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Prepared by QA Committee		
Issued by: Laboratory Manager	Revision Date: 9/14/2018	
Approved by Laboratory Director: Microbiologist-in-Chief	Annual Review Date: 5/1/2019	

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APPENDICES

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Appendix I [Analytical Process - Bacteriology Reagents Materials Media List QPCMI10001](#)

Appendix II [How to Set Up & Interpret a MIC Panel](#)

Appendix III [Isolate Notification and Freezing Table QPCMI16003](#)

Appendix IV

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METHICILLIN-RESISTANT *Staphylococcus aureus* (MRSA)

I. Introduction

These specimens are submitted to identify carriers of methicillin-resistant *S. aureus* (MRSA). Swabs may be submitted from any body site, but the most common are nasal, rectal and wound, or the combined nasal/axilla/groin/perineum (NAGP).

II. Specimen Collection and Transport

See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

III. Reagents/ Material/ Media

The OXOID Denim Blue Agar (DBLUE) contains a species-specific chromogen that turns *Staphylococcus aureus* colonies blue. As this chromogen is light sensitive, plates must be stored in their shipping boxes to protect them from unnecessary light exposure until use. After streaking, place directly into plastic bins inside the incubator shielded from light. No more than 4h light exposure by the final read is acceptable.

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)



IV. Procedure

A. Specimen Processing:

- a) Direct Examination: Not indicated
- b) Culture:

Media	Incubation
OXOID Denim Blue Agar (DBLUE)*	O ₂ , 37°C x 24 h -in the dark

*If multiple swabs from a single patient are received individually, then process as separate specimens. If multiple swabs from a single patient are received as a "bundle" with a single label and order number, then process all swabs in the bundle on a single "DBLUE" plate.

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- On Fridays and Saturdays, specimens will not be planted past 3 pm.
 - Any specimens received after this time will be held and planted the following morning. These will be stored in a basket labeled for this purpose in the planting refrigerator.
- On Sunday, specimens will be planted until 5:30 pm

B. Workflow and Culture Interpretations

1. Morning



- i. Check all plates in all bins and remove plates with blue colonies for work up. Separate DBLUE plates growing denim blue colonies (NOT blue hazes or dark blue pinpoint colonies) and replace plates with no blue colonies into their respective bins. Immediately replace cover on each bin to protect *S. aureus*-specific chromogen in the plates from excess light exposure. Return bins to incubator ASAP until final reads at 11 am, 3pm, 6pm and 10pm respectively.
- ii. For each plate with blue colonies, check each patient's MRSA and VRE history. Mark DBLUE and SUBBA with an asterisk if "PREV" MRSA and add "VANCS" if patient had VRE history *within last 3 months*. At media DBLUE, enter the amount of blue colonies present by pressing "Q" for keypad item "QUANT". Pick from the keypad the amount of growth (number of colonies if < or = 5, +/-, 1+, 2+ or 3+).
- iii. Separate "NEW" positive and "PREV" positive plates into different stacks. Order Vitek MS and call labels on all specimens that have isolated blue colonies. Set up Vitek MS on all blue colonies and subculture to BA for any blue colonies from "New" MRSA positives
- iv. Check "New" MRSA worklist for outstanding specimens from the previous day and ask for replant if any are not accounted for.
- v. On "New" positive patients, set up DENKAs on all isolates identified as *S.aureus* by Vitek MS.
- vi. Complete leftover old work from the previous day.
- vii. At 11:00 am, screen plates from the 8-11am bin. Plates with no blue colonies may now be batch finalized as "Negative – No methicillin-resistant *Staphylococcus aureus* (MRSA) isolated".

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i) **For NEW MRSA**

- a) If Vitek MS is negative for *S.aureus*, result as “Negative – No methicillin-resistant Staphylococcus aureus (MRSA) isolated” and status as “Final”.
- b) If MS identified as *S.aureus*, perform DENKA (Denka Seiken PBP2a agglutination test).
- c) If MS identified as *S.aureus* and DENKA+, <CTRL> “P” as “MRSA” and notify IC and ward as per Isolate Notification and Freezing Table QPCMI15003. Set up oxacillin screen (OXA), vancomycin screen (VANCS), Vitek GPAST and KB mupirocin (MUP) disc.
 When complete, interim for review as “MRSA”.
 Set up MUP E-test if MUP zone <19mm. Set up fusidic acid E-test if MRSA is resistant to both SXT and TET.
 Also set up BHIB for PF (MSH), SUBBA for PFGE at PHL (Baycrest), as appropriate and freeze (FRZ).

