

<b>NORTHWEST TERRITORIES</b> <b>Health and Social Services Authority</b>	<b>Laboratory</b> <b>Stanton Territorial Hospital</b> P.O. Box 10, 550 Byrne Road YELLOWKNIFE NT X1A 2N1	<b>Document Number:</b> MIC30300	
		<b>Version No:</b> 2.0	Page: 1 of 6
<b>Document Name:</b> MRSA Screen – Chromogenic Agar		<b>Distribution:</b> <b>Microbiology Culture Manual</b>	
		<b>Effective:</b> 11 January, 2017	
		<b>Date Reviewed:</b> 11 January, 2017 <b>Next Review:</b> 11 January, 2019	
<b>Approved By:</b> Jennifer G. Daley Bernier, A/manager, Laboratory Services		<b>Status:</b> <b>APPROVED</b>	

**PURPOSE:** To screen for *Methicillin Resistant Staphylococcus aureus* (MRSA) on admission, in Multi-resistant Organism (MRO) screens and from infected sites.

**SAMPLE INFORMATION:**

<b>Type</b>	Swab <ul style="list-style-type: none"> <li>Amie’s with or without charcoal</li> </ul>
<b>Source</b>	<ul style="list-style-type: none"> <li>Bilateral nasal swab</li> <li>Bilateral groin swab</li> <li>Other: drainages, wounds, sites of catheters, tracheostomy and other skin penetrating devices</li> </ul>
<b>Stability</b>	If the sample is received in the laboratory and processed greater than 48 hours from collection: <ul style="list-style-type: none"> <li>Add specimen quality comment: “Delayed transport may adversely affect pathogen recovery”.</li> </ul>
<b>Storage Requirements</b>	Room temperature
<b>Criteria for rejection and follow up action</b>	1. Unlabeled/mislabeled swabs 2. Dry swabs

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**REAGENTS and/or MEDIA:**

- Denim Blue agar (DEN) and Blood agar (BAP)
- Identification reagents: rapid Staph

**SUPPLIES:**

- Wooden sticks
- Disposable inoculation needles
- 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.

- 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. Universal precautions 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:**

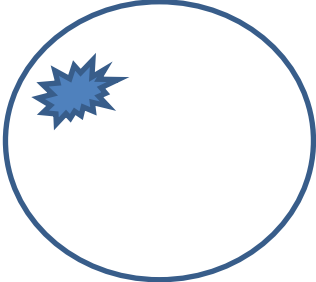
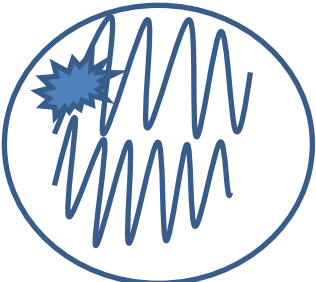
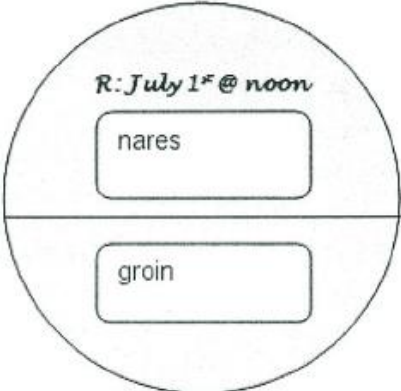
Refer to MIC60100 Non-Exempt Media Quality Control procedure

Refer to Quality Control manual for reagent quality control procedures

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**PROCEDURE INSTRUCTIONS:**

\*Note: MRSA swabs are set up twice a day, Monday → Friday at noon and 17:00. On weekends, they are set up once a day before 15:00.

Step	Action	
<b>Processing specimen for MRSA screening</b>		
<b>1</b>	<p>In the biosafety cabinet, inoculate the top-left corner of the Denim Blue agar from the swab, ensuring all surfaces of swab make contact with the agar.</p> 	
<b>2</b>	<p>Streak for confluent growth using a disposable inoculation needle.</p>  <p>Streak out to cover half the plate.</p>	
<b>3</b>	<p>Mark on Denim Blue plate:</p> <ul style="list-style-type: none"> <li>• “R” (for Read) followed by the date 24 hours from day of planting i.e.: July 1<sup>st</sup></li> <li>• Time of planting i.e.: noon</li> </ul> <p>Reason: Plates are read at approx. 18-24 hours after incubation.</p>	
<b>4</b>	<p>Incubate plate in O<sub>2</sub> incubator at 35° for 18-24 hours in separate batches depending on time of incubation.</p>	

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**INTERPRETATION OF RESULTS:**

Step	Action	
1	Remove culture plates after 18-24 hours incubation.	
2	Observe plates for denim blue colonies.	
		
3	<b>If:</b>	<b>Then:</b>
	No growth OR White colonies	<ul style="list-style-type: none"> <li>Record observations in the LIS.</li> <li>No workup required.</li> </ul>
	Atypical growth (i.e. colonies with blue “halos”, colonies not typical denim blue color)	<ul style="list-style-type: none"> <li>Record observations in the LIS.</li> <li>Should not be interpreted as MRSA.</li> <li>Subculture isolate to BAP to perform further identification testing (i.e. catalase, repeat rapid Staph, tub coag).</li> <li>Atypical colonies that identify as <i>Staphylococcus aureus</i> need to have GPS performed to confirm oxacillin resistance.</li> </ul>
	Denim Blue colonies seen	<ul style="list-style-type: none"> <li>Record observations in the LIS.</li> <li>If sufficient isolated colonies present, perform rapid Staph directly off Denim Blue agar.</li> <li>If sufficient isolated colonies are not present, subculture isolate to BAP to perform rapid Staph from the following day.</li> </ul>

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**REPORTING RESULTS:**

IF:	THEN:
No growth or white colonies present	<ul style="list-style-type: none"> <li>Report:  <b>“No Methicillin Resistant Staphylococcus aureus (MRSA) isolated”</b></li> </ul>
Denim blue colonies, rapid Staph positive	<ul style="list-style-type: none"> <li>Add organism: <b>“Staphylococcus aureus”</b></li> <li>Add quantity: <b>“Isolated”</b></li> <li>Use canned culture comment: <b>“***Methicillin Resistant***, This organism is cloxacillin resistant and is resistant to all beta-lactam agents”</b></li> <li>In order entry, copy report to Chief Medical Officer of Health (HPU1) and Infection Control (SOHS) if in-patient.</li> </ul>

**LIMITATIONS:**

- Heavy inoculation may lead to a blue/green haze appearance in the main inoculum which should not be interpreted as a positive result.
- 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 BA agar for further investigation.
- 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.

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**REFERENCES:**

- Clinical Microbiology Procedures Handbook, 4<sup>th</sup> edition, ASM Press, 2016
- 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, 11<sup>th</sup> edition, ASM Press, Washington, D.C.
- Oxoid Denim Blue agar package insert, May 2005

**REVISION HISTORY:**

REVISION	DATE	Description of Change	REQUESTED BY
1.0	11 Jan 2017	Initial Release	L. Steven
2.0	25 Apr 2018	Change to reflect new Vitek 2 instrument	L. Steven

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