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| **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: | Date Approved: |
| Director of Health Services |  |
| 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. Denim Blue is a selective chromogenic medium for the isolation and direct identification of MRSA. The presence of an optimized salt concentration and an antibiotic-antifungal mixture inhibit the growth of yeasts and the majority of Gram-negative and Gram-positive bacteria with the exception of MRSA.

**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 processing specimens for MRSA screen.

**SAMPLE INFORMATION:**

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| **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.

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

**PROCEDURE INSTRUCTIONS:**

**NOTE:** Monday to Friday: MRSA swabs are processed at noon and 17:00

Saturday and Sunday: MRSA swabs are processed before 15:00

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| **Step** | **Action** | |
| **Processing swabs for MRSA screening** | | |
| **1** | In the biosafety cabinet, inoculate the top-left corner of Denim Blue agar from the swab, ensuring all surfaces of swab make contact with the agar: | |
| **2** | Streak for confluent growth using a disposable inoculation needle:  Streak out to cover half the plate. | |
| **3** | Mark on Denim Blue plate:   * **R** (for read) followed by the read date (24 hours from day of planting) * Time of planting   (12:00 or 17:00)  **Reason:** Plates are read after  24 hours after incubation. | **GROIN**  **NARES**  **12:00**  **R: 30** |
| **4** | Incubate plate in O2 incubator at 35° for 18-24 hours in appropriate tray, depending on time of incubation. | |

**INTERPRETATION OF RESULTS:**

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| **Step** | **Action** | | |
| **1** | Remove culture plate after 18-24 hours incubation. | | |
| **2** | Observe plate for denim blue colonies: | | |
| **3** | **IF** | **THEN** | |
| No denim blue colonies seen at 18-24 hours | * Record observations in the LIS * Workup complete * MRSA not isolated | |
| Atypical growth  (i.e. colonies with blue “halos”, colonies not typical denim blue color) | * 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) | |
| **IF** | **THEN** |
| Results from above testing are not consistent with MRSA | * Workup complete * MRSA not isolated |
| Results from above testing are consistent with MRSA | * Workup complete * MRSA isolated |
| Denim Blue colonies seen | * Record observations in the LIS * Perform Staph latex test from DEN: | |
| **IF** | **THEN** |
| Staph latex test NEGATIVE | * MRSA not isolated |
| Staph latex test POSITIVE | * MRSA isolated |

**REPORTING INSTRUCTIONS:**

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| **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) and Stanton Infection Prevention and Control (SIPAC) if ER or In-patient * In order entry add ESO code “MRSA” |

**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.

**REFERENCES:**

* Clinical Microbiology Procedures Handbook, 4th 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, 11th edition, ASM Press, Washington, D.C.
* Oxoid Denim Blue agar package insert, May 2005

**APPROVAL:**

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Date

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**REVISION HISTORY:**

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| **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 |
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