Chapter 19
The Gram-Positive Bacilli of Medical Importance
19.1 Medically Important Gram-Positive Bacilli

Can be subdivided into three general groups, based on presence or absence of endospores and acid-fastness

Three general groups:

1. Endospore-formers
2. Non-endospore-formers
3. Irregular shaped and staining properties
19.2 Spore-Forming Bacilli

Genus *Bacillus*

Genus *Clostridium*

Genus *Sporolactobacillus*
General Characteristics of the Genus *Bacillus*

- Gram-positive, endospore-forming, motile rods
- Mostly saprobic
- Aerobic and catalase positive
- Versatile in degrading complex macromolecules
- Source of antibiotics
- Primary habitat is soil
- 2 species of medical importance:
  - *Bacillus anthracis*
  - *Bacillus cereus*
Bacillus Anthracis

- Large, block-shaped rods
- Central spores that develop under all conditions except in the living body
- Virulence factors – polypeptide capsule and exotoxins
- 3 types of anthrax:
  - **Cutaneous** – spores enter through skin, black sore-eschar; least dangerous
  - **Pulmonary** – inhalation of spores
  - **Gastrointestinal** – ingested spores
Figure 19.2 Cutaneous anthrax
Control and Treatment

• Treated with penicillin, tetracycline, or ciprofloxacin

• Vaccines
  – Live spores and toxoid to protect livestock
  – Purified toxoid; for high risk occupations and military personnel; toxoid 6 inoculations over 1.5 years; annual boosters
Figure 19.1 (a) *Bacillus anthracis*
**Bacillus Cereus**

- Common airborne and dustborne; usual methods of disinfection and antisepsis 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
- No treatment
- Increasingly reported in immunosuppressed
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
Figure 19.1 (b and c)
Gas Gangrene

- *Clostridium perfringens* most frequent clostridia involved in soft tissue and wound infections – *myonecrosis*
- Spores found in soil, human skin, intestine, and vagina
- Predisposing factors – surgical incisions, compound fractures, diabetic ulcers, septic abortions, puncture wounds, gunshot wounds
Virulence Factors

- Virulence factors
  - Toxins
    - Alpha toxin – causes RBC rupture, edema, and tissue destruction
  - Collagenase
  - Hyaluronidase
  - DNase
Figure 19.3 Growth of *Clostridium perfringens*
Pathology

- Not highly invasive; requires damaged and dead tissue and anaerobic conditions
- Conditions stimulate spore germination, vegetative growth and release of exotoxins, and other virulence factors
- Fermentation of muscle carbohydrates results in the formation of gas and further destruction of tissue
Figure 19.4 Myonecrosis
Treatment and Prevention

- Immediate cleansing of dirty wounds, deep wounds, compound fractures, and infected incisions
- Debridement of disease tissue
- Large doses of cephalosporin or penicillin
- Hyperbaric oxygen therapy
- No vaccines available
**Clostridium Difficile-Associated Disease (CDAD)**

- Normal resident of colon, in low numbers
- Causes antibiotic-associated colitis
  - Relatively non-invasive; treatment with broad-spectrum antibiotics kills the other bacteria, allowing *C. difficile* to overgrow
- Produces enterotoxins that damage intestines
- Major cause of diarrhea in hospitals
- Increasingly more common in community-acquired diarrhea
Treatment and Prevention

- Mild uncomplicated cases respond to fluid and electrolyte replacement and withdrawal of antimicrobials
- Severe infections treated with oral vancomycin or metronidazole and replacement cultures
- Increased precautions to prevent spread
Figure 19.6 Antibiotic-associated colitis
Tetanus

- *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
Pathology

• Spores usually enter through accidental puncture wounds, burns, umbilical stumps, frostbite, and crushed body parts

• Anaerobic environment is required for vegetative cells to grow and release toxin

• Tetanospasmin – neurotoxin causes paralysis by binding to motor nerve endings; blocking the release of neurotransmitter for muscular contraction inhibition; muscles contract uncontrollably

