric patients and IV drug abusers; neonates in developing countries



- Spores of *Clostridium tetani*, which causes tetanus, are present throughout the environment.
 - They germinate in tissue and elaborate the toxin **tetanospasmin, a potent neurotoxin.**
 - The toxin spreads along nerves to the central nervous system, where it binds to gangliosides, **suppresses the release of inhibitory neurotransmitters (GLYCINE AND GAMMA-AMINO BUTYRIC ACID) Across the synaptic cleft, and yields prolonged muscle spasms.**
 - Upper airway obstruction or involvement of the diaphragm, and organ dysfunction has emerged as the major cause of death.
 - More than 50% of tetanus cases follow minor injuries.
 - Tetanus is totally preventable: active immunity is induced with tetanus toxoid (formalinized tetanus toxin). Tetanus toxoid is part of routine childhood **DTaP** (diphtheria, tetanus, acellular pertussis) immunizations.

Clostridial Food Poisoning

- *Clostridium botulinum* – rare but severe intoxication usually from home canned food
- *C. botulinum* is distributed throughout the environment. The spores find their way into preserved or canned foods with low oxygen levels and nutrients that support growth.
- Seven antigenic varieties of toxin (A–G) are known. Types A, B, E, and F are the principal causes of human illness.
- Botulinum neurotoxins are the most potent toxins known. It is heat-labile, so properly heated food does not transmit botulism.

- Toxin is ingested and absorbed. **It acts on the peripheral nervous system blocks the release of acetylcholine, necessary for muscle contraction to occur (flaccid paralysis).**
- Once the toxin is bound, the process is irreversible. The symptoms include **dysphagia, dry mouth, diplopia, and weakness or inability to breath.**
- Botulism should be treated with antitoxin.
- **Infant botulism follows the ingestion of spores. Honey is a common vehicle for spread of the spores in infants.**
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Gram-Positive Irregular Non-Spore-Forming Bacilli

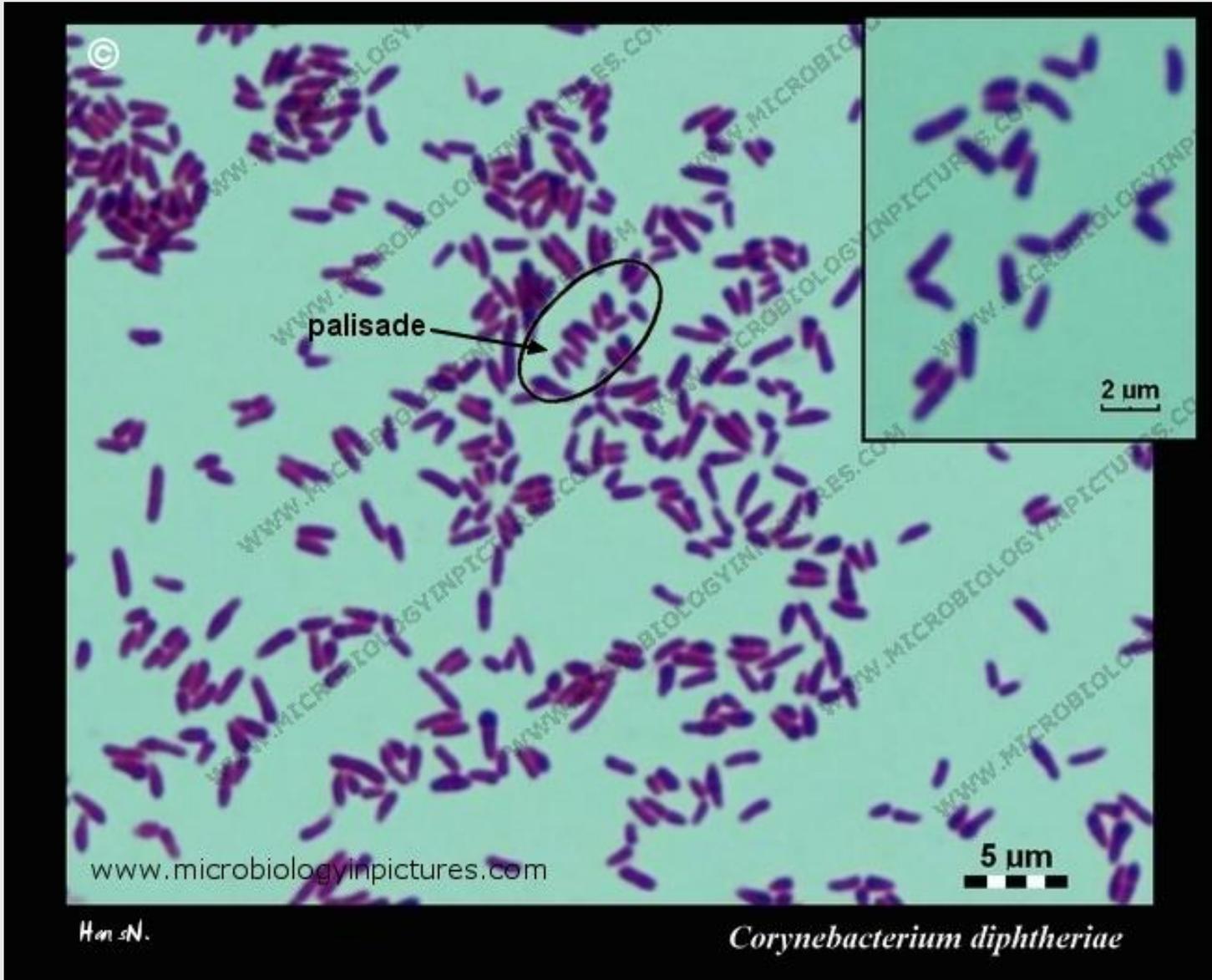
Medically important genera:

