**Micrococcus** species

- **Gram Stain**
  - Gram positive cocci
  - Characteristically in tetrads
  - Usually larger than *Staphylococcus* species

- **Colony morphology**
  - *Micrococcus luteus* = yellow pigment
  - *Micrococcus roseus* = pink pigment

- **Biochemical tests**
  - Catalase positive
  - Modified oxidase positive
Staphylococcus aureus

- **Gram Stain**
  - Gram positive cocci in clusters

- **Colony morphology**
  - Round, smooth, white, creamy colonies with beta-hemolysis
  - May have yellow pigment with extended incubation

- **Biochemical tests**
  - Catalase positive
  - Coagulase positive

- **Pathology**
  - Wound & skin infections, toxic shock syndrome, food poisoning, pneumonia, osteomyelitis, bacteremia, etc.
Staphylococcus epidermidis

- **Gram Stain**
  - Gram positive cocci in clusters

- **Colony morphology**
  - Round, smooth, white, creamy colonies, no hemolysis

- **Biochemical tests**
  - Catalase positive
  - Coagulase negative
  - Novobiocin “S”

- **Pathology**
  - Normal skin flora, nosocomial infections, prosthetic valve endocarditis, catheters, septicemia
**Staphylococcus saprophyticus**

- **Gram Stain**
  - Gram positive cocci in clusters
- **Colony morphology**
  - Round, smooth, white, creamy colonies, no hemolysis
  - May have yellow pigment
- **Biochemical tests**
  - Catalase positive
  - Coagulase negative
  - Novobiocin “R”
- **Pathology**
  - UTI’s in young, sexually active females
Microdase (Modified Oxidase) Test

• Rapid test
  – Two minutes for reaction
  – Detects enzyme – cytochrome oxidase
  – Based on oxidase reaction but contains DMSO to release cytochrome oxidase

• Used to differentiate between
  – *Staphylococcus* species
    • No color change = negative for cytochrome C
  – *Micrococcus* species
    • Blue color = positive for cytochrome C
Bacitracin ("A", "BC") Disk

- Overnight disk diffusion test
- Concentration of bacitracin is 0.04 UI
- Used to differentiate between
  - *Micrococcus* species = "S" (zone of no growth)
  - *Staphylococcus* species = "R" (growth right up to disk)
Coagulase Tube Test

• Tests for free coagulase – an extracellular enzyme that causes a clot to form when bacterial cells are incubated in rabbit plasma

• Used to differentiate between
  – *Staphylococcus aureus* = positive (clot at 4 hours)
    • Some strains of *Staph. aureus* may lyse the clot (due to staphylokinase) so it is important to check reaction every ½ hour for the first 4 hours
  – Other *Staphylococcus* species = negative (no clot at 24 hours)
    • Report as “Coagulase negative *Staphylococcus* species”
Mannitol Salt Agar

• Media classified as selective and differential
  – Selective for *Staphylococcus* species (↑ NaCl)
  – Differential for the fermentation of mannitol, the sole carbohydrate in the media
    • Mannitol fermented → ↓ pH → indicator turns pink to yellow

• *Staphylococcus aureus* = positive, yellow colonies surrounded by yellow zone
  – Note: some coag neg *Staph* can be +

• Other *Staphylococcus* species & *Micrococcus* species = negative, reddish colonies – no color change of media
DNase Agar

- Media classified as differential
- Inoculum is a dime-sized area of growth
- Used to detect the production of an active DNase exoenzyme by aerobic bacteria
- Used to differentiate
  - *Staphylococcus aureus* = positive, pink color around growth
  - Other *Staphylococcus* species = negative, no color change
Novobiocin Disk

- Disk diffusion test using novobiocin (antibiotic) at a concentration of 5 µg for the presumptive identification of *Staphylococcus saprophyticus* in urine cultures

- *Staphylococcus saprophyticus* = “R”
  - Organism grows up to the disk or a zone ≤ 16 mm

- Other *Staphylococcus* species = “S”
  - Organism’s growth is inhibited by antibiotic with a zone > 16 mm