 If VITEK SXT=R SUPPRESS SXT and confirm result by KB BEFORE reporting. A POP-UP will remind you: “Dsxt=R//uncommon susceptibility result. Suppress and verify w/ KB”
- d) If MS identified as *S.aureus* but DENKA-negative, CTRL “P” as “MRSA presumptive identification, confirmation to follow” and notify IC/ward as per [Isolate Notification and Freezing Table QPCMI16003](#), set up OXA/VANCS/MUP/VT GP- AST and set up KB (from 0.5 McFarland suspension) with cefoxitin disc.
- e) After overnight incubation, record cefoxitin KB result and perform [Induced DENKA](#) from colonies that grew closest to the cefoxitin disc.

If induced DENKA is positive, notify IC/ward of confirmed “MRSA”. Document other test results and FRZ, setting up BHIB (for PFGE) or SUBBA for PGFE at PHL when appropriate and status the test as “Interim” for review.

If induced DENKA is negative, refer to How to [Detect MRSA/BORSA](#) section in the susceptibility manual.

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f) Send [to NML](#) in batches when requested by IC for CNISP surveillance

ii) **For PREVIOUS MRSA (MRSA within prior 3 months)**

- a) If Vitek MS identified as *S. aureus*, or Pastorex Staph-Plus is positive, check patient VRE history. If patient has had any VRE, (within the last 3 months) and there is sufficient growth of blue colonies, set up VANCS. If no positive VRE history, report as “MRSA with quantitation”; assign “Interim” status for review.
- b) If Vitek MS identified as NOT *S. aureus*, suppress ID and finalize as “Negative - No methicillin-resistant Staphylococcus aureus (MRSA) isolated”.

iii) If SUBBA grows **an organism other than staphylococcus**, document organism and supplementary tests performed and finalize as “Negative - No methicillin-resistant Staphylococcus aureus (MRSA) isolated”.

2. Between 2:30 and 3:00pm

- a) Remove the >11am-3pm bin, batch report DBLUE with no blue colonies as “Negative – No methicillin-resistant Staphylococcus aureus (MRSA) isolated”.
- b) For newly visible blue colonies, check each patient’s MRSA and VRE history. Mark DBLUE and SUBBA with an asterisk if “PREV” MRSA, and if sufficient, perform “VANCS” if patient was previously VRE positive. Inoculate SUBBA and incubate in O₂ overnight.
- c) For “New” Positives, subculture to SUBBA and incubate overnight.

3. At 6pm and 10pm



- a) The evening technologist will batch-report DBLUE with no blue colonies as “Negative - No methicillin-resistant Staphylococcus aureus (MRSA) isolated” at 6pm from the >3pm-6pm and at 10pm from the >6-10pm bins, respectively.
- b) For newly visible “Previous” positive or “New” positive, the evening technologist will subculture to SUBBA only.

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V. Reporting

Negative report: “Negative - No methicillin-resistant *Staphylococcus aureus* (MRSA) isolated”

Positive report: “Methicillin-Resistant *Staphylococcus aureus*” with quantitation and appropriate susceptibilities and comments for new cases ([Refer to Susceptibility Testing Manual](#)).

Scant growth (1-5 colonies) - Upon Infection Control request to replant into BHIB (2mL):

- Confirmed by replanting original specimen in broth

Add ISOLATE Comment:

“MRSA confirmed by broth enrichment culture.”

LIS Code: “\MRSc

- NOT confirmed by replanting original specimen in broth:

1. Change original isolate to an alpha isolate

2. Add TEST Comment

“No MRSA isolated by broth enrichment culture. The previous report of “MRSA isolated” was not confirmed by broth enrichment culture suggesting that the previous report reflects contamination or a very low level positive result. Please send another screening swab as clinically indicated.”

LIS Code: “}MRSC”

VI. References



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VANCOMYCIN-RESISTANT ENTEROCOCCI (VRE)

TEMPORARY PROCEDURE CHANGE IN EFFECT: Effective Immediately until further notified

Procedure modification: Please set up Cepheid VRE PCR on **ANY** amount of purple colonies from BVRE plate.

Introduction

These specimens are submitted to identify carriers of vancomycin-resistant *E. faecium* and/or *E. faecalis* (VRE). Swabs may be submitted from any body site (other than nasal and axilla), but most commonly are collected from the rectum.

Specimen Collection and Transport



See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

Specimen Rejection Criteria

Nasal and axilla swabs will not be processed for VRE. Refer to [Reporting](#) in **Section VI** for the appropriate reporting comment.