- *Corynebacterium*
- *Propriobacterium*
- *Mycobacterium*
- *Actinomyces*
- *Nocardia*

- Pleomorphic; stain unevenly
- 20 genera; *Corynebacterium*, *Mycobacterium*, and *Nocardia* greatest clinical significance
- Some possess mycolic acids

Corynebacterium diphtheriae

- Gram-positive irregular bacilli (Chinese letters)
- Virulence factors assist in attachment and growth.
 - diphtherotoxin – exotoxin
 - 2 part toxin – part B binds and induces endocytosis; part A inhibit protein synthesis



Han sN.

- Aerobic or facultatively anaerobic
- Small, pleomorphic (club-shaped), gram-positive bacilli that appear in short chains (“V” or “Y” configurations) or in clumps resembling “Chinese letters”
- Cells contain metachromatic granules (visualize with methylene blue stain)
- Lysogenic bacteriophage encodes for potent exotoxin in virulent strains

Pathogenic Corynebacterial Species

- *Corynebacterium diphtheriae*
- *Corynebacterium jeikeium*
- *Corynebacterium urealyticum*

Corynebacterium urealyticum

- Urinary tract infections (UTI's); rare but important
- Urease hydrolyzes urea; release of NH_4^+ , increase in pH, alkaline urine, renal stones

Corynebacterium diphtheriae

- Respiratory diphtheria (pseudomembrane on pharynx) and cutaneous diphtheria
- Prototype A-B exotoxin acts systemically
 - Toxoid in DPT and TD vaccines
- Selective media: cysteine-tellurite; serum tellurite; Loeffler's media
- *C. gravis*, *C. intermedius*, and *C. mitis* differentiated by colonial morphology

Epidemiology and Pathology

- Reservoir of healthy carriers; potential for diphtheria is always present
- Most cases occur in non-immunized children living in crowded, unsanitary conditions.
- Acquired via respiratory droplets from carriers or actively infected individuals

2 stages of disease:

