Coagulase Testing

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 Principle: Staphylococcus aureus is the only significant human pathogen that produces the enzyme coagulase. Coagulase is an enzyme that changes soluble fibrinogen into soluble fibrin. When *Staphylococcus aureus* is combined with rabbit plasma, soluble fibrin clumps are formed. Bound coagulase is indicated by a positive result in the slide test and free coagulase is indicated by a positive result in the tube test.

• Purpose: Differentiate *Staphylococcus aureus* from other *Staphylococcus species*.

Procedure:

- Slide test:
 - Place a drop of rabbit plasma on a clean glass slide.
 - Place a drop of distilled water on a separate area of the glass slide.
 - Emulsify a colony of the organism in the distilled water using a sterile inoculating loop. This is the negative control.
 - Emulsify a colony of the organism in the rabbit plasma using a sterile inoculating loop.
 - Rock the slide back and forth gently for one minute.
 - Record results:
 - Positive: white clumps in plasma
 - Negative: no clumps

Procedure:

- Tube test:
 - Place 0.5 mL of rabbit plasma in a test tube.
 - Place 0.5 mL of distilled water in a test tube.
 - Add one inoculating loopful of the organism being tested to each tube.
 - Incubate the test tubes at 37 C for intervals of 30 minutes.
 Every 30 minutes check the test tubes. Tubes that are negative after 4 hours should be incubated at room temperature for 18 to 24 hours.
 - Record results:
 - Positive: fibrin clot formation in the test tube
 - Negative: no formation of fibrin clot

Other Information

- The coagulase tube test detects the free coagulase. Free coagulase is an extracellular toxin that reacts in the presence of CRF (coagulase reacting factor) that resembles thrombin and changes fibrinogen to fibrin.
- There are several strains of Staphylococcus aureus that produce staphylokinase, a fibrinolysin that slows the fibrin clotting process. Incubation for 18 – 24 hours will catch the slower process and catch false negatives.
- Other tests for *Staphylococcus aureus* are growth and fermentation on MSA. *S. aureus* can grow in 6.5% to 10% NaCl and ferments the mannitol. Fermentation is seen by the yellow color in the agar as the phenol red indicator changes color due to the acid produced.