Pathogenic Gram-Positive Cocci and Bacilli

http://www.healthmap.org/en

The Three Domains of Life

Domain characteristics
What is GC ratio?

- The proportion of a cell’s DNA that is guanine and cytosine
- Used for taxonomy
  
  \[
  \frac{G + C}{A + T + C + G} \times 100\%
  \]
- GC ratios vary from 20% to 80% among prokaryotes
Summary of Selected Gram-Positive Bacteria

Table 11.2.1

<table>
<thead>
<tr>
<th>Characteristics of Selected Gram-Positive Bacteria</th>
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<td>Positive Bacteria</td>
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Staphylococcus

- Normal members of every human’s microbiota
- Can be opportunistic pathogens

Structure and Physiology

- Gram-positive cocci, nonmotile, facultative anaerobes
- Cells occur in grapelike clusters
- Salt-tolerant – allows them to tolerate the salt present on human skin
- Tolerant of desiccation
- Allows survival on environmental surfaces (fomites) biofilms on catheters

Structure and Physiology

- Two species are commonly associated with staphylococcal diseases in humans
  - *Staphylococcus aureus* - the more virulent strain that can produce a variety of diseases depending on the site of infection
  - *Staphylococcus epidermidis* - normal microbiota of human skin that can cause opportunistic infections in immunocompromised patients or when introduced into the body (i.e. artificial heart valves)
Pathogenicity
- "Staph" infections result when staphylococci breach the body's physical barriers
- Entry of only a few hundred bacteria can result in disease
- Pathogenicity results from three features
  1. Structures that enable it to evade phagocytosis
  2. Production of enzymes
  3. Production of toxins

Structural Defenses Against Phagocytosis
- **Protein A** coats the cell surface (Sa)
  - Interferes with humoral immune responses by binding to class G antibodies (on Fc regions)
- Bound **coagulase** (Sa)
  - Converts the soluble blood protein fibrinogen in insoluble fibrin molecules that form blood clots
  - Fibrin clots hide the bacteria from phagocytic cells

Structural Defenses Against Phagocytosis
- Synthesize loosely organized polysaccharide slime layers (Sa and Se) (often called capsules)
- Inhibit chemotaxis of and phagocytosis by leukocytes
- Facilitates attachment of Staphylococcus to artificial surfaces
Enzymes: see chapter 14

- **Cell-free coagulase** *(Sa = Staphylococcus aureus)*
  - Triggers blood clotting
- **Hyaluronidase** *(Sa)*
  - Breaks down hyaluronic acid, enabling the bacteria to spread between cells
- **Staphylokinase** *(Sa)*
  - Dissolves fibrin threads in blood clots, allowing *S. aureus* to free itself from clots

- **Lipases**
  - Digest lipids, allowing staphylococcus to grow on the skin’s surface and in cutaneous oil glands
- **β-lactamase**
  - **Breaks down penicillin**
  - Allows the bacteria to survive treatment with β-lactam antimicrobial drugs
Toxins

- *Staphylococcus aureus* produces toxins more frequently than *S. epidermidis*
- **Cytolytic toxins**
  - Disrupts the cytoplasmic membrane of a variety of cells
  - **Leukocidin** can lyse leukocytes specifically
- **Exfoliative toxins**
  - Causes the patient’s skin cells to separate from each other and slough off the body

**Table 19.1**

<table>
<thead>
<tr>
<th>Factor</th>
<th><em>S. aureus</em></th>
<th><em>S. epidermidis</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein A</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Coagulase</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Catalase</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Hyaluronidase</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Staphylocinase</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Lipase</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>β-lactamase (penicillinase)</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

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Staphylococcal Diseases:
Three categories

1. Noninvasive disease:
   - Food poisoning from
   - the ingestion of
   - enterotoxin-contaminated
   - food
   - Intoxication

