PROGRAM Standard Operating Procedure – Laboratory Services	
Title: MIC31000 – MRSA Screen	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:
Specimens are submitted to identify carriers of methicillin-resistant *Staphylococcus aureus* (MRSA). Swabs may be submitted from any body site, but most common are nasal, groin and wound swabs. Combined nasal/axilla/rectal/perineum swabs may also be processed. MRSA may occasionally be found exclusively in urine or sputum and specimens from such sites should be processed after consultation with Infection Prevention and Control.

Chromogenic MRSA Screening Agar is selective medium for the isolation of Methicillin Resistant Staphylococcus aureus (MRSA). The medium uses a chromogen which yields a denim blue colour as a result of phosphatase activity. This enzyme is present in all MRSA. The antibiotic solution in the medium is selective for *Staphylococcus aureus*, containing compounds that inhibit the growth of competitor organisms while some encourage the production of MRSA markers.

PURPOSE/RATIONALE:
To screen for Methicillin Resistant *Staphylococcus aureus* (MRSA) on admission and as part of Multi-Resistant Organism (MRO) screens.

SCOPE/APPLICABILITY:
This procedure applies to Medical Laboratory Technologists (MLTs) processing specimens for MRSA screen.

SAMPLE INFORMATION:

Type	Swab • Amie's with or without charcoal
Source	• Bilateral nasal swab • Bilateral groin swab • MRO screen: any site

Stability	If the sample is received in the laboratory and processed greater than 48 hours from collection: • Add specimen quality comment: "Delayed transport may adversely affect pathogen recovery"
Storage Requirements	Room temperature
Criteria for rejection	1. Unlabeled/mislabeled swabs 2. Specimen container label does not match patient identification on requisition 3. Duplicate specimens obtained with same collection method from same collection location within 24 hours

REAGENTS and/or MEDIA:

- Denim Blue agar (DEN) and Blood agar (BA)
- Identification reagents: gram stain, catalase, Staph latex test and tube coagulase

SUPPLIES:

- Disposable inoculation needles
- Wooden sticks

EQUIPMENT

- Biosafety
- cabinet
- 35° ambient air incubator
- Vitek 2 and supplies

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.

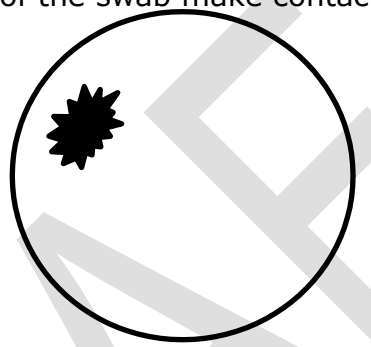
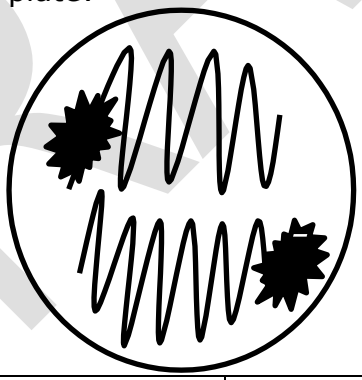
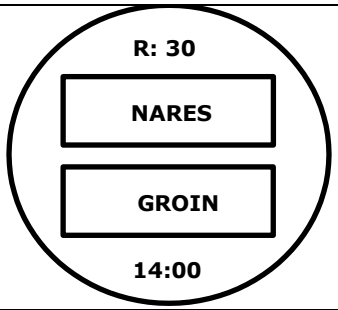
All patient specimens are assumed to be potentially infectious. Routine Practices must be followed. Since viable micro-organisms are used, all cultures must be handled with appropriate precautions. All equipment in contact with cultures should be decontaminated by appropriate methods.