Reagents/ Material/ Media

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)

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Procedure

A. Processing of Specimen:

Refer to Specimen Processing Procedure MI_SM_PROC

- a) Direct Examination: Not indicated
- b) Culture in non-outbreak setting:

Media	Incubation
Brilliance VRE Agar (BVRE)	O ₂ , 37°C x 36hrs in the dark

B. Culture for **VRE PCR positive samples** in outbreak setting:



Media	Incubation time (all O ₂ at 37°C)
i) Place 500uL (0.5 mL mark of transfer pipette) of the eSwab transport medium into: - 2 mL Brain Heart Infusion broth (BHIB) Place 30uL (1 drop from transfer pipette) of the eSwab transport medium onto: - Brilliance VRE Agar (BVRE)	overnight on shaker 36h in the dark
ii) If BVRE is no growth after overnight incubation, subculture 1 drop from BHIB to: - Brilliance VRE Agar (BVRE)	36h in the dark

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C. Workflow and Interpretation of cultures:

Workflow is described in the Bench Workflow Manual.

Process specimens as per WASPLab Screening, Reading and Picking Manual.

For specimens processed offline:

- a) Label new bin for Planting incubator
- b) Read BVRE plates planted from the previous day, separating plates growing purple or blue colonies. Read 36 hrs. plates separating plates growing purple or blue colonies.

VRE cultures will be read at 18hrs, 30hrs and a final reading at 36hrs for VRE *faecium* & *faecalis*

Colonies on Brilliance VRE Agar:

Isolate:	Colony colour:
<i>Enterococcus faecium</i>	Purple to Royal Blue colour on entire colony, moist
<i>Enterococcus faecalis</i>	Denim Blue
CNST	Blue (if grown)
Yeast	Light blue (if grown)
<i>Enterococcus gallinarum</i>	Blue (if grown)
Lactobacilli	Light blue/pink (if grown)

A. Royal Blue and Purple colonies:

Check history of patient who's specimen's are growing purple colonies.

- a) If patient is a **“New” positive with ≥ 5 purple colonies**; perform PCR and Vitek MS
 - a) Inoculate a spot on Vitek MS slide for ID
 - b) Pick purple colonies and emulsify them in 0.5 mL saline
 - c) Using the same swab, inoculate a vial of PCR sample reagent and set up Cepheid PCR
 - d) Using the 0.5mL emulsified saline, inoculate a SUBBA and ¼ BVRE (SBVRE).

Note: For isolates with unsuccessful ID results, report as Vancomycin-Resistant E.faecium, confirmation to follow (with vanA/B positive result)



- b) If patient is **“New” positive with < 5 colonies**
 - Pick colony(ies) and emulsify into 0.5 mL saline
 - Using the same swab, inoculate a SUBBA and ¼ BVRE

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c) If patient is a “**Previous**” **positive** (≤ 3 months)

- Set up Vitek MS, VANCS, PP,

Follow : Table 1 [VRE Workup Guide –PURPLE COLONIES](#) for further work up.

B. Samples growing Denim/Light Blue colonies:

Observe quantity of suspect colony growth.

a) **Scant growth:** inoculate colonies into 0.5mL saline and onto ¼ BVRE (SBVRE)

b) **Moderate/Heavy growth:**

- Inoculate a spot on Vitek MS for ID.
- If *enterococcus faecium* or *faecalis* (or gpc chains when Vitek MS fails) emulsify colonies in 0.5 mL saline and use that swab to inoculate a vial of PCR sample reagent and set up Cepheid PCR.
- Using the 0.5mL emulsified saline, inoculate a SUBBA and ¼ BVRE (SBVRE).

Note: For isolates with unsuccessful ID results, report as Vancomycin-Resistant E.faecium, confirmation to follow (with vanA/B positive result)

Follow : Table 2 [VRE Workup Guide – BLUE](#) for further work up.

C. No Royal Blue and Purple or Denim/Light Blue colonies:

Re-incubate negative plates for further incubation as needed.



Enter “__hr: No purple or blue” and status as “Prelim”.

Finalize 36 hr culture as negative. (See VRE reporting section)

Read and report old work. Communicate to ward and/or infection control if necessary as per [Isolate Notification and Freezing Table QPCMI16003](#)

Continue to scan BVRE plates and process any that are now growing purple or blue colonies.

Check new VRE worklist after all plates are prelimmed for any missing plates. Document if plate is not found and ask for replant.