2. Cutaneous disease
   - Various skin conditions including scalded skin syndrome, impetigo, folliculitis, and furuncles

Impetigo: small, flattened, red patches forming pus
More cutaneous diseases

- Folliculitis (i.e. sty)
- Furuncle (boil)
- Carbunkle

3. Systemic Staphylococcal Diseases/symptoms

- **Toxic shock syndrome** - TSS toxin is absorbed into the blood and causes shock
- **Bacteremia** - presence of bacteria in the blood
- **Endocarditis** - occurs when bacteria attack the lining of the heart (50% mortality)
- **Pneumonia** - inflammation of the lungs in which the alveoli and bronchioles become filled with fluid
- **Osteomyelitis** - inflammation of the bone marrow and the surrounding bone
Diagnosis, Treatment

**Diagnosis**
- Detection of Gram-positive bacteria in grapelike arrangements isolated from pus, blood, or other fluids

**Treatment**
- Methicillin is the drug of choice to treat staphylococcal infections
  - Is a semisynthetic form of penicillin and is not inactivated by β-lactamase

**MRSA**

MRSA: methicillin-resistant Staphylococcus aureus (MRSA) has become the most frequent identifiable cause of skin and soft-tissue infections among patients treated at hospital emergency departments in several metropolitan areas across the US, investigators report.  S. aureus was isolated from 322 (76%) of the patients. Of these, 78% were MRSA. Thus, MRSA was isolated from 59% of the patients. Methicillin-sensitive S. aureus (MSSA) was the second most common cause, affecting 17% of patients. Streptococcal infections made up most of the remaining bacterial isolates. No organism was isolated from 9% of patients.

From Medscape online
Prevention
- Hand antisepsis is the most important measure in preventing nosocomial infections.
- Also important is the proper cleansing of wounds and surgical openings, aseptic use of catheters or indwelling needles, an appropriate use of antiseptics.

Streptococcus
- Gram-positive cocci, arranged in pairs or chains, that are facultative anaerobes.

Streptococcus: Serotype classification
- Often categorized based on the Lancefield classification.
  - Divides the streptococci into serotype groups based on the bacteria’s surface antigens.
  - Lancefield groups A and B include the significant streptococcal pathogens of humans.
S. pyogenes forms white colonies surrounded by zone of beta-hemolysis on blood agar plates.

Group A Streptococcus: 
*Streptococcus pyogenes*
  
**Beta hemolytic**
  
- Pathogenic strains often form a capsule
- Group A streptococci generally only cause disease in certain situations
  - 1. Normal microbiota are depleted
  - 2. Large inoculum enable the streptococci to establish themselves before antibodies are formed against them
  - 3. Specific immunity is impaired

**Pathogenicity**
  
- Structural components
  - Protein M, which interferes with opsonization and lysis of the bacteria (via C')
  - Hyaluronic acid capsule, which acts to camouflage the bacteria
- Enzymes
  - Streptokinases, deoxynucleases (much DNA in pus), and C5a peptidase all facilitate the spread of streptococci through tissues
- Pyrogenic (erythrogenic) toxins that stimulate macrophages and helper T cells to release cytokines, causing fever (shock) in lysogenized strep
- Streptolysins lye red blood cells, white blood cells, and platelets
Group A Streptococcal Diseases

- **Pharyngitis ("strep throat")** - inflammation of the pharynx
- **Scarlet fever** - rash that begins on the chest and spreads across the body
- **Pyoderma (erysipelas)** - confined, pus-producing lesion that usually occurs on the face, arms, or legs
- **Streptococcal toxic shock syndrome** - bacteremia and severe multisystem infections
Erysipelas: spreads to lymph nodes

Necrotizing Fasciitis; flesh-eating Strep

More Group A Streptococcal Diseases
- **Necrotizing fasciitis** - toxin production destroys tissues and eventually muscle and fat tissue
- **Rheumatic fever** - inflammation that leads to damage of heart valves muscle (autoimmune)
- **Glomerulonephritis** - inflammation of the glomeruli and nephrons which obstruct blood flow through the kidneys
Diagnosis

- Observation of Gram-positive bacteria in short chains or pairs or immunological tests that identify the presence of group A streptococcal antigens
- Streptococci are normally in the pharynx so their presence in a respiratory sample is of little diagnostic value

Treatment and Prevention

- Treatment
  - Penicillin is very effective
  - Also susceptible to bacitracin
- Prevention
  - Antibodies against M protein provide long-term protection against future infection of *S. pyogenes*, but only if it is the same strain

Group B Streptococcus: *Streptococcus agalactiae*

- Gram-positive cocci that divide to form chains
- Distinguished from group A streptococcus by its buttery colonies and smaller zone of beta-hemolysis on blood agar plates and its resistance to bacitracin
- **Normally colonizes the lower gastrointestinal, genital, and urinary tracts**
Pathogenicity and Diseases

**Pathogenicity**
- Often infects newborns who have not yet formed type-specific antibodies and whose mothers are uninfected (and so do not provide passive immunity).
- Produces various enzymes whose roles in causing disease is not yet understood.

