PROGRAM Standard Operating Procedure – Laboratory Services	
Title: MIC51100 – Beta-Lactamase 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 beta-lactamase test is used to rapidly detect the production of beta-lactamase in colonies of *Neisseria gonorrhoeae*, *Moraxella catarrhalis*, *Staphylococcus* spp., *Enterococcus* spp., *Haemophilus influenzae* and anaerobic bacteria.

PURPOSE/RATIONALE:
This standard operating procedure describes how to perform the beta-lactamase test.

SCOPE/APPLICABILITY:
This procedure applies to Medical Laboratory Technologists (MLTs) performing the beta-lactamase test.

SAMPLE INFORMATION:

Type	Few, well isolated colonies

REAGENTS and/or MEDIA:

Type	BD BBL Cefinase Disks
Stability and Storage Requirements	• Store unopened package at -20°C to 8°C • After opened, store in air tight container containing desiccant at -20°C to 8°C • Do not use the cartridge if the disks appear orange or red in color

SUPPLIES:
- Glass microscope slides
- Forceps
- Sterile saline
- Sterile pipettes
- Wooden sticks
- Disposable loops

SPECIAL SAFETY PRECAUTIONS:

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

- Ensure that appropriate hand 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 weekly:
  - Positive: *Staphylococcus aureus* ATCC 29213
  - Negative: *Haemophilus influenzae* ATCC 10211
- A TQC order is automatically generated on Wednesdays to record the QC results

PROCEDURE INSTRUCTIONS:

<table>
<tr><th>Step</th><th colspan="4">Action</th></tr>
<tr><td colspan="5">Performing the beta-lactamase test</td></tr>
<tr><td>1</td><td colspan="4">Remove disks from the fridge and bring to room temperature.</td></tr>
<tr><td>2</td><td colspan="4">Using forceps remove the required number of disks from the dispenser and place on a microscope slide. Use 1 disk per organism.</td></tr>
<tr><td>3</td><td colspan="4">Using a sterile pipette, moisten each disk with a drop of sterile saline.</td></tr>
<tr><td>4</td><td colspan="4">With a disposable loop or wooden stick, pick several similar colonies from the agar plate and smear onto the surface of the disk.</td></tr>
<tr><td>5</td><td colspan="4">Observe the disk for up to 5 minutes for a colour change. For Staphylococci, observe the disk for up to 60 minutes.</td></tr>
<tr><td rowspan="4">6</td><td>Organism</td><td>Result</td><td>Time</td><td>Interpretation</td></tr>
<tr><td>Staphylococcus aureus</td><td>Positive</td><td>1 hour</td><td>Resistant to penicillin, ampicillin Probably susceptible to cephalothin, methicillin, oxacillin and other penicillinase-resistant penicillins</td></tr>
<tr><td>Enterococcus faecalis</td><td>Positive</td><td>5 minutes</td><td>Resistant to penicillin and ampicillin</td></tr>
<tr><td>Haemophilus influenzae</td><td>Positive</td><td>1 minute</td><td>Resistant to ampicillin Susceptible to cephalosporins</td></tr>
</table>

	Neisseria gonorrhoeae *Moraxella catarrhalis*	Positive	1 minute	Resistant to penicillin

INTERPRETATION OF RESULTS:

IF	THEN
Yellow to red colour at site of inoculation	**Beta-lactamase = Positive**
No colour change on the disk	**Beta-lactamase = Negative**

LIMITATIONS:

1. The efficacy of this test in predicting the β-lactam resistance of microorganisms other than *Neisseria gonorrhoeae*, *Haemophilus influenzae*, *Moraxella catarrhalis*, *Staphylococci*, *Enterococci* and certain anaerobic bacteria is unproven.
2. Resistance to β-lactam antibiotics has been, on rare occasions, reported in some of the above organisms without the production of β-lactamases. In these cases, resistance mechanisms such as permeability barriers have been postulated. Therefore, the β-lactamase test should be used as a rapid supplement and not a replacement for conventional susceptibility testing.
3. For some strains of *Staphylococci*, particularly *S.epidermidis*, an inducible β-lactamase has been described that might result in a false-negative β-lactamase reaction with a strain which is resistant to penicillin or ampicillin.

REFERENCES:

1. BD BBL. (2018-09). *Paper Disks for the Detection of ß-Lactamase Enzymes* 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