**Introductory Medical Microbiology**

**Laboratory Notes**

**BIOL 2161L**

# CATALASE TEST

Catalase is an enzyme that decomposes hydrogen peroxide into oxygen and water.  Excluding the *Streptococci*, most aerobic and facultative anaerobic bacteria possess catalytic activity.

**Methodology**Hydrogen peroxide forms as one of the oxidative end products of aerobic carbohydrate metabolism.  Catalase converts hydrogen peroxide into water and oxygen.  The catalase test is commonly used to differentiate streptococci (negative) for staphylococci (positive).

**Specimen Requirements**Well -isolated colonies of an 18-24 hour culture.

**Reagents and Equipment**

3% Hydrogen peroxide, Clean glass slide, Dropper, Bacteriological Loop

**Controls**Positive:  *Staphylococcus aureus*  
Negative:  *Streptococcus sp.*

**Procedure:**1. With loop or applicator stick, transfer cells from the center of a well-isolated colony to a glass slide.  
2. Add 1-2 drops of the 3% Hydrogen peroxide to the bacterial cells  
      *Note:  It is recommended to use this order instead of adding the organism to the reagent,  
         particularly if iron containing loops are used, due to false positives*

**Interpretation**Positive: rapid, appearance of sustained gas bubbles  
Negative:  No gas bubble production

# COAGULASE TEST

**Coagulase** catalyzes the conversion of fibrinogen to fibrin in blood plasma.  The network of fibrin formed in host tissue infected with coagulase producing organisms serves to protect the bacterium from the defenses of the host.  This characteristic is a primary indicator of virulence among staphylococci. This test can be used to differentiate *Staphylococcus aureus* from *Staphylococcus epidermidis* as well as streptococcus organisms.  Coagulation within 24 hours is indicative of *Staphylococcus aureus.*

**Materials**

* Well-isolated colonies
* BD BBL Staphlyoslide Test Latex (Latex beads that have been sensitized with IgG and human fibrinogen
* BD BBL Staphyloslide Control Latex (Latex beads that are sensitized)
* Test cards
* Sterile inoculating loops/needles for mixing

**Procedure**1. Your instructor will provide you with a card that contains two circles. One circle will be indicated as (T) and will contain the text latex beads (sensitized) and the other will contain the control (C) latex beads (sensitized).   
2. Using a sterile loop or needle, transfer a colony of the test isolate into the circle. Mix this into the test latex reagent and spread to cover the complete area of the circle. Repeat this step with the same colony using another sterile loop or needle for the control latex reagent.   
3. Gently rock the card allowing the mixture to flow slowly over the entire test ring area.  
4. Observe for agglutination for up to 20 seconds.

5. Dispose of cards and needles in the bleach containers and/or biohazard bags provided.

**Interpretation**Positive result will agglutinate (clump):  *Staphylococcus aureus*  
Negative result will NOT agglutinate:  *Staphylococcus epidermidis*, *Streptococcus sp.*