**Diseases**
- Most often associated with neonatal bacteremia, meningitis, and pneumonia.
- Immunocompromised older patients are at risk from group B streptococcal infections.

Diseases

Diagnosis, Treatment,

**Diagnosis**
- **Agglutination and ELISA** test utilizing antibodies directed against the bacteria's distinctive cell wall polysaccharide.
- Bacitracin **does not** inhibit its growth.

**Treatment**
- Penicillin G, sometimes in combination with an aminoglycoside.

Prevention

**Prevention**
- Prophylactic administration of penicillin at birth to children whose mothers' urinary tracts are colonized with group B streptococci.
- Immunization of women against group streptococci to prevent infection of future children.

Alpha-Hemolytic Streptococci: The Viridans Group
- Lack group-specific carbohydrates and cannot be grouped by the Lancefield system
- Many produce a green pigment when grown on blood media
- Normally inhabit the mouth, pharynx, GI tract, genital tract, and urinary tract
- One of the causes of dental caries and dental plaques
- If enter the blood can cause meningitis and endocarditis

Streptococcus pneumoniae
- Gram-positive cocci that most commonly forms pairs but may also form chains
- Forms unpigmented, alpha-hemolytic colonies when grown on blood agar (anaerobic incubation produces beta-hemolytic colonies)
- Normally colonizes the mouths and pharynx but can cause disease if travels to the lungs
- Disease is highest in children and the elderly
Summary of streptococci

### Pathogenicity

- **Phosphorylcholine** – stimulates host cells to phagocytize the bacteria
- **Polysaccharide capsule** – protects the bacteria from digestion after endocytosis
- **Protein adhesin** – mediates binding of the cells to epithelial cells of the pharynx
- **Secretory IgA protease** – destroys IgA
- **Pneumolysin** – lyses epithelial cells and suppresses the digestion of the endocytized bacteria

### Diseases

- **Pneumococcal pneumonia** – bacteria multiply in the alveoli of the lower lung causing damage to the alveolar lining and producing an inflammatory response
- **Bacteremia and endocarditis** – bacteria in the bloodstream or in the lining of the heart
- **Pneumococcal meningitis** – bacteria that have spread to the meninges
capsules

Otitis media

- Sinusitis and otitis media – bacteria invade the sinuses or middle ear, often following a viral infection

Diagnosis, Treatment, and Prevention

- Diagnosis
  - Gram-strain of sputum smears
  - Quellung reaction – anti-capsular antibodies cause the capsule to swell, confirming the presence of bacteria

- Treatment
  - Penicillin

- Prevention
  - Vaccine made from purified capsular material
  - Newer PCV conjugate is more effective for children

http://www.vaccineinformation.org/pneumchild/qandavax.asp
**Enterococcus**
- Previously classified as group D streptococci but differed enough to be reclassified as a separate genus
- Form short chains and pairs and lack a capsule
- Found in the human colon but are rarely pathogenic at this site
- Can cause disease if they are introduced into other parts of the body, such as the urinary tract or bloodstream

**Enterococcus**
- Important cause of nosocomial infections
- Treatment is difficult because enterococci are often resistant to antimicrobials: VRE (Vancomycin Resistant Enterococcus)
- Prevention is difficult, especially in a healthcare setting, where patients' often have weakened immune systems

**Bacillus (genus)**
- Gram-positive bacilli, that occurs singly, in pairs, or in chains
- Forms endospores
- *Bacillus anthracis* is a strict pathogen of animals and humans
  - Primarily a disease of herbivores, but humans can contract the disease from infected animals
  - Humans contract the bacteria via one of three routes:
    - Inhalation of spores
    - Inoculation of spores into the body through a break in the skin
    - Ingestion of spores
Pathogenicity and Diseases