• Death most often due to paralysis of respiratory muscles
Figure 19.7 The events in tetanus
Figure 19.8 Neonatal tetanus
Treatment and Prevention

• Treatment aimed at deterring degree of toxemia and infection and maintaining homeostasis
• Antitoxin therapy with human tetanus immune globulin; inactivates circulating toxin but does not counteract that which is already bound
• Control infection with penicillin or tetracycline; and muscle relaxants
• Vaccine available; booster needed every 10 years
Clostridial Food Poisoning

- *Clostridium botulinum* – rare but severe intoxication usually from home canned food
- *Clostridium perfringens* – mild intestinal illness; second most common form of food poisoning worldwide
Botulinum Food Poisoning

- Botulism – intoxication associated with inadequate food preservation
- *Clostridium botulinum* – spore-forming anaerobe; commonly inhabits soil and water
Pathogenesis

• Spores are present on food when gathered and processed
• If reliable temperature and pressure are not achieved air will be evacuated but spores will remain
• Anaerobic conditions favor spore germination and vegetative growth
• Potent toxin, **botulin**, is released
• Toxin is carried to neuromuscular junctions and blocks the release of acetylcholine, necessary for muscle contraction to occur
• Double or blurred vision, difficulty swallowing, neuromuscular symptoms
Figure 19.9 Physiological effects of botulism toxin
Infant and Wound Botulism

• Infant botulism – caused by ingested spores that germinate and release toxin; flaccid paralysis

• Wound botulism – spores enter wound and cause food poisoning symptoms
Treatment and Prevention

- Determine presence of toxin in food, intestinal contents or feces
- Administer antitoxin; cardiac and respiratory support
- Infectious botulism treated with penicillin
- Practice proper methods of preserving and handling canned foods; addition of preservatives
Clostridial Gastroenteritis

- *Clostrium perfringens*
- Spores contaminate food that has not been cooked thoroughly enough to destroy spores
- Spores germinate and multiply (especially if unrefrigerated)
- When consumed, toxin is produced in the intestine; acts on epithelial cells, acute abdominal pain, diarrhea, and nausea
- Rapid recovery
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<th>Motility</th>
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<td>Aerobe</td>
<td>−</td>
<td>Cutaneous anthrax, Pulmonary anthrax</td>
<td>Antibiotics, Vaccines for high risk</td>
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<td><em>Bacillus cereus</em></td>
<td>Facultative anaerobe</td>
<td>+</td>
<td>Food poisoning</td>
<td>None; disease self-limiting</td>
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<td><em>Clostridium perfringens</em></td>
<td>Strict anaerobe</td>
<td>−</td>
<td>Gas gangrene, Food poisoning (mild)</td>
<td>Debridement; antibiotics; oxygen therapy, None; disease self-limiting</td>
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<tr>
<td><em>Clostridium difficile</em></td>
<td>Strict anaerobe</td>
<td>+/-</td>
<td>Antibiotic-associated colitis</td>
<td>Withdrawal of antibiotics; administration of probiotics</td>
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<td><em>Clostridium tetani</em></td>
<td>Strict anaerobe</td>
<td>+</td>
<td>Tetanus</td>
<td>Vaccination; passive immunization</td>
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<td>Botulism</td>
<td>Antitoxin</td>
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19.3 Gram-Positive Regular Non-Spore-Forming Bacilli

Regular: stain uniformly and do not assume pleomorphic shapes

Medically important:

• *Listeria monocytogenes*
• *Erysipelothrix rhusiopathiae*
Listeria Monocytogenes

- Non-spore-forming gram-positive
- Ranging from coccobacilli to long filaments
- 1-4 flagella
- No capsules
- Resistant to cold, heat, salt, pH extremes, and bile
- Virulence attributed to ability to replicate in the cytoplasm of cells after inducing phagocytosis; avoids humoral immune system
Figure 19.10
Multiplication cycle of *Listeria monocytogenes*
Epidemiology and Pathology

