National Department of Health

Title: Tube coagulase test

ID: G_90_T_2_A

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Changes to the last authorized version:

Version	Date issued	Changes

1. Purpose and Scope

This document describes the Coagulase test procedure used to differentiate Staphylococcus sp. This test detects the ability of bacteria to clot plasma by the action of the enzyme, coagulase.

2. Principle/Clinical application

This test is used to differentiate the species within the genus Staphylococcus.
Staph. aureus is Coagulase Positive.

Coagulase is an enzyme produced by microorganisms that bind plasma fibrinogen- causing the organisms to agglutinate or plasma to clot.

The Tube coagulase test detects free coagulase which is an extracellular protein enzyme. The clotting mechanism involves activation of a Coagulase Reacting Factor (CRF) in the rabbit plasma to form a coagulase-CRF complex. This complex in turn reacts with fibrinogen to produce the clot.

3. Responsibilities

- Staff performing coagulase require specific training and demonstrated competency.
- Staff performing Coagulase are responsible for the setup, reading and recording of the coagulase result.
- Staff are required to test and record Coagulase positive and negative controls daily.

4. Specimen

- Test isolated colonies of Staph. species (Gram positive cocci and catalase positive)

5. Safety

For safety aspects, please review this document G_10_Info_3 Laboratory Biosafety

6. Equipment and Materials

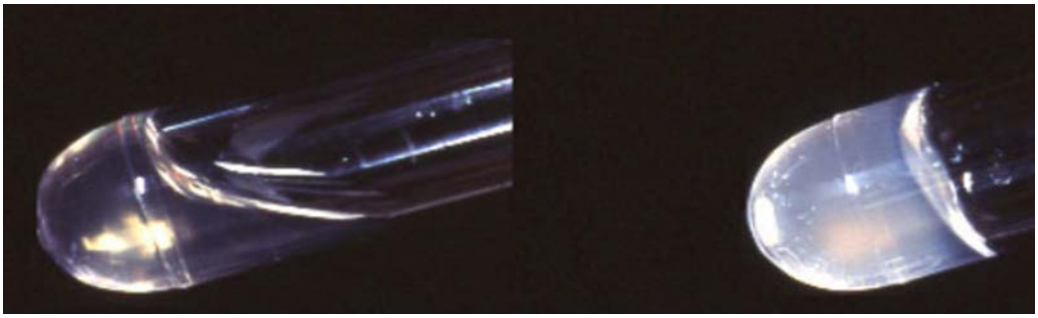
- Commercially available Rabbit plasma
- Distilled Water to dilute Rabbit Plasma
- Quality Control organisms: S. aureus ATCC 29213 & S. epidermidis ATCC 12228
- 5ml Glass or plastic tubes
- Test tube rack
- Plastic disposable pipettes
- Marking pen
- 37^{o}C Waterbath or incubator
- Gloves
- Loops for inoculation
- Loop steriliser
- Timer

7. Procedure

7.1 Use commercially available plasma and dilute according to manufacturer's instructions
7.2 Label the test tubes with the organism to be tested as well as the control organisms

7.3 Add 0.25 mL of plasma to each tube
7.4 Emulsify representative colony/colonies of the test organism in the plasma.

7.5 Incubate at 35-37°C and examine hourly up to 4hr. Do not shake or agitate the tube.



8. Results Recording

- Record all results onto paper worksheet with registered lab number and patient identification.
- Record results into LIMS

9. Interpretation

- A positive coagulase will be demonstrated by the formation of a clot which gels the whole contents of the tube or forms a loose web of fibrin.
- If tubes are still negative at 4 hrs- place tubes at room temperature overnight. Leaving tubes at 37C >4hrs can result in false negatives results.

10. Quality Control

- Staph aureus ATCC 29213 - Coagulase Positive
- S. epidermidis ATCC 12228 – Coagulase Negative
- Record the QC results on the Bench Reagent QC Worksheet

11. Related Documents

- Bench Reagent QC Worksheet G_90_WS_1
- Laboratory Biosafety Info Sheet G_10_Info_3

12. References

- DMDP Coagulase Test SOP- Joanne Letchford 2013
- UK Standards for Microbiology Investigations https://www.gov.uk/uk-standards-for-microbiology-investigations-smi-quality-and-consistency-in-clinical-laboratories