Pathogenicity
- Anthrax toxin

Diseases
- Anthrax is the only disease caused by *Bacillus anthracis*
- Anthrax can have three clinical manifestations
  - Gastrointestinal anthrax
    - Rare in humans
    - Intestinal hemorrhaging and eventually death
  - Cutaneous anthrax
    - Produces a ulcer called an eschar and toxemia
Inhalation Anthrax

- Inhalation anthrax
- Rare in humans
- Spores germinate in the lungs and secrete toxins that are absorbed into the bloodstream
- High mortality rate
- Bioterrorism in US mail

Diagnosis, Treatment, and Prevention

- **Diagnosis**
  - Presence of large, nonmotile, Gram-positive bacilli in clinical samples of the lungs or skin
- **Treatment**
  - Ciproflaxacin and many other antimicrobials are effective against *B. anthracis*
- **Prevention**
  - Control the disease in animals
  - An anthrax vaccine is available but requires multiple doses and boosters

Clostridium: description

- **Gram-positive, anaerobic, endospore-forming bacillus**
- Ubiquitous in soil, water, and the gastrointestinal tracts of animals and humans
- The presence of endospores allows for survival in harsh conditions
**Clostridium perfringens**
- Commonly grows in the digestive tracts of animals and humans
- Produces 11 toxins that have various effects on the body and can result in irreversible damage

**Diseases**
- **Food poisoning**
  - Benign disease characterized by abdominal cramps and watery diarrhea
- **Gas gangrene** (11 toxins produced)
  - Endospores are introduced into the body through some traumatic event
  - The endospores germinate and cause necrosis that is often accompanied by foul-smelling gaseous bacterial waste products
**Diagnosis, Treatment**

- **Diagnosis**
  - The presence of more than $10^5$ bacteria in a gram of food or $10^6$ cells per gram of feces indicates the involvement of *Clostridium* in food poisoning
  - Gas gangrene is usually diagnostic by itself

- **Treatment**
  - Food poisoning is self-limited
  - Gas gangrene is treated by removing the dead tissue and administering large doses of antitoxin and penicillin

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**Prevention**

- Difficult to prevent because it is so common
- Proper cleaning of wounds can often prevent gas gangrene

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**Clostridium difficile: Cdiff**

- Common member of the intestinal microbiota
- Opportunistic pathogen in patients treated with broad-spectrum antimicrobial drugs
  - Minor infections can result in a self-limiting explosive diarrhea
  - Serious cases can cause pseudomembranous colitis
  - Can result in perforation of the colon, leading to massive internal infection by fecal bacteria and eventual death
**Clostridium difficile**
- Diagnosed by isolating the organism from feces or by demonstrating the presence of toxins via immunoassay
- Minor infections are usually resolved by discontinuing use of the antimicrobial drug in use
- Serious cases are treated with antibiotics
- Proper hygiene is critical for limiting nosocomial infections

**Clostridium botulinum**
- **Anaerobic, endospore-forming, Gram-positive bacillus**
- Common in soil and water
- Botulism results when the endospores germinate and produce botulism toxin
- The different botulism toxins (A through G) are among the deadliest toxins known

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**Figure 19.14**
Diseases

- Botulism is not an infection, but an intoxication caused by the botulism toxin.

Three forms of botulism

- Food-borne botulism
  - Usually occurs due to the consumption of toxin in home-canned foods or preserved fish.
  - Can result in a progressive paralysis that results in death due to the inability to inhale.

- Infant botulism
  - Results from the ingestion of endospores, which germinate and colonize the infant's gastrointestinal tract due to the lack of sufficient numbers of normal microbiota.
  - Symptoms include constipation and "failure to thrive"; paralysis and death are rare.

- Wound botulism
  - Wound becomes contaminated with endospores.
  - Symptoms are the same as with food-borne botulism.

Diagnosis, Treatment

Diagnosis
- Symptoms of botulism are diagnostic.
- Confirm diagnosis by culturing the organism from food, feces, or the patient's wound.

