PURPOSE:

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

SAMPLE INFORMATION:

Type	Swab • Amie's with or without charcoal
Source	• Bilateral nasal swab • Bilateral groin swab • Other: drainages, wounds, sites of catheters, tracheostomy and other skin penetrating devices
Stability	If the sample is received in the laboratory and processed greater than 48 h from collection: • Add specimen quality comment "Delayed transport may have compromised the recovery of organism"
Storage Requirements	Room temperature
Criteria for rejection and follow up action	1. Unlabeled/mislabeled swabs 2. Dry swabs

REAGENTS and/or MEDIA:

- Denim Blue Agar and Blood Agar
- Deification reagents: rapid staph

SUPPLIES:

- Wooden applicator sticks
- Disposable inoculation needles
- 35° ambient air incubator
- Biosafety cabinet

SPECIAL SAFETY PRECAUTIONS:

Containment Level 2 facilities, equipment and operational practices for work involving infectious or potentially 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 where 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.

QUALITY CONTROL:

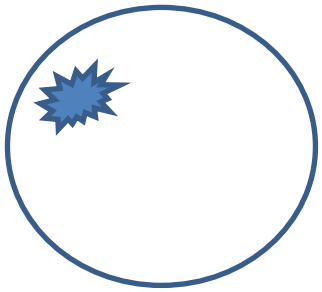
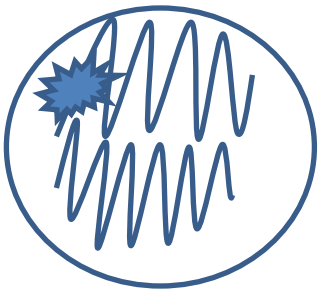
Refer to MIC60100 Non-Exempt Media Quality Control procedure

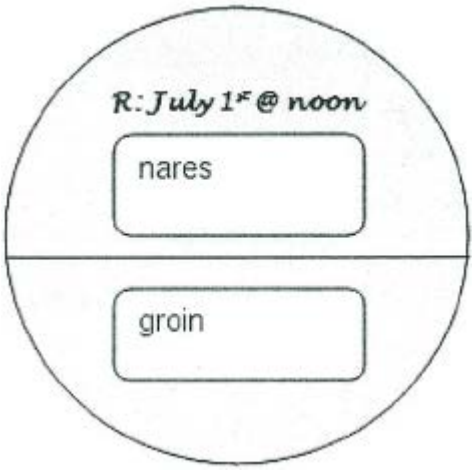
Refer to Quality Control manual for reagent quality control procedures

PROCEDURE INSTRUCTIONS:

Follow the steps in the table below:

*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
Processing specimen for MRSA screening	
1	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. 
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 date 24 hours from day of planting i.e.: July 1st • Time of planting i.e.: noon Reason: Plates are read at approx. 18-24 hours after incubation.	
4	Incubate plate in O_2 incubator at 35° for 18-24 hours in separate batches depending on time of incubation.	

INTERPRETATION OF RESULTS:

Follow the steps in the table below to interpret the MRSA culture:

ACTION
• Remove cultures at 18-24 hours from O_2 incubator • Observe plates for denim blue colonies   

IF	THEN
No growth OR White colonies	• Log results into LIS. • Report "No Methicillin Resistant Staph aureus (MRSA) isolated".
Atypical growth (e.g. colonies with blue "halos", blue colonies not typical denim blue color)	• Should not be interpreted as possible MRSA. • Do further testing to rule out MRSA (i.e. rapid staph/tube coag/GPS/smear/catalase, etc.) • Atypical colonies that identify as *Staphylococcus aureus* need a GPS to confirm oxacillin resistance.
Denim Blue colonies seen	• If sufficient isolated colonies present, perform rapid staph directly off the Denim Blue plate. • If no isolated colonies or in doubt of the color, subculture colonies to BA agar. Perform rapid staph off BA sub plate.
IF	**THEN**
Rapid Staph POSITVE Known history	• Log results into LIS. • Add organism: "*Staphylococcus aureus*". • Growth quantity: "Isolated". • Culture comment: "***Methicillin Resistant***, This organism is cloxacillin resistant and is resistant to all beta-lactam agents". • Go to Order Entry; copy report to Chief Medical Officer of Health (HPU) and Infection Control Nurse (SOHS) if in-patient.

IF	THEN
Rapid Staph POSITVE First time isolated	• Log results into LIS. • Add organism: "*Staphylococcus aureus*". • Growth quantity: "Isolated". • Culture comment: "***Methicillin Resistant***, This organism is cloxacillin resistant and is resistant to all beta-lactam agents" • Go to Order Entry; copy report to the Chief Medical Officer of Health (HPU) and Infection Control Nurse (SOHS) if in-patient • In the patient demographics field click on ESO and add "MRSA Positive"
Rapid Staph NEGATIVE	• Log results into LIS. • Report "No Methicillin Resistant Staph aureus (MRSA) isolated".

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.

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

REVISION HISTORY:

REVISION	DATE	Description of Change	REQUESTED BY
1.0	11 Jan 2017	Initial Release	L. Steven