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# Staphylococci and other grampositive cocci

key points of the lecture

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# Staphylococci

- Facultatively anaerobic
- Catalase positive
- Gram-positive cocci - clusters

- resistant to dry conditions
- resistant to high salt concentrations
  
- also be found as part of the normal flora of the skin and other sites such as the upper respiratory tract (in the nose)

# Medical important species

Coagulase positive – *Staphylococcus aureus*

Coagulase negative – *S. epidermidis*, *S. saprophyticus*

# Coagulase test

- The test identifies whether an organism is able to produce coagulase (virulence factor)
- Coagulase is an enzyme that clot blood plasma
  - by converting fibrinogen to fibrin
  - The fibrin meshwork surrounds the bacterial cells or infected tissues
- Allows detection of staphylococci and its species
  - Coagulase positive → *S. aureus*
  - Coagulase negative → *S. epidermis* & *S. saprophyticus*

# Coagulase test

- Most strains of *S.aureus* produce one or two types of coagulase:
  - bound coagulase (localized on the surface of the cell wall)
  - free coagulase (localized intracellularly → secreted)

# *Staphylococcus aureus*

- major pathogen within the genus
- causes a wide range of major and minor infections in man
- ability to clot blood plasma by the action of the enzyme coagulase
- cocci are usually arranged in grapelike clusters
- nonsporing, non-motile and usually non-capsulate

# Cultivation – blood agar

- grow on many types of agar for 24 h at 37°C,
- individual colonies are circular with a smooth, shiny surface
- colonies are often pigmented (golden-yellow, hence the '*aureus*')
- Beta-hemolysis



# Blood Agar Culture

## → Cultivation on BA, colour of colonies

- *S. aureus* usually displays → grey & golden yellow colonies
- *S. epidermidis* → white colonies
- *S. saprophyticus* → white colonies

## → Hemolysis:

- *S. aureus* → beta-hemolytic
- *S. epidermidis* → non-hemolytic
- *S. saprophyticus* → non-hemolytic

# Mannitol salt agar (MSA)

- Mannitol salt agar (MSA) is both a selective and differential media used for the identification of Staphylococci
- **7.5 % Salt ( NaCl)** → Allows growth of most Staphylococci  
→ This salt-concentration is too high for most other bacteria inhibiting their growth
- **Mannitol** → Allows identification of *S.aureus*  
→ *S.aureus* metabolizes mannitol producing acid → changing the pH and color → Red to Yellow

Yellow → Mannitol positive (*S.aureus* )

Red → Mannitol negative (*S. epidermidis*)

# Pathogenesis

- *Staph. aureus* is present in the nose of 30% of healthy people and may be found on the skin
- It causes infection most commonly at sites of lowered host resistance:
  - such as damaged skin (e.g. surgical site infection) or
  - mucous membranes (e.g. ventilator-associated pneumonia)

# The most common infections caused by *S. aureus*

## 1) Localized skin infection:

- a) Small, superficial abscesses involving sweat- or sebaceous glands or hair follicles (**folliculitis**)
- b) Subcutaneous abscesses (**furuncles** or **boils**) that form around foreign bodies
- c) Larger, deeper infections (**carbuncles**) that can lead to bacteremia

## 2) Diffuse skin infection-impetigo (pyoderma)

This is a superficial, spreading, crusty skin lesion → usually seen in children

## 3) Deep, localized infections

Osteomyelitis (acute and chronic infection of the bone marrow)

Arthritis (acute infection of the joint space - "**septic joint**")

## 4) Other infections:

**Acute endocarditis, septicemia and necrotizing pneumonia**

The most common  
infections caused by  
*S. epidermis*

- Hospital-acquired infections !!!
- Surgical implants, such as:
  - Prosthetic implants
  - Heart valves
  - Catheters
  - Indwelling devices

The most common  
infections caused by  
*S. saprophyticus*

- Urinary tract infections in females, e.g. cystitis

# Virulence factors

## **Enterotoxins:** (superantigen)

within a few hours, induce the symptoms of staphylococcal food poisoning: nausea, vomiting and diarrhoea

## **Toxic shock syndrome toxin (TSST-1):** (superantigen)

Potent activators of T lymphocytes resulting in the liberation of cytokines such as tumour necrosis factor, and it binds with high affinity to mononuclear cells.

## **Epidermolytic toxins**

commonly produced by strains that cause blistering diseases.

- pemphigus neonatorum
- scalded skin syndrome

## **Panton-Valentine leukocidin (PVL)**

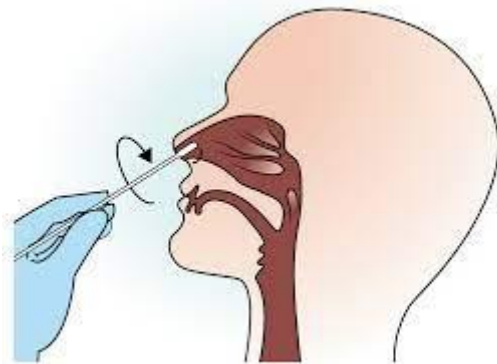
described in the context of community-acquired MRSA