Treatment
- Can involve three approaches:
  - Repeated washing of the intestinal tract to remove Clostridium.
  - Administer antibodies against botulism toxin to neutralize toxin in the blood before it can bind to neurons.
  - Administer antimicrobials drugs to kill clostridia in infant botulism cases.
Prevention
- Proper canning of food to prevent contamination
- Infants should not consume honey under the age of 1

Clostridium botulinum
- Gram-negative, spore-forming
- Ubiquitous in soil, dust, and the GI tract of animals and humans
- Tetanus results when the bacterial endospores germinate and produce tetanus toxin

Clostridium tetani
- Endospore-forming, obligately anaerobic, Gram-positive bacilli
- Ubiquitous in soil, dust, and the GI tract of animals and humans
- Tetanus results when the bacterial endospores germinate and produce tetanus toxin
- Tetanus results in spasms and contractions that can result in death because patients can’t exhale
**Diagnosis, Treatment,**

- **Diagnosis**
  - Characteristic muscular contraction
  - The bacteria is rarely isolated from clinical samples because it grows slowly and is sensitive to oxygen

- **Treatment**
  - Thorough cleaning of wounds to remove endospores
  - Passive immunization with immunoglobulin (antitoxin) directed against the toxin (exotoxin)
  - Administration of antimicrobials
  - Active immunization with tetanus toxoid
Prevention

- Immunization with tetanus toxoid

Listeria

- Gram-positive non-spore-forming, coccobacillus
- Found in soil, water, mammals, birds, fish, and insects
- Enters body in contaminated food and drink
- Psychrophilic: grows in refrigerator
- Listeria produces no toxins or enzymes
- Virulence is directly related to the bacteria’s ability to live within cells
- Pregnant woman to fetus
- Leads to premature delivery or meningitis
Diagnosis, Treatment, and Prevention

- **Diagnosis**
  - Presence of the bacteria in the cerebrospinal fluid
  - Rarely seen by Gram-staining because so few *Listeria* cells are required to produce disease

- **Treatment**
  - Most antimicrobial drugs inhibit *Listeria*

- **Prevention**
  - Difficult because the organism is ubiquitous
  - At risk individuals should avoid undercooked vegetables, unpasteurized milk, undercooked meat, and all soft cheeses

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*Corynebacterium*

- **G+, pleomorphic, nonendospore-forming**
- Ubiquitous on plants and in animals and humans
- Colonizes the skin and the respiratory, gastrointestinal, urinary, and genital tract

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**Disease**

- *Corynebacterium diphtheriae*, the cause of diphtheria, is the most widely known
  - Transmitted from person to person via respiratory droplets or skin contact
  - Endemic in poor parts of the world that lack adequate immunization
  - Diphtheria toxin is responsible for the signs and symptoms of diphtheria
  - Diphtheria toxin inhibits polypeptide synthesis which results in cell death
  - Infections are asymptomatic or produce mild respiratory disease in immune or partially immune individuals
Disease

- Severe respiratory infections of nonimmune patients produce the signs and symptoms of diphtheria.
  - Pseudomembrane results from fluid that has thickened and adheres throughout the respiratory tract.
  - The pseudomembrane can completely occlude the respiratory passages and cause suffocation.
- Cutaneous diphtheria causes cell death and formation of a pseudomembrane on the skin.

Diagnosis, Treatment, and Prevention

- **Diagnosis**
  - Initial diagnosis is based on the presence of pseudomembrane.
  - Absolute identification is based on the Elek test.
    - Antibodies against the toxin react with toxin in a sample of fluid from the patient.
- **Treatment**
  - Administration of antitoxin to neutralize toxin before it binds to cells.
  - Penicillin and erythromycin kills the bacteria.
- **Prevention**
  - Immunization with the DPT vaccine.