- Primary reservoir is soil and water; animal intestines
- Can contaminate foods and grow during refrigeration
- **Listeriosis** – most cases associated with dairy products, poultry, and meat
- Often mild or subclinical in normal adults
- Immunocompromised patients, fetuses, and neonates; affects brain and meninges
  - 20% death rate
Diagnosis and Control

• Culture requires lengthy cold enrichment process
• Rapid diagnostic tests using ELISA, immunofluorescence, and DNA analysis
• Ampicillin and trimethoprim/sulfamethoxazole
• Prevention – pasteurization and cooking
Erysipelothrix Rhusiopathiae

- Gram-positive rod widely distributed in animals and the environment
- Primary reservoir – tonsils of healthy pigs
- Enters through skin abrasion, multiplies to produce erysipeloid, dark red lesions
- Penicillin or erythromycin
- Vaccine for pigs
Figure 19.11 Erysipeloid on hand
19.4 Gram-Positive Irregular Non-Spore-Forming Bacilli

Irregular: pleomorphic, stain unevenly

Medically important genera:

- Corynebacterium
- Propionibacterium
- Mycobacterium
- Actinomyces
- Nocardia
• 20 genera; *Corynebacterium, Mycobacterium,* and *Nocardia* greatest clinical significance

• All produce catalase, possess mycolic acids, and a unique type of peptidoglycan
Corynebacterium Diptheriae

- Gram-positive irregular bacilli
Epidemiology

- 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
Figure 19.13
Incidence and case fatality of diphtheria
Pathology

2 stages of disease:

1. Local infection – upper respiratory tract inflammation
   - Sore throat, nausea, vomiting, swollen lymph nodes; pseudomembrane formation can cause asphyxiation

2. Diptherotoxin production and toxemia
   - Target organs – primarily heart and nerves
Diagnostic Methods

• Pseudomembrane and swelling indicative
• Stains
• Conditions, history
• Serological assay
Figure 19.14
Diagnosing diphtheria
Treatment and Prevention

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

- *Propionibacterium acnes* most common
- Gram-positive rods
- Aerotolerant or anaerobic
- Nontoxigenic
- Common resident of pilosebaceous glands
- Causes acne
19.5 Mycobacteria: Acid-Fast Bacilli

- Gram-positive irregular bacilli
- Acid-fast staining
- Strict aerobes
- Produce catalase
- Possess mycolic acids and a unique type of peptidoglycan
- Do not form capsules, flagella, or spores
- Grow slowly
### TABLE 19.3 Differentiation of Important *Mycobacterium* Species

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<th>Primary Habitat</th>
<th>Disease in Humans</th>
<th>Treatment</th>
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<th>Pigmentation**</th>
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The mycobacteria are grouped into major categories by their growth rate and their pigment production.

*Growth rate is rapid (R), occurring in less than 7 days, or slow (S), occurring in more than 7 days.

**Photochromogens (PP) develop yellow to dark orange pigment in the presence of light; scotochromogens (PS) synthesize pigment in darkness; and nonpigmented forms (NP) have no color.
Figure 19.15 Microscopic morphology of mycobacteria
**Mycobacterium Tuberculosis**

- **Tubercle bacillus**
- Produces no exotoxins or enzymes that contribute to infectiousness
- Virulence factors – contain complex waxes and cord factor that prevent destruction by lysosomes or macrophages
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; highest rate in U.S. occurring in recent immigrants
- Bacillus very resistant; transmitted by airborne respiratory droplets
Course of Infection and Disease

- 5% to 10% of infected people develop clinical disease
- Untreated, the disease progresses slowly; majority of TB cases contained in lungs
- Clinical tuberculosis divided into:
  - Primary tuberculosis
  - Secondary tuberculosis (reactivation or reinfection)
  - Disseminated (extrapulmonary) tuberculosis
Figure 19.17 (a)
Staging of tuberculosis