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VRE Identification:

Rule out VRE as below:

Table 1 VRE Workup Guide –PURPLE COLONIES



NEW Purple/Royal Blue Colonies		PREVIOUS + (<3months) Purple cols
BVRE >5cols 24/48 hours	BVRE – scant, <5 cols	BVRE (any amount)
1. Set up vanA/vanB Cepheid PCR and MS and SBVRE and SUBBA 2. Cepheid – Positive <ul style="list-style-type: none"> • Report according to ID as Entfar or Entfer with comment <ul style="list-style-type: none"> vanA gene positive OR vanB gene positive ▪ If Cepheid vanB Positive, Roche PCR must be done for MSH patients only (within 24 hours) • Notify ICP/ward • Set up Etest, VANCS <ul style="list-style-type: none"> ▪ Vanco Etest $\geq 8\mu\text{g/mL}$, VANCS-growth, SBVRE-growth, <ul style="list-style-type: none"> ○ Report Entfar or Entfer with phenotype comment ▪ Vanco Etest $\leq 4\mu\text{g/mL}$, VANCS - NG, SBVRE - NG, <ul style="list-style-type: none"> ○ Perform PCR again from etest plate to confirm presence of vanA ○ Report as vanco sensitive entvaa or entfva ○ Report with comment: “Vanco susceptible phenotype” • PFGE (MSH only) & FRZ 	1. Set up SBVRE and SUBBA 2. SBVRE - NG <ul style="list-style-type: none"> • Report as No VRE 3. SBVRE - Any growth <ul style="list-style-type: none"> • Set up Cepheid PCR & MS <ul style="list-style-type: none"> ▪ Cepheid Positive: proceed as #2 (BVRE >5 cols). ▪ Cepheid Negative: proceed as #3 BVRE >5 cols. (growth on SBVRE) 	1. Set up MS and VANCS 1. VANCS – Growth <ul style="list-style-type: none"> • Report Entfar or Entfer with comment 2. VANCS – NG <ul style="list-style-type: none"> • If “Previous” entfar or entfer: report as NO VRE • If “Previous” entvaa, perform Cepheid: <ul style="list-style-type: none"> ▪ Cepheid vanA Positive: report as entvaa ▪ Cepheid Negative: report as NO VRE *Set up etests for Vanco and Teico every 3 months from original isolate to confirm phenotype

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NEW Purple/Royal Blue Colonies		PREVIOUS + (<3months) Purple cols
BVRE >5cols 24/48 hours	BVRE – scant, <5 cols	BVRE (any amount)
3. Cepheid – Negative <ul style="list-style-type: none"> • SBVRE - NG <ul style="list-style-type: none"> ▪ Report as No VRE • SBVRE - GROWTH <ul style="list-style-type: none"> ▪ Set up Vanco/Teico Etests, VANCS ▪ Vanco Etest $\geq 8\mu\text{g/mL}$, VANCS - growth, <ul style="list-style-type: none"> ○ Add comment non vanA/B to isolate ○ Send for van genotyping & FRZ ○ Notify ICP 		



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Table 2 VRE Workup Guide – BLUE COLONIES

Blue Colonies (SCANT /LIGHT growth)			Blue Colonies (HEAVY)
Set up SBVRE on any amount of blue cols growing			Vitek MS ID of <i>Enterococcus faecium</i> or <i>Enterococcus faecalis</i> .
NG on SBVRE	Scant Growth on SBVRE	Mod-Heavy Growth on SBVRE	
Report – No VRE	Set up VANCS ‘PP’	Set up Cepheid PCR & MS	Set up Cepheid PCR , SBVRE and SUBBA
	1. VANCS – No growth <ul style="list-style-type: none"> • Report No VRE 2. VANCS - Growth <ul style="list-style-type: none"> • Set up MS • Perform Cepheid PCR, Etests. <ul style="list-style-type: none"> ▪ Proceed as Mod-Heavy Growth • Notify ICP/ward 	1. Cepheid – Positive <ul style="list-style-type: none"> • Follow NEW Purple >5 Cepheid positive workflow. 2. Cepheid – Negative <ul style="list-style-type: none"> • Set up Etests & VANCS • If Vanco Etest $\geq 8\mu\text{g/mL}$, VANCS - growth, add comment: ‘non vanA/B’ to isolate • Send to NML for van genotyping & FRZ • Notify ICP 	1. Cepheid – Positive <ul style="list-style-type: none"> • Follow NEW Purple >5 Cepheid positive workflow. 2. Cepheid – Negative <ul style="list-style-type: none"> • Set up Etests & VANCS • If Vanco Etest $\geq 8\mu\text{g/mL}$, VANCS - growth, add comment: ‘non vanA/B’ to isolate • Send to NML for van genotyping & FRZ • Notify ICP