*Mycobacterium* characteristics

- High G+C nonendospore-forming pathogen.
- Cell wall contains large amounts of a waxy lipid called mycolic acid.
- The unusual cell wall results in a number of unique characteristics.
  - Airborne pathogen resists dessication.
  - Slow growth.
  - Protection from lysis once the bacteria are phagocytized.
  - Capacity for intracellular growth.
  - Resistance to Gram-staining, detergents, many antimicrobial drugs, and dessication.
Acid-fast

Figure 19.21 - Overview

Mycobacterium: the Three main mycobacterial diseases
- Tuberculosis
- Leprosy
- Opportunistic infections in AIDS patients

Tuberculosis (TB)
- Respiratory disease cause by *Mycobacterium tuberculosis*
- Cases are declining in the United States but it is pandemic in other parts of the world
- Virulent strains of *M. tuberculosis* contain the cell wall component, cord factor, that is necessary to cause disease
Tuberculosis (TB)

- Three types of tuberculosis
  - Primary TB
    - Results from the initial infection with M. tuberculosis
  - Secondary TB
    - Reestablishment of an active infection after a period of dormancy
  - Disseminated TB
    - Results when the infection spreads throughout the body
Diagnosis

- Tuberculin skin test identifies individuals with previous exposure to M. tuberculosis by the presence of a hard, red swelling at the test site.
- Chest x-rays are used to identify individuals with active disease.

Treatment and Prevention

- Treatment: Treatment with common antimicrobials is difficult because the bacteria grow slowly and can live within macrophages. Combination therapy must be used for a number of months to treat the disease.
- Prevention: Prophylactic use of antibacterial drugs is used to treat patients who have shown a conversion from a negative to a positive skin test or were exposed to active cases of tuberculosis. Immunization with BCG vaccine is used in countries where TB is common.

Case distribution

- Inactive contagious cases reported in the U.S.A.
- Percentage of cases by race or ethnic origin in the U.S.A. (2009)
Leprosy

- Caused by *Mycobacterium leprae*
- Bacteria have never been grown in cell-free culture (grow on armadillos)
- Cases of leprosy are becoming relatively rare
- Transmission is via person-to-person contact or through a break in the skin

Leprosy

- Two different forms of disease
  - Tuberculoid leprosy: Nonprogressive disease that is characterized by loss of sensation in regions of the skin
  - Lepromatous leprosy: Produces gradual tissue destruction that results in the loss of facial features, digits, and other body structures

Diagnosis, Treatment

- Diagnosis
  - Based on the signs and symptoms of the disease
  - Loss of sensation in skin lesions in the case of tuberculoid leprosy
  - Disfigurement in the case of lepromatous leprosy
- Treatment
  - Treatment with a combination of antimicrobial drugs
  - Lifelong treatment is sometimes needed
Prevention

- Primarily prevented by limiting exposure to the pathogen
- BCG vaccine provides some protection

Mycobacterial Infections in AIDS Patients

- Mycobacterium avium-intracellularare is the most common mycobacterial infection among AIDS patients in the United States
- Infections are a result of ingestion of contaminated food or water
- Infections can simultaneously affect almost every organ and result in massive organ failure
- Treatment is difficult due to the disseminated nature of the infection

Propionibacterium

- Small, Gram-positive rods that are often found on the skin
- Propionibacterium acnes is the species most commonly involved in human infections
- Causes much of the acne of adolescents and young adults
- May also be an opportunistic pathogen
- Treatment often involves the use of antimicrobial drugs though many cases require no treatment
**Nocardia asteroides**

- Common inhabitant of soils rich in organic matter
- Produces opportunistic infections in numerous sites
  - Pulmonary infections
    - Develop from inhalation of the bacteria
    - Produce pneumonia
  - Cutaneous infections
    - Result from introduction of the bacteria into wounds
    - Produce mycetoma, a painless, long-lasting infection characterized by swelling, pus production, and draining sores

**Central nervous system infections**
- Result from the spread of the bacteria in the blood

**Prevention of nocardial disease**
- Involves avoiding exposure to the bacterium in soil
**Actinomyces**

- Normal member of the surface microbiota of human mucous membranes
- Produces opportunistic infections of the respiratory, gastrointestinal, urinary, and female genital tracts
- Actinomycosis results when the bacteria enters breaks in the mucous membrane
- Disease is characterized by the formation of many abscesses connected by channels in the skin or mucous membranes
- Diagnosis of actinomycosis can be difficult because other organisms cause similar diseases

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**Actinomycoses**

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