MHD Streptococcus and Enterococcus

STREPTOCOCCI AND ENTEROCOCCI

• Reading Assignment: Murray et al., Medical Microbiology, 8th ed., 2016. Chapter 19

Streptococci and Enterococci

OBJECTIVES

• Name the species of streptococci associated with Lancefield groups A and B
• Describe the structural and functional characteristics that distinguish streptococci from staphylococci
• List clinical manifestations of Streptococcus pyogenes infection.
• Name two diseases that occur as sequelae to streptococcal infections and describe their clinical presentation
• Name virulence factors associated with Streptococcus pyogenes and the mechanisms by which they enhance infection
OBJECTIVES
• Describe the limitations of Rapid Group A Detection kits
• Explain the role of S. agalactiae in causing neonatal disease and how neonatal disease may be prevented.
• Name the species included in the Anginosus Group Strep
• Identify the key morphologic and phenotypic tests used for identification of S. pneumoniae
• List the two major species of Enterococcus causing infection in humans and explain their association with vancomycin resistance.
• List 3 most common infections caused by enterococci

After working all day on the docks, I
Most carefully take off my socks. I
Will make an inspection
To look for infection
With Strep, or with Staph
—they're both cocci.

Adapted from a poem by Sheila B on OEDILF
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Staphylococci are **catalase +**
Streptococci/Enterococci are **catalase -**
**Classification of Streptococci**

- Hemolytic properties:
  - Complete hemolysis (Beta)
  - Incomplete hemolysis (Alpha)
  - No hemolysis (Gamma)
- Agar with sheep’s blood

**Lancefield Typing**

- Antigenic differences in group-specific polysaccharides located in the bacterial cell wall
  - Group specific carbohydrate
  - A, B, C, F, G most commonly used
  - A-H, K-U
- Primarily for beta Strep, but some alpha Strep/Enterococcus will type by group
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Lancefield Typing by Latex Agglutination

Streptococci
Important Species
- *Streptococcus pyogenes* (Group A)
- *Streptococcus agalactiae* (Group B)
- Other beta hemolytic streptococci
- Viridans group streptococci
- *Streptococcus pneumoniae*

Case 1
- A 15 year old girl presents to an urgent care clinic
- She describes sore throat, pain on swallowing and she has a temperature of 38.3°C
- She as a negative rapid antigen test for “strep throat”
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Case 1 Diagnosis
Acute pharyngitis caused by *Streptococcus pyogenes*
- Fever, sore throat, headache, cervical lymphadenopathy (swollen lymph nodes), exudate
- Difficult to differentiate from viral infection
- 5% asymptomatic carriers
- Transmitted by respiratory droplets
- Rapid antigen testing is not 100% sensitive

*Streptococcus pyogenes* (GAS)
Key Characteristics
- Gram positive cocci in chains
- Beta-hemolytic colonies

Streptococci
Habitat
- Found in mucous membranes
  - Nasopharynx
  - Throat
  - Vagina
  - Gut
- Significant opportunistic pathogen under appropriate conditions
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What’s in the Bug Tool Box?

- Adhere
- Evade
- Destroy

What’s in the Bug Tool Box?

- Adhere = M Protein, F Protein
- Evade = M Protein, capsule
- Destroy = Exotoxins, Enzymes

Streptococcus pyogenes

Virulence Factors

- M protein (>80 serotypes)
  - Binds to epidermal cells
  - allows bacteria to survive
  - Strains without M protein are avirulent
  - Antiphagocytic
- Degrades complement C3b
- Antibodies to M protein activate complement and kill the bacteria
**S. pyogenes**

**Infections**

- **Erysipelas**
  - Spreading erythema with well-demarcated edge on the face
  - Fever and lymphadenopathy
  - Lesions often on face and often with accompanying streptococcal pharyngitis

- **Impetigo**
  - Pyoderma
  - Young children, warm months, poor hygiene
  - Associated with trauma / insect bites
  - Pustule with yellow crust
  - Appears on face or extremities
  - Not the same strains as those that cause pharyngitis
**Streptococcus pyogenes**

**Virulence Factors**
- Enzymes (Destroy) — The “A ses”
- Streptokinase
  - Cleavage of fibrinogen and fibrin
  - “clot buster”
  - Facilitates spread in infected tissues

**Streptococcus pyogenes**

**Virulence Factors**
- Enzymes (Destroy)
- Hemolysins — The “Lysin” Family
  - Lyse RBCs, leukocytes
  - Death of phagocytic cell
  - Spread of bacteria in tissues
  - Streptolysin S:
  - Streptolysin O:

**Case 2**
- An 8 year old boy presents to his pediatrician
- He has a low-grade fever and a diffuse erythematous rash over his chest, that developed 2 days after he complained of a painful sore throat
- An exudate was present over the tonsillar area of this throat and covered his tongue
- He has a positive rapid antigen test
Case 2 Diagnosis

Scarlet Fever
- Complication of streptococcal pharyngitis
- A rash first appears as tiny red bumps on the chest and abdomen
- Fine, red, and rough-textured blanches upon pressure
- Appears 12–48 hours after fever

Scarlet Fever
- Generally starts on the chest, armpits, and behind the ears
- Accentuation of the rash in skin folds (Pastia’s lines) and circumoral pallor

Scarlet Fever
Characterized by:
- Sore throat
- Fever
- Bright red tongue with a "strawberry" appearance
- The rash begins to fade three to four days after onset and desquamation begins.
Case 3

- A 35 year old man presents to the emergency department
- He reports 3 days of malaise, diffuse myalgia (muscle ache), and low grade fever
- Over the next several hours the pain became excruciating
- Extensive necrotizing fasciitis was present on surgical exploration
Case 3 Diagnosis

Necrotizing Fasciitis cause by *S. pyogenes*

- Strep infection that occurs deep in the subcutaneous tissues
- Spreads along the fascial planes
- Extensive destruction of the muscle and fat
- “Flesh eating” bacteria
- Systemic toxicity, mortality exceeds 50%

*S. pyogenes* Infections

- Toxic Shock-like Syndrome
  - Multisystem organ failure (heart, respiratory tract, kidney)
  - SPE toxins are similar to Staph aureus TSST-1
  - Unlike patients with staph toxic shock, cultures are usually positive for Group A Strep
**Streptococcus pyogenes**

**Virulence Factors**

- Streptococcal pyrogenic exotoxins (SPE)
  - Three distinct heat labile toxins (A, B, C)
  - Called “superantigens” that stimulate cytokine response leading to shock and organ failure
  - Strep toxic shock-like syndrome
  - Responsible for the rash in scarlet fever “erythrogenic exotoxin”

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**Post-Streptococcal Sequelae**

- Rheumatic Fever
  - Non-suppurative inflammatory disease occurs 1-5 weeks after strep pharyngitis
  - Inflammation in the heart, joints, skin or central nervous system
  - Fever, carditis, subcutaneous nodules, chorea, polyarthritis
  - Attacks reoccur into adulthood
  - Characteristic cardiac lesions = Aschoff bodies and valvular damage leads to possible endocarditis later in life

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**Post-Streptococcal Sequelae**

- Acute Glomerulonephritis
  - Edema, hypertension, hematuria, proteinuria
  - Occurs after skin / respiratory infection
  - Certain M types are “nephritogenic”
  - Antigen + antibody + C’ deposited in glomeruli seen on kidney biopsy
Case 4

- An infant male was delivered spontaneously at term
- Examinations were normal during the first week of life
- On day 13 the baby was admitted to the hospital with generalized seizures
- Cerebral spinal fluid was collected by lumbar puncture and a Gram positive coccus was isolated by culture in lab

Case 4 Diagnosis

Late-Onset Group B Strep Neonatal Disease

- Early Onset neonatal disease
  - First week of life
  - Bacteremia, pneumonia, or meningitis
- Late onset neonatal disease
  - 1 week to 3 months of age
  - Bacteremia with meningitis

Streptococcus agalactiae (GBS)

Key Characteristics

- Beta-hemolytic colonies
- Gram positive cocci in chains
Streptococcus agalactiae

• Part of normal flora in throat, vaginal and GI tract
• Infections in Pregnant Women
• Infections in Men and Non-Pregnant Women
  – Neonatal pneumonia, sepsis, meningitis
  – Skin and wound infections in adult diabetic patients
  – Endocarditis

Streptococcus agalactiae

Virulence Factors

• Maternal colonization of vagina or rectum exposes baby at delivery
• Lack of protective maternal antibody
• Sialic acid on polysaccharide capsule inhibits C allowing organisms to multiply