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Table 3 VRE Workup Guide – Cepheid PCR + from E- swab directly

1. Phone/email ward and ICP as per Isolate Notification and Freezing table QPCMI15003.
2. For new or previous VRE patients where NO isolate has been isolated yet proceed as below:

Subculture to BHIB broth and BVRE and incubate overnight		
If BVRE is No growth, Subculture BHIB to BVRE		
NG	Scant Growth (purple or blue colonies)	Mod-Heavy Growth (purple or blue colonies)
Report as “No VRE isolated after broth enhancement”	1. Sub to SUBBA and SBVRE <ul style="list-style-type: none"> • Set up MS and Vanco/Teico etest, VANCS • Proceed as Mod-Heavy Growth 	1. Set up MS and Vanco/Teico etest, VANCS <i>Do not set up Cepheid PCR from BVRE plate</i> <ul style="list-style-type: none"> • If Vanco R/Teico R report as vanA phenotype with comment • If Vanco R/Teico S report as vanB phenotype with comment • If Vanco S/Teico S, do Cepheid from Etest plate <ul style="list-style-type: none"> ▪ Cepheid Negative: report as No VRE after broth enhancement ▪ Cepheid Positive, vanA Positive: report as Entvaa with comment <u>!vaAi</u>

[Vancomycin and teicoplanin for Enterococcus phenotype](#)

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VRE PFGE:

If patient is from MSH only, subculture isolate to Brain Heart Infusion Broth for pulsed-field (PFGE).

VI. Reporting

Negative Report: “Negative - No Vancomycin-Resistant Enterococci (VRE) isolated”

Positive Report:

New Positive VRE Patients

Day 1

- **PCR direct from BVRE plate - with isolate**

ISOLATE: “*Enterococcus (faecium or faecalis)*-vancomycin resistant” “isolated”

ISOLATE COMMENT:

“This organism is positive for the vanAorB gene as tested by the Cepheid vanA/B GenXpert Assay (for research only).

~Phenotypic confirmation to follow.” Isolate Comment Code \vaAg or \vaBg

- **PCR direct from BVRE plate - no isolate, from sweep**

ADD ISOLATE COMMENT:

“PCR from a sweep of growth on the plate is positive for the vanA gene by the Cepheid vanA/B GenXpert Assay (for research use only) but a distinct vancomycin-resistant or vancomycin-susceptible *Enterococcus* species that is vanA positive cannot be found.”

Isolate Comment Code \vaAp

Day 2

- Vancomycin=R, Teicoplanin=R:

“*Enterococcus faecium or faecalis* -vancomycin resistant” “isolated”

ISOLATE COMMENT (Code \vaA):

“This organism has a vanA phenotype.”

- Vancomycin=R, Teicoplanin=S:

“*Enterococcus faecium (or faecalis)* -vancomycin resistant”



ISOLATE COMMENT (Code \vaB):

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“This organism has a vanB phenotype.”

Previous VRE Positive Patients:

Enterococcus (faecium or faecalis)—vancomycin-resistant isolated.

ISOLATE COMMENT (Code: \vapr):

“The Cepheid vanA/B GenXpert Assay was not completed as this patient has had VRE isolated within the past 3 months that has had molecular characterization.”

Vancomycin=S, vanA gene-positive VRE

- Isolate from IC VRE Culture Screen

- 1) Change previous isolate code of entfar to **entvaa** - “*Enterococcus faecium* - vanA gene positive” “isolated”

ISOLATE COMMENT (Code: \vaAi) – “This organism is positive for vanA gene by the Cepheid vanA/B GenXpert Assay (for research use only) but has a vancomycin susceptible phenotype.

The effectiveness of vancomycin in this setting is uncertain and is not recommended. Please contact Infectious Diseases or Medical Microbiology for treatment advice.”

Remove previous duplicated ISOLATE COMMENT.

- 2) Change previous isolate code of entfer to **entfva** - “*Enterococcus faecalis* - vanA gene positive” “isolated”

Vancomycin MIC =>8 by macro Etest, vanA/B-negative by PCR

“*Enterococcus faecium or faecalis*” “isolated”

ISOLATE COMMENT (Code: \vanI):



“This organism has reduced susceptibility to vancomycin but is negative for *vanA* and *vanB* genes as tested by the Cepheid vanA/B GenXpert Assay (for research use only).

~This organism has been sent to the National Microbiology

~Laboratory for further testing and results will be

~reported when available.”