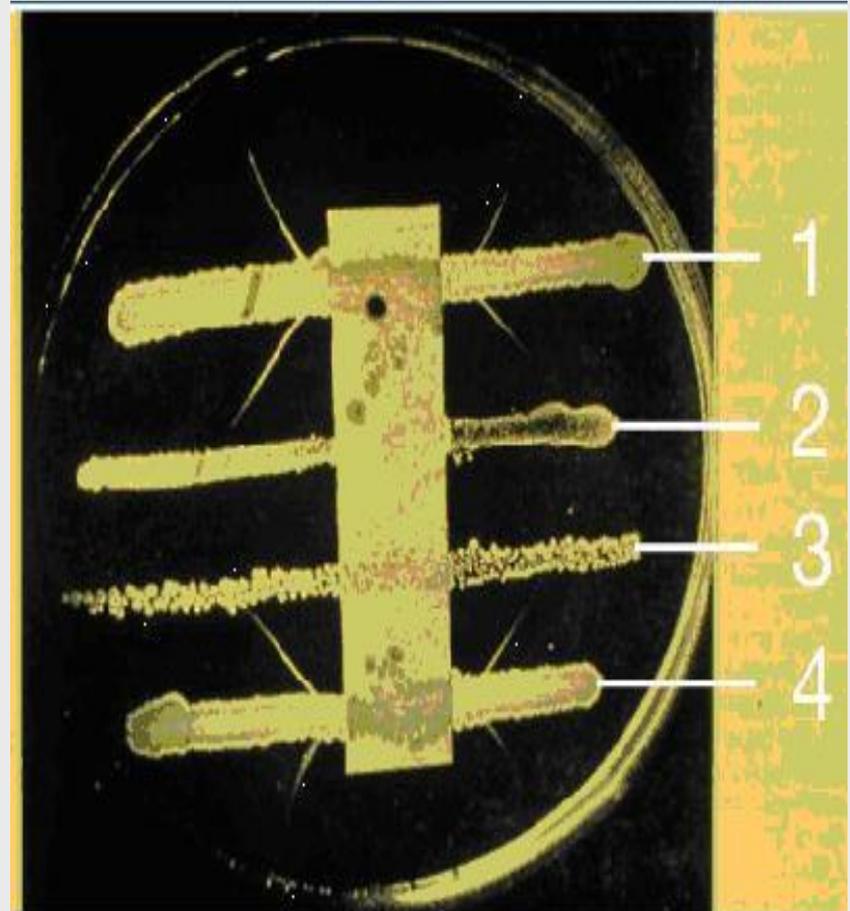
1. Local infection –upper respiratory tract inflammation
 - sore throat, nausea, vomiting, swollen lymph nodes;
pseudomembrane formation can cause asphyxiation
2. **Diphtherotoxin** production and toxemia
 - target organs primarily heart and nerves

Diagnostic Methods

- Pseudomembrane and swelling
- Staining
- Elick test
- Serological assay

Treatment and Prevention

- Antitoxin
- Penicillin or erythromycin
- Prevented by toxoid vaccine series and boosters



Mycobacteria: Acid-Fast Bacilli

- The mycobacteria are rod-shaped, aerobic bacteria
- Although they do not stain readily, after being stained, they resist decolorization by acid and are therefore called “acid-fast” bacilli”. The **Ziehl-Neelsen technique** of staining is employed
- *Mycobacterium tuberculosis* causes tuberculosis
- *Mycobacterium leprae* causes leprosy.
- *Mycobacterium avium-intracellulare* and other nontuberculous (NTM) mycobacteria frequently infect patients with AIDS

Genus *Mycobacterium*

- Mycobacteria are rich in lipids. These include mycolic acids ,waxes, and phosphatides.
- Strict aerobes
- Produce catalase
- Do not form capsules, flagella or spores
- Grow slowly

Acid-fast bacilli



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Epidemiology of Tuberculosis