QUALITY CONTROL:

- Refer to MIC60040-Culture Media Quality Control procedure
- Refer to Test Manual for reagent quality control procedures

PROCEDURE INSTRUCTIONS:

- Monday to Friday: MRSA swabs are processed at 14:00
- Saturday and Sunday: MRSA swabs are processed at 14:00

<table>
<tr><th>Step</th><th>Action</th></tr>
<tr><td colspan="2">Processing swabs for MRSA screen</td></tr>
<tr><td>1</td><td>In the biosafety cabinet:
• Inoculate top-left corner of DEN with the swab
• Ensure all surfaces of the swab make contact the agar:
 
• Streak for isolated growth using a disposable inoculation needle to cover half of the plate:
  </td></tr>
<tr><td>2</td><td>Mark on Denim Blue plate:
• R (for read) followed by the read date (24 hours from day of planting)
• Time of planting (14:00)
Reason: Plates are read after 24 hours after incubation.
  </td></tr>
<tr><td>3</td><td>Incubate the media:
• Place DEN in the O_2 incubator in appropriate tray</td></tr>
</table>

INTERPRETATION OF RESULTS:

<table>
<tr><th>Step</th><th colspan="3">Action</th></tr>
<tr><td>1</td><td colspan="3">• Observe DEN plate at 18 to 24 hours (8:00 to 14:00)
• Examine for denim blue colonies</td></tr>
<tr><td rowspan="5">2</td><td>IF</td><td colspan="2">THEN</td></tr>
<tr><td>No denim blue colonies seen at 18-24 hours</td><td colspan="2">• Record observations in the LIS
• Workup complete
• MRSA not isolated</td></tr>
<tr><td rowspan="3">Atypical growth (i.e., colonies with blue “halos”, colonies not typical denim blue color)</td><td colspan="2">• Record observations in the LIS
• Subculture isolate to BA plate
• From BA plate, perform:
1. Gram stain (Gram-positive cocci)
2. Catalase (positive)
3. Staph latex test (positive)
4. Tube coagulase (positive)
5. GPS (cefoxitin screen positive)</td></tr>
<tr><td>IF</td><td>THEN</td></tr>
<tr><td>Results from above testing are not consistent with MRSA</td><td>• Workup complete
• MRSA not isolated</td></tr>
<tr><td>Results from above testing are consistent with MRSA</td><td>• Workup complete
• MRSA isolated</td></tr>
<tr><td>Denim Blue colonies seen</td><td colspan="2">• Record observations in the LIS
• Perform Staph latex test from DEN:</td></tr>
<tr><td></td><td>IF</td><td>THEN</td></tr>
<tr><td></td><td>Staph latex test NEGATIVE</td><td>• MRSA not isolated</td></tr>
<tr><td></td><td>Staph latex test POSITIVE</td><td>• MRSA isolated</td></tr>
</table>

REPORTING INSTRUCTIONS:

IF	REPORT
MRSA not isolated	• Report:**“No Methicillin Resistant Staph aureus (MRSA) isolated”**
MRSA isolated	• Add organism: **“Staphylococcus aureus”** • List quantitation as **“Isolated”** • The following isolate comment will be added: **&cx00** • In order entry, copy report to OCPHO (HPU1) • In order entry, copy report to appropriate IPAC ward if ER or In-patient • In order entry add ESO code “MRSA”

NOTE:
STH IPAC ward is **SIPAC**. IRH IPAC ward is **IIPAC**. Territorial IPAC ward is **TIPAC**

LIMITATIONS:

1. Heavy inoculation may lead to a blue/green haze appearance in the main inoculum which should not be interpreted as a positive result.
2. Some Bacillus species may produce an atypical, very dark navy-blue colored colony with a halo and crenated edge. Aerococcus species may also appear as dark navy-blue colonies. If in doubt, subculture colonies to Blood agar for further investigation.
3. Incubation beyond 24 hours can result in false positive results. Suspicious colonies detected on a second day of incubation must be sub cultured for additional identification testing.

CROSS-REFERENCES:

- MIC60040-Culture Media Quality Control

REFERENCES:

1. Leber, A. (2016). *Clinical microbiology procedures handbook.* (4th ed.) Washington, D.C.: ASM Press
2. Jorgensen J.H., Pfaller M.A., Carroll K.C., Funke G., Landry M.L., Richter S.S., Warnock D.W. (2015). *Manual of Clinical Microbiology, 11th edition*. Washington, D.C: ASM Press
3. Oxoid. (May 2005). *Denim Blue Agar (Chromogenic MRSA Screening Agar)* package insert

APPROVAL:

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Date

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REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	11 Jan 17	Initial Release	L. Steven
2.0	30 Nov 18	Updated to include new Vitek 2 instrument	L. Steven
3.0	30 Dec 20	Procedure reviewed and added to NTHSSA policy template	L. Steven