Confirmation from NML:

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Negative – Add the following statement as an ‘Updated Report’: “The previously reported organism has no vancomycin resistance genes as tested by the National Microbiology Laboratory, Winnipeg, Specimen No. xxxx”

Positive – *Enterococcus faecalis or faecium* - vancomycin-resistant “isolated”
ISOLATE COMMENT (Code: vanE):

“This organism is positive for the *vanE* gene as reported by the National Microbiology Laboratory... NML Specimen No. xxx”

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RESISTANT GRAM NEGATIVE BACILLI

I. Introduction

These specimens may be submitted to identify carriage of drug-resistant Gram negative bacilli, to determine cross-transmission between patients or to identify an environmental source of patient infection.

II. Specimen Collection and Transport

Any specimen may be submitted, although rectal swabs and environmental are the most common. Swabs should be transported in an Eswab or Amies transport medium. If a delay in transport or processing is anticipated, the swabs should be kept at 4°C.

III. Reagents/Materials/Media

See [Analytical Process - Bacteriology Reagents Materials Media List QPCMI10001](#)

IV. Procedure

A. Processing of Specimen:

- a) Direct Examination: Not indicated
- b) Culture:

Media	Incubation
-------	------------

For *Enterobacteriaceae* with fluoroquinolone and/or aminoglycoside resistance but susceptibility to cefpodoxime:

MacConkey Agar (Mac) –no antibiotic	O ₂ , 35°C x 18 h
-------------------------------------	------------------------------

For *Serratia marcescens* outbreaks,

CTCZ – with colistin	O ₂ , 35°C x 18 h
----------------------	------------------------------

B. Interpretation of cultures:



1. Read cultures plates after 18 to 24 hours of incubation.

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2. Workup requested organism as per [Bacteria Workup Manual](#)
3. Set up susceptibility as per [Susceptibility Manual](#)
4. Communicate with requesting Infection Control Practitioner or Microbiologist as appropriate and freeze all positive isolates unless otherwise directed. PFGE will only be performed on request from Infection Control.

For *Serratia* Screen:

1. Read culture plates after 18 to 24 hours of incubation.
2. For *Serratia marcescens*, work-up NLF, LLF or orange-red pigmented colonies only. Perform Vitek MS.
 - Phone ward and email ICP if *Serratia marcescens* is isolated.
3. Set up susceptibility as per [Susceptibility Manual](#).
4. Previously positive *Serratia marcescens* specimens only require a meropenem screen to be set up.
5. If *Serratia* is isolated, freeze and set up BHIB for PFGE.

N.B. Susceptibilities can be referred for 3 months

V. Reporting

Negative report: “No <requested organism> isolated”

Positive report: “<requested organism> isolated”
 Report their susceptibility results as per Susceptibility Manual.
 Add Isolate comment: “Susceptibility testing results are provided for infection control purposes only.” \ICSN

VI. References



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ESBL and Carbapenemase SCREEN

I. Introduction

These specimens are submitted to identify *Klebsiella* species, *Escherichia coli* and *Proteus mirabilis* with acquired extended spectrum β -lactamases as well as carbapenemases from any *Enterobacteriaceae*.

ESBL testing is only performed on specimens from pregnant patients, specimens originating from mothers and baby units or upon special request.

II. Specimen Collection and Transport

See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

III. Reagents/Materials/Media

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)

IV. Procedure

A. Processing of Specimen:

- a) Direct Examination: Not indicated
- b) Culture:

Media	Incubation
ESBL Isolation Agar with 2 μ g/ml cefpodoxime	O ₂ , 37°C x 18-24 hours

B. Interpretation of cultures:



1. Examine plates after 18-24 hours of incubation for any growth of an *Enterobacteriaceae*.
2. If no *Enterobacteriaceae* are isolated, report as “Negative - no ESBL or Carbapenemase producing organisms isolated.”

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3. For all LF and oxidase negative NLF *Enterobacteriaceae* colony types, set up Vitek MS for identification.
4. Should an isolate ID as an *E.coli*, *Klebsiella spp.*, or *P.mirabilis*, check patient history.
 - For a patient with no prior history or with “Previous” positive (>3months) history of *E.coli*, *Klebsiella spp.*, or *P.mirabilis* in an IC sample set up ‘KB IC ESBL’.
 - If a previous positive ESBL was isolated within the last 3 months, set up **Meropenem Screen**, only by disk diffusion. Refer to the previous sample’s date that susceptibilities were reported. Report isolate with phrase

“Phenotypic screening suggests this organism is ESBL POSITIVE as previously confirmed on “yyyy.mm.dd”. LIS isolate comment code \ESBP

Report with Test Comment:

“Negative Carbapenemase screen - No carbapenemase producing organisms isolate. POSITIVE for ESBL screen”.