- Exposure to TB droplets
  - Inhaled into lungs
    - No infection in 10–20% of cases
    - Lung macrophages engulf bacilli, infection occurs in 80–90% of cases; TB test +
    - Infection cleared by immune system in 90–95% of cases; TB test –
      - Latency; bacilli go dormant in lungs; carrier state without symptoms may last for many years; TB test +
      - Recurrent disease; tubercles break down; bacilli are released into lung cavities and circulation; TB test +
    - Primary TB disease with tubercles, symptoms occur in 5–10% of cases within 2 years; TB test +
  - Infection and disease can clear with long-term treatment; TB test + or –
  - Disease spreads to extrapulmonary sites such as kidney, bones, brain with severe morbidity; TB test +

(a) Infection  Disease  Infection free
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 – tubercle
• If center of tubercle breaks down into necrotic caseous lesions, they gradually heal by calcification
Figure 19.17 (b) Section of a tubercle

- Epithelioid cells
- Multinucleate giant cell
- Caseous necrosis (tubercle bacilli at center)
- Granuloma (fibroblast) cells
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

• These complications are grave
Diagnosis

1. *In vivo* or tuberculin testing

   **Mantoux test** – local intradermal injection of purified protein derivative (PPD); look for red wheal to form in 48-72 hours – **induration**; established guidelines to indicate interpretation of result based on size of wheal and specific population factors

2. X-rays

3. Direct identification of acid-fast bacilli in specimen

4. Cultural isolation and biochemical testing
Figure 19.18  
Skin testing for tuberculosis
Figure 19.19 X-ray of secondary tubercular infection
Figure 19.20 Fluorescent acid-fast stain of *Mycobacterium tuberculosis*
Management and Prevention of TB

• 6-24 months of 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
Mycobacterium Leprae: The Leprosy Bacillus

- Hansen’s bacillus/Hansen’s Disease
- Strict parasite – has not been grown on artificial media or tissue culture
- Slowest growing of all species
- Multiplies within host cells in large packets called globi
- Causes leprosy, a chronic disease that begins in the skin and mucous membranes and progresses into nerves
Epidemiology and Transmission of Leprosy

- Endemic regions throughout the world
- Mechanism of transmission is not fully verified
- Not highly virulent; appears that health and living conditions influence susceptibility and the course of the disease
- May be associated with specific genetic marker
Figure 19.21 Leprosy lesions
Course of Infection and Disease

• Macrophages phagocytize the bacilli, but a weakened macrophage or slow T cell response may not kill bacillus

• Incubation from 2-5 years; if untreated, bacilli grow slowly in the skin macrophages and Schwann cells of peripheral nerves

• 2 forms possible:
  – **Tuberculoid** – asymmetrical, shallow lesions, damage nerves – results in local loss of pain reception
  – **Lepromatous** – a deeply nodular infection that causes severe disfigurement of the face and extremities, widespread dissemination
Figure 19.22
Diagnosing

- Combination of symptomology, microscopic examination of lesions, and patient history
- Numbness in hands and feet, loss of heat and cold sensitivity, muscle weakness, thickened earlobes, chronic stuffy nose
- Detection of acid-fast bacilli in skin lesions, nasal discharges, and tissue samples
Figure 19.24 Feather test for leprosy
Treatment and Prevention

• Treatment by long-term combined therapy
• Prevention requires constant surveillance of high-risk populations
• WHO sponsoring a trial vaccine
Infections by Non-Tuberculosis Mycobacteria (NTM)

• *M. avium* complex – third most common cause of death in AIDS patients
• *M. kansaii* – pulmonary infections in adult white males with emphysema or bronchitis
• *M. marinum* – water inhabitant; lesions develop after scraping on swimming pool concrete
• *M. scrofulaceum* – infects cervical lymph nodes
• *M. paratuberculosis* – raw cow’s milk; recovered from 65% of individuals diagnosed with Crohn’s disease
Figure 19.25 Chronic swimming pool granuloma
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19.6 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
Figure 19.26 Symptoms and signs of actinomycosis
Figure 19.27 Nocardiosis