PROGRAM Standard Operating Procedure – Laboratory Services	
Title: MIC50200 – Tube Coagulase Test	Policy Number:
Program Name: Laboratory Services	
Applicable Domain: Lab, DI and Pharmacy Services	
Additional Domain(s):	
Effective Date:	Next Review Date:
Issuing Authority: Director of Health Services	Date Approved:
Accreditation Canada Applicable Standard: N/A	

GUIDING PRINCIPLE:
The tube coagulase test is used to differentiate *Staphylococcus* spp. by determining the organism's ability to clot plasma using the enzyme coagulase.

PURPOSE/RATIONALE:
This standard operating procedure describes how to perform the tube coagulase test.

SCOPE/APPLICABILITY:
This procedure applies to Medical Laboratory Technologists (MLTs) performing the tube coagulase test.

SAMPLE INFORMATION:

Type	Several, well-isolated colonies that are: • Gram-positive cocci • Catalase positive • 18 to 24 hours old

REAGENTS and/or MEDIA:

Type	BD BBL Coagulase Plasma, Rabbit with EDTA
Reagent Preparation	• Reconstitute lyophilized reagent with 15 mL of sterile water
Stability and Storage Requirements	• Store unopened lyophilized coagulase plasma at 2°C to 8°C • Store reconstituted plasma at 2°C to 8°C for up to 14 days

SUPPLIES:

- Sterile water
- 10 mL plastic syringe
- Glass test tubes
- Sterile pipettes
- Disposable loops

EQUIPMENT

- 35° ambient air incubator

SPECIAL SAFETY PRECAUTIONS:

Containment Level 2 facilities, equipment, and operational practices for work involving infectious or potential infectious materials or cultures.

- Ensure that appropriate hang hygiene practices be used.
- Lab gown must be worn when performing activities with potential pathogens.
- Gloves must be worn when direct skin contact with infected materials is unavoidable.
- Eye protection must be used when there is a known or potential risk of exposure of splashes.
- All procedures that may produce aerosols, or involve high concentrations or large volumes should be conducted in a biological safety cabinet (BSC).
- The use of needles, syringes and other sharp objects should be strictly limited.

All patient specimens are assumed to be potentially infectious. Routine Practices must be followed. Since viable micro-organisms are used, all cultures must be handled with appropriate precautions. All equipment in contact with cultures should be decontaminated by appropriate methods.

QUALITY CONTROL:

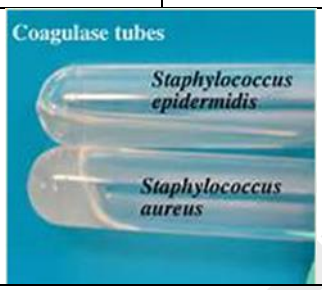
- Quality control is performed daily:
  - Positive: *Staphylococcus aureus* ATCC 25923
  - Negative: *Staphylococcus epidermidis* ATCC 12228
- A TQC order is automatically generated daily to record the QC results

PROCEDURE INSTRUCTIONS:

Step	Action
Performing the tube coagulase test	
1	Using a sterile pipette, add approximately 0.5 mL of rehydrated coagulase reagent to a labelled glass test tube.
2	Using a disposable loop, thoroughly emulsify several colonies from a non-inhibitory agar plate in the tube of plasma and mix gently.
3	Incubate in the O_2 incubator for up to 6 hours.
4	Examine the tubes periodically by gently tipping the tube; avoid shaking the tube, which could cause breakdown of the clot and false negatives.
5	If no clot is visible after 6 hours, leave at room temperature overnight and observe for clot formation.

INTERPRETATION OF RESULTS:

IF	THEN
Any degree of clot formation	**Tube coagulase = Positive**
No clot formation	**Tube coagulase = Negative**



LIMITATIONS:

1. Some species of organisms utilize citrate in their metabolism and will yield false-positive reactions for coagulase activity. Normally, this does not cause problems since the coagulase test is performed almost exclusively on *Staphylococci*. However, it is possible that bacteria that utilize citrate may contaminate *Staphylococcus* cultures on which the coagulase test is being performed. These contaminated cultures may, upon prolonged incubation, give false-positive results due to citrate utilization.
2. Some strains of *S.aureus* produce staphylokinase, which may lyse clots. If the tubes are not read until 24 hours of incubation, false-negative results may occur.
3. Do not use plasma if a heavy precipitate or clot has formed before inoculation.
4. Do not shake or agitate the tube during the test. This may cause a breakdown of the clot, which will not reform upon additional incubation.
5. False negative coagulase reactions may occur if the test culture is older than 18 to 24 hours or if there is scant inoculum.
6. False-positive results may occur if the test isolate is removed from agar containing high concentrations of salt. Use only strains grown on non-selective media.

REFERENCES:

1. BD BBL. (2017-02). *Coagulase Plasmas* package insert

APPROVAL:

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Date

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REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	04 Apr 19	Initial Release	L. Steven
2.0	30 Jun 21	Procedure reviewed and added to NTHSSA policy template	L. Steven