5. For all other *Enterobacteriaceae* set up **Meropenem Screen** only.
6. For CRE work up, refer to [CRE Flowchart and Procedure](#)

V. **Reporting**

When **ESBL screen** is requested, report both ESBL and Carbapenemase comments where applicable.

Negative report for both ESBL and carbapenemase:

“Negative - No extended-spectrum-beta-lactamase producing (ESBL) or carbapenemase-producing organism isolated”

Positive reports:

Positive for both ESBL and Carbapenemase:

At TEST Window:

POSITIVE for ESBL screen
 POSITIVE Carbapenemase Screen



At ISOLATE Window:

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“*Escherichia coli*” or “*Klebsiella species*” or “*Proteus mirabilis*” isolated **with** one of the following ISOLATE COMMENT:

“The susceptibility pattern suggests that this organism contains a class A extended spectrum beta-lactamase (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains class A and C extended spectrum beta-lactamases (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains a class C extended spectrum beta-lactamase (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains an inducible class C extended spectrum beta-lactamase (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains an extended spectrum beta-lactamase (ESBL) other than class A or C.”

AND

From keypad: ESBL: \ICSN “Susceptibility testing results are provided for infection control purposes only.”

AND

Final Positive CRE Result by CARB-R PCR:

“_____ carbapenemase gene DETECTED by Cepheid Xpert CARBA-R Assay (for research use only). This assay is able to detect NDM, KPC, OXA48, OXA181, OXA232, IMP-1, and VIM carbapenemase genes.” Isolate Comment Code: \CPC+

OR

Preliminary CRE Result:

Isolate Comment: [\CNML](#)



AND

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Send updated, Final Result once NML report is available

Negative report:

- a. **Suppress the isolate**
- b. Add the following comment in the TEST window for **NOT CONFIRMED** carbapenemase:
Add TEST COMMENT code [\KPCN](#)
- c. Enter the NML results to the LIS ISOLATE Breakpoint panel **kpercon**.
- d. E-mail or call Infection Control Practitioner and ward as per [Isolate Notification Table](#).

Positive report:



- a. “Updated Report”
- b. Add the following isolate comment for **CONFIRMED** carbapenemase:
- c. Add ISOLATE COMMENT code [\KPCP](#)
- d. Enter the NML results to the LIS ISOLATE Breakpoint panel **kpercon**.
- e. E-mail or call Infection Control Practitioner and ward as per [Isolate Notification Table](#).

Negative report for carbapenemase but POSITIVE for ESBL:

At TEST Comment: “Negative Carbapenemase Screen - No carbapenemase-producing organism isolated”
POSITIVE ESBL Screen”

At ISOLATE Window:

“*Escherichia coli*” or “*Klebsiella species*” or “*Proteus mirabilis*” “isolated” **with** ISOLATE COMMENT: “The susceptibility pattern suggests that this organism contains a class A extended spectrum beta-lactamase (ESBL).” **OR** “The susceptibility pattern suggests that this organism contains class A and C extended spectrum beta-lactamases (ESBL).” **OR** “The susceptibility pattern suggests that this organism contains a class C extended spectrum beta-lactamase (ESBL).” **OR** “The susceptibility pattern suggests that this organism contains an inducible class C extended spectrum beta-lactamase (ESBL).” **OR** “The susceptibility pattern

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suggests that this organism contains an extended spectrum beta-lactamase (ESBL) other than class A or C.”

Report appropriate sensitivity results as per [Susceptibility Manual](#)

Previous ESBL Positive Patient:

Negative report for carbapenemase but POSITIVE for ESBL:

At TEST Comment: “Negative Carbapenemase Screen - No carbapenemase-producing organism isolated”
 POSITIVE ESBL Screen”

At ISOLATE Window:

“*Escherichia coli*” or “*Klebsiella species*” or “*Proteus mirabilis*” “isolated” **with ISOLATE COMMENT:** “Phenotypic screening suggests this organism is ESBL POSITIVE

as previously confirmed on “yyyy.mm.dd”.”