Group B Strep Infection Prevention

• To prevent perinatal GBS disease, cultures should be performed on vaginal/rectal swabs collected at 35 to 37 weeks gestation
• Lab Plays Important Role
  – Combined vaginal/rectal swab improves isolation rates by 40% over vaginal swab alone. Alert physician when cervical or vaginal specimens received
  – Use of selective enrichment broth (LIM) will increase GBS isolation by 50%; slows down time to result
Other Beta Hemolytic Strep

*Streptococcus dysgalactiae*
- Group C
  - Associated with veterinary infections
  - Pharyngitis in college age patients
  - Sepsis
- Group G
  - Pharyngitis
  - Sepsis in neonates and elderly

Other Beta Hemolytic Strep

*Anginosus group Streptococci*
- Group F
  - Associated with abscesses
Case 5

- A 68 year old female presents to the emergency department
- She developed fever, chills, weakness, and a productive cough with pleuritic chest pain 3 days prior to admission
- At the time of admission she was febrile and in moderate respiratory distress
- Chest radiograph showed infiltrates with pleural effusions

Case 5 Diagnosis

Pneumonia caused by *Streptococcus pneumoniae*

- Normal flora of human upper respiratory tract
- After aspiration, bacteria multiply in alveolar spaces
- Lower lobes = lobar pneumonia
- Onset is abrupt
- Patients usually have productive cough and chest pain (pleurisy)

*Streptococcus pneumoniae*

**Key Characteristics**

- Gram positive diplococci (pairs)
- Alpha-hemolytic colonies
- Optochin S
**Streptococcus pneumoniae**

**Epidemiology**
- Most common cause of community acquired acute bacterial pneumonia
  - Estimated 500,000 cases annually
- Predisposing conditions
  - Alcoholism
  - Diabetes mellitus
  - Chronic lung disease
  - Chronic renal disease
  - Certain malignancies

**Streptococcus pneumoniae**

**Virulence Factors**
- More than 90 known serotypes
- Polysaccharide capsule is an important virulence factor
- Capsular type-specific antibody is protective = vaccine target
- Prevents phagocytosis
- Pneumolysin – damages ciliated cells and activates alternative complement pathway

**S. pneumoniae Capsule**
**Pneumococcal Disease**

- Bacteremia most common clinical presentation among children younger than 2 years
- Most common cause of bacterial meningitis in the U.S.
- Highest rate of meningitis among children younger than 2 years
- Paranasal sinusitis and otitis media
- Meningitis, which is usually secondary to one of the former infections
- Osteomyelitis, septic arthritis, endocarditis, peritonitis, cellulitis and brain abscesses.
- Prevention through vaccination (capsular serotypes)

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**Viridans Streptococci**

- Alpha or gamma hemolytic
  - Lack hemolysins and toxins of beta strep
- Normal flora of upper respiratory tract
- Major cause of dental caries
- Opportunistic pathogen causing sepsis in neutropenic cancer patient
- Important cause of endocarditis

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**Viridans Streptococci**

- Sanguis Group:
  - S. sanguis, S. parassanguis, S. gordonii, S. sinensis
- Mitis Group:
  - S. mitis, S. oralis, S. crista
- Mutans Group:
  - S. mutans, S. sobrinus
- Salivarius Group:
  - S. salivarius, S. vestibularis, S. infantarius
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**Bovis Group Streptococcus**

- Alpha hemolytic
- Causes bacteremia, meningitis, and both native- and prosthetic-valve endocarditis
- Isolation of Strep Bovis Group S. gallolyticus ss. gallolyticus from blood is associated with carcinoma of colon
- Group D
  - S. gallolyticus ss. gallolyticus (S. bovis biotype I)
  - S. gallolyticus ss. pasteurianus (S. bovis biotype II.2)
  - S. infantarius ss. Infantarius (S. bovis II.1)

**Anginosus Group Streptococcus**

- S. anginosus, S. constellatus, S. intermedius
- Abscess formation in deep tissue
- Can be alpha or beta hemolytic
- (Group C, F, G)
- Usually commensals isolated from mouth, oropharynx, GI tract and vagina
- Have been isolated from 56-81% of brain abscesses either in pure or mixed culture
Case 6

- A 65 year old man has been hospitalized for several weeks post surgery
- He develops symptoms consistent with urinary tract infection including pain and fever
- He was recently catheterized as part of a postsurgical complications
- A Gram positive coccus is recovered from his urine on culture in the laboratory

Case 6 Diagnosis

Catheter Associate Urinary Tract Infection (CAUTI) caused by Enterococcus faecalis
- Urinary tract is most common site of infection
- Often associated with catheterization
- Often a nosocomial pathogen
  - Originates from patient’s bowel flora
  - Spread from patient to patient
- Other sites of infection include: mixed bacterial wound infections and decubitus ulcers, sepsis, endocarditis

Enterococcus sp.