Virulence factor	Activity
<b>Cell wall polymers</b> Peptidoglycan  Teichoic acid	Inhibits inflammatory response; endotoxin-like activity  Phage adsorption; reservoir of bound divalent cations
<b>Cell surface proteins</b> Protein A Clumping factor Fibronectin-binding protein	Reacts with Fc region of IgG Binds to fibrinogen Binds to fibronectin
<b>Exoproteins</b> α-Lysin β-Lysin γ-Lysin δ-Lysin Panton-Valentine leucocidin Epidermolytic toxins Toxic shock syndrome toxin  Enterotoxins  Coagulase  Staphylokinase Lipase Deoxyribonuclease	} Impairment of membrane permeability; cytotoxic effects on phagocytic and tissue cells  Dermo-necrotic and leucocidal Cause blistering of skin Induces multi-system effects; superantigen effects  Induce vomiting and diarrhoea; superantigen effects  Converts fibrinogen to fibrin in plasma  Degrades fibrin Degrades lipid Degrades DNA

# Laboratory diagnosis

- following specimens should be collected:

- Pus
- Sputum
- Faeces or vomit
- Remains of implicated foods
- Blood
- Mid-stream urine
- Anterior nasal and perineal swabs



# Methods of laboratory testing

- Microscopy
- Cultivation on BA
- Cultivation on MSA
- Catalase test
- Coagulase tests
- Biochemical identification
- ATB susceptibility testing



# Treatment

Resistance is common

Active agents	Agents lacking useful activity
Penicillins <sup>a</sup> (e.g. flucloxacillin)	Aztreonam
Cephalosporins (e.g. cefuroxime)	Polymyxins
Aminoglycosides <sup>b</sup> (e.g. gentamicin)	Mecillinam
Tetracyclines (e.g. doxycycline)	Nitroimidazoles
Macrolides (e.g. clarithromycin)	Quinolones <sup>c</sup>
Lincosamides (clindamycin)	
Glycopeptides (vancomycin & teicoplanin)	
Fluoroquinolones <sup>c</sup> (moxifloxacin)	
Rifampicin <sup>b</sup>	
Fusidic acid <sup>b</sup>	
Trimethoprim	
Chloramphenicol	
Carbapenems (e.g. meropenem)	
Oxazolidinones (linezolid)	
Lipopeptide (daptomycin)	

Greenwood, D., Barer, M., Slack, R., Irwing, W. Medical Microbiology. 18.ed. Edinburgh: Elsevier Saunders, 2012.778 s.

# MRSA

- MRSA is increasingly prevalent in hospitals
- Flucloxacillin and vancomycin or teicoplanin are the agents of choice to treat methicillin-susceptible and methicillin-resistant *Staph. aureus* infections, respectively but newer options include linezolid and daptomycin.
- Coagulase-negative staphylococci (such as *Staph. epidermidis*) are major pathogens involving prosthetic implants such as intravascular lines or cardiac valves; the pathogenesis involves biofilm production.

# Key points - general

- Staphylococci are commonly found on the skin of healthy individuals. *Staph. aureus* is present in the nose of 30% of healthy people but can cause infections where there is lowered host resistance.
- Many virulence factors have been described for *Staph. aureus*.
- Organisms spread from colonized sites (e.g. skin) by hands, clothing, dust and desquamation from the skin.

# Enterococcus

- They have natural habitat in the human intestines
- most commonly associated with human disease are:
  - *E. faecalis*
  - *E. faecium*

# Enterococcus

- The diseases with which they are associated are:
  - urinary tract infection
  - infective endocarditis
  - biliary tract infections
  - suppurative abdominal lesions
  - peritonitis

- *E. faecalis* and *E. faecium* are important causes of wound and urinary tract infection in hospital patients and may cause sporadic outbreaks.
- Bacteremia carries a poor prognosis as it often occurs in patients with major underlying pathology and in those who are immunocompromised.

# LABORATORY DIAGNOSIS

- ability to grow on bile-containing media
- Gram-positive cocci
- Group D streptococci

# Therapy

- Enterococci are naturally resistant to cephalosporins.
- Sensitivity to penicillins and other antibiotics varies widely, and clinical isolates must be tested for their susceptibility.
- Vancomycin resistance has been observed in enterococci and is a problem in some hospitals.



# Peptostreptococcus

- Gram-positive cocci
- Non-sporing
- **Anaerobic**
- Found as a part of normal physiological flora:
  - Skin
  - Mouth
  - Genitourinary tract
  - Gastrointestinal tract
- Most are found as part of the flora of the bowel and are not usually considered to be significant in infections.

# Infections with anaerobic cocci

- isolated from infections in various parts of the body, particularly from abscesses
- often found in association with other anaerobic, facultatively anaerobic or aerobic organisms
- mixed infections

# Therapy

- In many infections caused by anaerobes the most important aspect of treatment is surgical.
- This often involves drainage of pus from abscesses, curettage and removal of necrotic tissue.
- For minor infections surgical drainage alone may be sufficient
- In many cases antimicrobial chemotherapy is also indicated:
  - The main groups of agents used are the penicillins and the nitroimidazoles, particularly metronidazole.

# Sources

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