

COAGULASE TEST

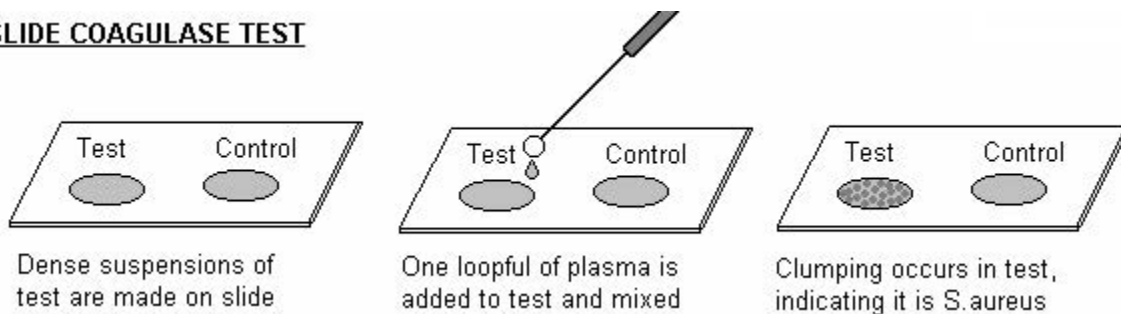
Staphylococcus aureus is known to produce coagulase, which can clot plasma into gel in tube or agglutinate cocci in slide. This test is useful in differentiating *S.aureus* from other coagulase-negative staphylococci. Most strains of *S.aureus* produce two types of coagulase, free coagulase and bound coagulase. While free coagulase is an enzyme that is secreted extracellularly, bound coagulase is a cell wall associated protein. Free coagulase is detected in tube coagulase test and bound coagulase is detected in slide coagulase test. Slide coagulase test may be used to screen isolates of *S.aureus* and tube coagulase may be used for confirmation. While there are seven antigenic types of free coagulase, only one antigenic type of bound coagulase exists. Free coagulase is heat labile while bound coagulase is heat stable.

SLIDE COAGULASE TEST:

Principle: The bound coagulase is also known as clumping factor. It cross-links the α and β chain of fibrinogen in plasma to form fibrin clot that deposits on the cell wall. As a result, individual coccus stick to each other and clumping is observed.

Procedure: Dense suspensions of Staphylococci from culture are made on two ends of clean glass slide. One should be labeled as “test” and the other as “control”. The control suspension serves to rule out false positivity due to autoagglutination. The test suspension is treated with a drop of citrated plasma and mixed well. Agglutination or clumping of cocci within 5-10 seconds is taken as positive. Some strains of *S.aureus* may not produce bound coagulase, and such strains must be identified by tube coagulase test.

SLIDE COAGULASE TEST



TUBE COAGULASE TEST:

Principle: The free coagulase secreted by *S.aureus* reacts with coagulase reacting factor (CRF) in plasma to form a complex, which is thrombin. This converts fibrinogen to fibrin resulting in clotting of plasma.

Procedure: Three test tubes are taken and labeled “test”, “negative control” and “positive control”. Each tube is filled with 0.5 ml of 1 in 10 diluted rabbit plasma. To the tube labeled test, 0.1 ml of overnight broth culture of test bacteria is added. To the tube labeled positive control, 0.1 ml of overnight broth culture of known *S.aureus* is added and to the tube labeled negative control, 0.1 ml of sterile broth is added. All the tubes are incubated at 37°C and observed up to four hours. Positive result is indicated by gelling of the plasma, which remains in place even after inverting the tube. If the test remains negative until four hours at 37°C, the tube is kept at room temperature for overnight incubation.

Application: Coagulase test is used to identify and differentiate *S.aureus* from coagulase negative staphylococci. While slide coagulase test is useful in screening, tube coagulase test is useful in confirmation of coagulase test. Not all *S.aureus* strains produce coagulase; such rare strains are identified by thermonuclease test. Some coagulase negative staphylococci such as *S.lugdensis* and *S.schleiferi* are known to give positive slide coagulase test while *S.hyicus* and *S.intermedius* are known to give positive tube coagulase test.

TUBE COAGULASE TEST