Key Characteristics

- Gram positive diplococci (pairs)
- Alpha-hemolytic colonies
- Not MRSA

Key Characteristics

- Gram positive diplococci (pairs)
- Alpha-hemolytic colonies
- Not MRSA
**Enterococcus sp.**

- Formally Group D Streptococci
  - separate genus now
- Enteric bacteria
- *E. faecalis* “pertaining to feces”
- *E. faecium* “of feces”
- Do not possess a broad range of virulence factors like Staph and Strep
- Inherently resistant to many commonly used antibiotics

**Vancomycin Resistant Enterococci (VRE)**

- Increasing incidence in hospital especially ICU or chemotherapy patients
- Most often in *E. faecium* in U.S.
  - Plasmid-mediated, high or low-level resistance to vancomycin
  - vanA or vanB gene
- Infection prevention risk

**Lab Tools**

- Gram stain
- The Lysins
- Group Specific Carbohydrate
Comparison of Staph vs Strep

- **Staphylococci**
  - Catalase positive
  - Cocci in clusters
  - Grows in minimal media
  - Grows best 35-37°C
  - Prefers aerobic atmosphere

- **Streptococci**
  - Catalase negative
  - Cocci in pairs and chains
  - Requires complex media
  - Grows best 35-37°C
  - Prefers anaerobic or carbon dioxide atmosphere
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**Beta Strep**

**Viridans Strep**

**S. pneumoniae** and **Enterococcus sp.**

- Gram-positive, lancet-shaped cocci (elongated cocci with a slightly pointed outer curvature).
- Usually seen as pairs of cocci (diplococci), but they may also occur singly and in short chains.
Identification of Streptococci/Enterococci

- Hemolytic properties:
  - Complete hemolysis (Beta)
  - Incomplete hemolysis (Alpha)
  - No hemolysis (Gamma)

Lancefield Typing

Latex Agglutination
(mostly beta, sometimes alpha)

S. pneumoniae

Bile Solubility Test
- Sodium deoxycholate
- Distinguishes S. pneumoniae from all other viridans streptococci.
- S. pneumoniae is bile soluble
- Sodium deoxycholate (2% in water) will lyse the pneumococcal cell wall.
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**S. pneumoniae**

- Optochin
  - Strains are sensitive to the chemical optochin (ethylhydrocupreine hydrochloride).
  - Optochin sensitivity = presumptive identification of *S. pneumoniae*
  - Some pneumococcal strains are optochin-resistant

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**PYR Test**

- Quick spot test
- Enterococcus
- Group A strep

PYR (L-pyrrolidonyl-β-naphthylamide) impregnated discs serve as a substrate for detection of pyrrolidonyl peptidase.

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**Antibody Detection**

- Anti-Streptolysin O Antibodies
  - Latex agglutination
- Appear 3-4 weeks after initial exposure to Group A Strep
- Useful for confirming diagnosis of rheumatic fever
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GAS Laboratory Testing

<table>
<thead>
<tr>
<th>Rapid Antigen Tests</th>
<th>Nucleic Acid Amplification Tests</th>
<th>Culture</th>
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<tbody>
<tr>
<td>Proper swab collection Procedure followed</td>
<td>Proper swab collection Procedure followed</td>
<td>Proper swab collection Hold cultures 48 h</td>
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A 7-year-old child presents with a fever, pain in his ankles, knees and wrist, and a new heart murmur. His mother said that he complained of a "sore throat" last month, but the symptoms resolved without taking him to the pediatrician. A rapid screening test for strep throat is negative. His most likely diagnosis is:

A. Toxic shock-like syndrome
B. Rheumatic fever
C. Scarlet fever
D. Puerperal fever
E. Acute glomerulonephritis

An organism is isolated from the blood of a 65 year-old male patient with a diagnosis of probable bacterial endocarditis. The organism displays streptococcus-like morphology on gram stain and is catalase-negative. On blood agar the colonies appear gamma hemolytic and are PYR positive. Patient was being treated with vancomycin plus an aminoglycoside with no response. This isolate is likely to be:

A. Streptococcus pyogenes
B. An Enterococcus species
C. Group B Streptococcus
D. A member of the "Streptococcus milleri" group
E. A Staphylococcus species