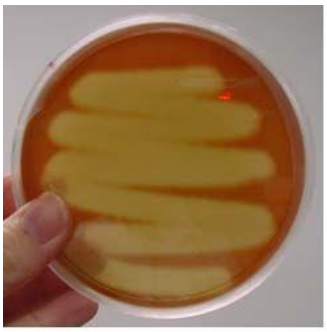
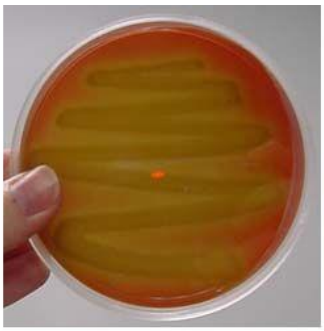
Throat and Skin Substitute

Throat Culture Using Blood Agar

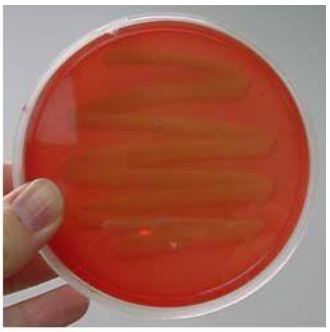
Blood agar plates (BAP) are TSA plus 5% sheep's blood. The blood enriches the agar allowing fastidious microbes to grow. A fastidious microbe is one that requires extra nutrients to grow. Blood agar is also differential. The type of hemolysis can be determined. There are three types of hemolysis. Alpha-hemolysis is the partial break-down of the hemoglobin. The agar turns a brownish/greenish color. Alpha-hemolysis is always present in throat cultures. Beta-hemolysis is the complete break-down of the hemoglobin. The agar turns clear. It is usually clear enough to see thru. Some common bacteria are beta-hemolytic such as many *Staphylococcus* species and *Bacillus cereus*. Gamma-hemolysis is no hemolysis. The blood agar is still nice and red under the bacterial growth. (α = alpha, β = beta, γ = gamma)



Beta Hemolysis



Alpha Hemolysis



Gamma Hemolysis

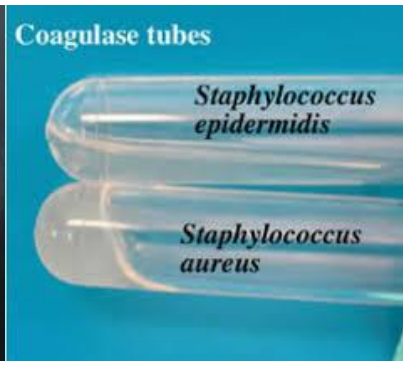
Skin Culture Using MSA

To differentiate the three classic species of *Staphylococcus* (*S. aureus*, *S. epidermidis, S. saprophyticus*), three tests are performed: MSA, coagulase, and novobiocin sensitivity.

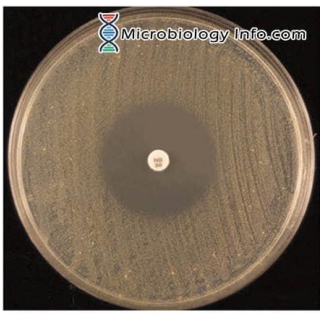
MSA – all *Staph.* grows on MSA. *S. aureus* ferments mannitol producing acid and turning the agar yellow.

Coagulase – *S. aureus* produces coagulase that cleaves fibrinogen to fibrin. Fibrin associates to form clots.

Novobiocin Sensitivity – *S. saprophyticus* is resistant to novobiocin and has little or no zone of inhibition.



NOVOBIOCIN



Staphylococcus saprophyticus
Resistant (less than 16 mm)

Staphylococcus aureus
Sensitive (greater than 16 mm)

Take Home Lesson: If given a blood agar plate, you should be able to recognize the different types of hemolysis (alpha, beta, gamma). If given a mannitol salt agar plate, coagulase tube inoculated with *Staphylococcus*, you should be able to differentiate *S. aureus* from *S. epidermidis*. You should refresh your knowledge of the purpose of mannitol salt agar.

Test	*Staphylococcus aureus*	*Staphylococcus epidermidis*	*Staphylococcus saprophyticus*
Mannitol Fermentation	**+, yellow agar, growth**	**-, no yellow agar, growth**	**-, no yellow agar, growth**
Coagulase	**+, clot forms**	**-, no clot**	**- no, clot**
Novobiocin Sensitivity	**Susceptible (>=22mm zone)**	**Susceptible (>=22mm zone)**	**Resistant (<=17mm zone)**