- Predisposing factors include: inadequate nutrition, debilitation of the immune system, poor access to medical care, lung damage, and genetics.
- Estimate 1/3rd of world population and 15 million in U.S. carry tubercle bacillus
- Bacillus very resistant; transmitted by airborne respiratory droplets

PATHOGENESIS

- **Mycobacteria are emitted in droplets <25 m in diameter when infected persons cough, sneeze, or speak.**
 - **The droplets evaporate leaving organisms to be deposited in alveoli.**
 - **Once inside the alveoli, the host's immune system responds by release of cytokines and lymphokines that stimulate monocytes and macrophages.**
 - **Mycobacteria begin to multiply within macrophages.**
 - **After 1–2 months of exposure, pathogenic lesions associated with infection, appear in the lung.**

Pathology

Two principal lesions

1. **Exudative type**—this consists of an acute inflammatory reaction, with edema fluid, polymorphonuclear leukocytes, and, later, monocytes around the tubercle bacilli (in lung). It may lead to massive necrosis of tissue; or it may develop into the second (productive) type of lesion.

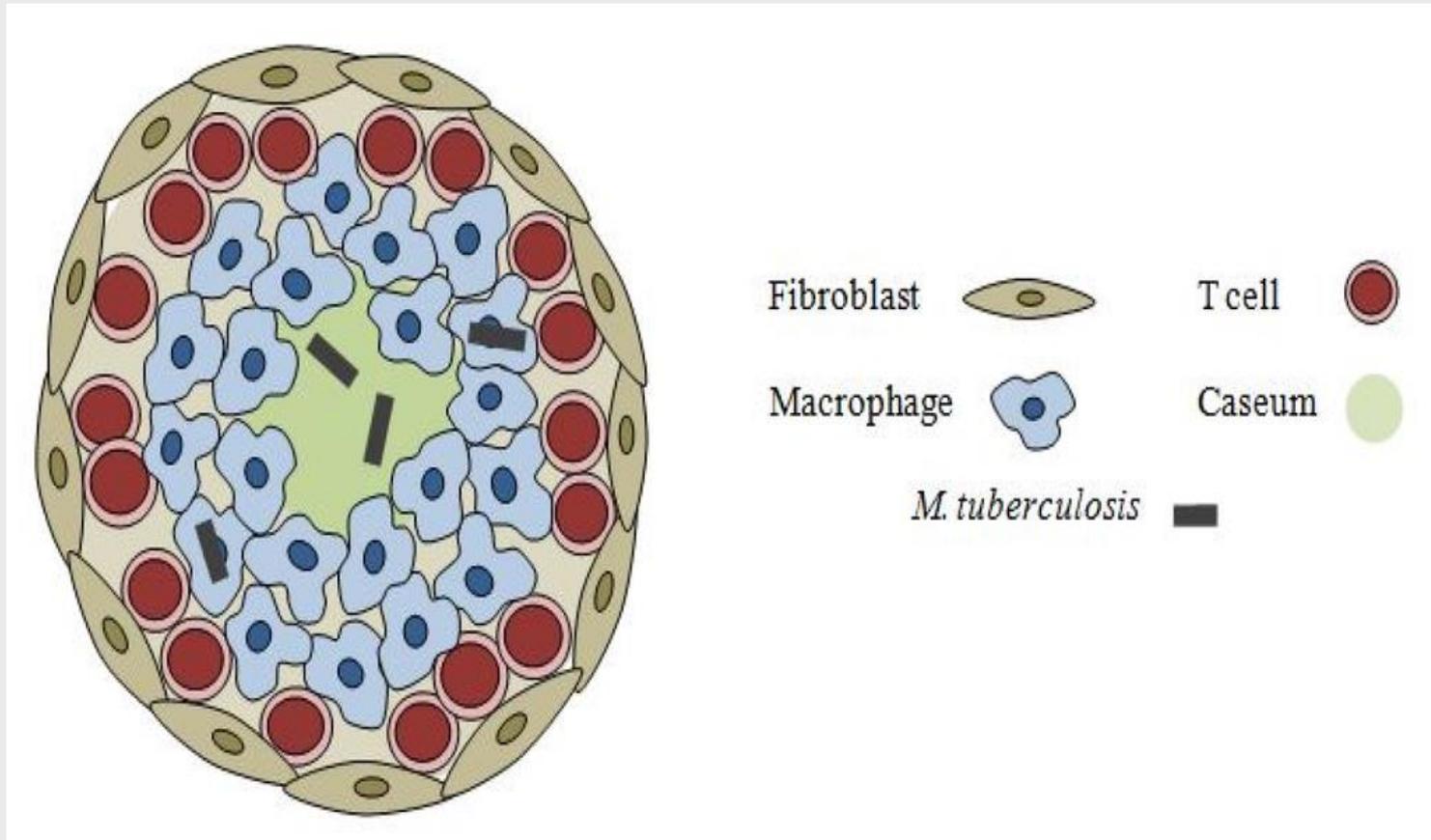
2. **Productive type**—when fully developed, this lesion, a chronic granuloma, consists of three zones:

(1) a central area of large, multinucleated giant cells containing tubercle bacilli

(2) a mid zone of pale epithelioid cells

(3) a peripheral zone of fibroblasts, lymphocytes, and monocytes.

Later, peripheral fibrous tissue develops, and the central area undergoes caseation necrosis. Such a lesion is called a tubercle. A caseous tubercle may break into a bronchus, empty its contents there, and form a cavity.



Course of Infection and Disease

- In the first infection, tubercle bacilli always spread from the initial site via the lymphatics to the regional lymph nodes.
- **The bacilli may spread farther and reach the bloodstream, which in turn distributes bacilli to all organs (miliary distribution).**
- Clinical tuberculosis divided into:
 - primary tuberculosis
 - secondary tuberculosis (reactivation or reinfection)
 - disseminated tuberculosis

Primary TB

- Infectious dose **10 cells**
- Phagocytosed by alveolar macrophages and multiply intracellularly
- After 3-4 weeks immune system attacks, forming **tubercles** (granulomas consisting of a central core containing bacilli surrounded by WBCs that undergoes caseation necrosis)

Secondary TB

- If patient doesn't recover from primary tuberculosis, reactivation of bacilli can occur.
- Tubercles expand and drain into the bronchial tubes and upper respiratory tract.
- Gradually the patient experiences more severe symptoms.
 - violent coughing, greenish or bloody sputum, fever, anorexia, weight loss, fatigue
- Untreated, 60% mortality rate

Extrapulmonary TB

- During secondary TB, bacilli disseminate to regional lymph nodes, kidneys, long bones, genital tract, brain, and meninges.

Clinical Findings

- Fatigue, weakness, weight loss, fever, and night sweats may be signs of tuberculous disease.
- Pulmonary involvement giving rise to chronic cough and spitting of blood usually is associated with far-advanced lesions.

Prevention & Control

Immunization: Various **living avirulent tubercle bacilli**, particularly BCG (**bacillus Calmette-Guérin, an attenuated bovine organism**), have been used to induce a certain amount of resistance in those heavily exposed to infection.

Management and Prevention of TB

- Treatment for 6-24 months with at least 2 drugs from a list of 11
- One pill regimen called *Rifater* (isoniazid, rifampin, pyrazinamide)
- Vaccine based on attenuated bacilli Calmet-Guerin strain of *M. bovis* used in other countries

Actinomycetes: Filamentous Bacilli

- Genera *Actinomyces* & *Nocardia* are nonmotile filamentous bacteria related to mycobacteria.
- May cause chronic infection of skin and soft tissues
- *Actinomyces israelii* – responsible for diseases of the oral cavity, thoracic or intestines - **actinomycoses**
- *Nocardia brasiliensis* causes pulmonary disease similar to TB.