LIS isolate comment code: \ESBP

Negative report for ESBL but POSITIVE for carbapenemase:

At TEST Comment: “Negative ESBL Screen- No extended spectrum beta-lactamase - producing organism (ESBL) isolated”
 POSITIVE Carbapenemase Screen”

At ISOLATE Window:

Report isolate comment as per [Reporting Section](#)



Previous Carbapenemase Positive Patient:

At TEST Comment: “Negative ESBL Screen- No extended spectrum beta-lactamase – producing organism (ESBL) isolated POSITIVE Carbapenemase Screen”

OR

“POSITIVE ESBL Screen and POSITIVE Carbapenemase Screen”



At ISOLATE Window: Report isolate along with Isolate Comment:

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“Phenotypic testing suggests this organism is carbapenemase POSITIVE as previously confirmed on “yyyy.mm.dd”.”
Isolate Comment code \CREP

VI. Reference

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Carbapenemase (CRE) SCREEN (without ESBL Screen)

I. Introduction

These specimens are submitted to identify carbapenemases from any *Enterobacteriaceae*.

II. Specimen Collection and Transport

See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

III. Reagents/Materials/Media

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)

IV. Procedure

A. Processing of Specimen:

- a) Direct Examination: Not indicated
- b) Culture:

Media	Incubation
ESBL Isolation Agar with 2 µg/ml cefpodoxime	O ₂ , 37°C x 18-24 hours

B. Interpretation of cultures:

See [IC Carbapenemase Testing Flowchart](#)



1. Examine plate after 18-24 hours of incubation for any growth of an *Enterobacteriaceae*.
2. If no *Enterobacteriaceae* are isolated, report as negative for CRE.
3. For all *Enterobacteriaceae* colony types, set up a meropenem screen disk diffusion test.
4. If isolates >25mm (susceptible) by “MEMS” disk diffusion, report as negative for CRE.
5. For all Meropenem Screen R (<25mm) by disk diffusion, Set up Vitek MS
 - If the isolate is not identified as *Enterobacteriaceae*, report as negative for CRE.

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- If the isolate is identified as *Enterobacteriaceae*, set up β CARBA (BCARB)
 - a) **If β CARBA (BCARB) is negative:**
 - i. Report isolate with the following TEST COMMENT: \NCRB
 - ii. Phone or e-mail IC and ward as [per Isolate Notification Table](#).
 - iii. Set up Rosco with Temocillin (breakpoint panel kpcros)
 - If **Temocillin = S** and Rosco disks show no potentiation, send out report as NO CRE.
 - **Suppress the isolate**
 - Send Updated Report
 - Add TEST Comment for **NOT CONFIRMED** carbapenemase: [\NCRE](#)
 - E-mail or call Infection Control Practitioner and ward as per [Isolate Notification Table](#)..
 - If **Temocillin = R** OR Rosco shows potentiation to MRBO or MRDP,
 - report isolate with the following ISOLATE Comment: [\CNML](#)
 - Phone or e-mail IC and ward as per [Isolate Notification Table](#). Send isolates to NML ASAP (Cannot send on Friday)
 - Order the LIS ISOLATE Breakpoint panel **kpcrcon**.
 - Freeze isolate
 - b) **If β CARBA (BCARB) is positive:**
 - **Previous CRE positive (\leq 6 months)**
 - i. At TEST Comment: “POSITIVE Carbapenemase Screen”
At ISOLATE Window: Report isolate along with Isolate Comment: [\CREP](#)
 - Phone or e-mail IC and ward as per [Isolate Notification Table](#).
 - **New ?positive CRE**
Notify ICP and send out isolate with comment [\PCRB](#)

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- i. Set up Cepheid CARBA-R PCR (CARBR)
 - If Cepheid CARBA-R PCR (CARBR) is negative
 - Report isolate with the following ISOLATE Comment:
[\pCRB](#)
 - Phone or e-mail IC and ward as per [Isolate Notification Table](#).
 - Send isolates of to NML ASAP (Cannot send on Friday)
 - Order the LIS ISOLATE Breakpoint panel **kpcrcon**
 - Set up Rosco with Temocillin (panel kpcros). For epidemiology purposes only. Record and suppress kpcros results.
 - If Cepheid CARBA-R PCR (CARBR) is positive
 - Report gene identified by Cepheid using ISOLATE Comment:
[\CPC+](#)
 - Phone or e-mail IC and ward as per [Isolate Notification Table](#).
 - Send to NML and PHOL in batches when requested for and .
 - Freeze isolate

V. Reporting

See [Carbapenemase Testing Reporting](#).

VI. Reference

1. **Clinical and Laboratory Standards Institute** 2016 Performance Standards for Antimicrobial Susceptibility Testing; Documents M100-S26, M2-A12, M7-A10 CLSI, Wayne, PA.
2. **QMP-LS Bacteriology Consensus Practice Recommendations** – Antimicrobial Susceptibility Reporting Toronto ON: QMP-LS QView. c2011

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RESISTANT *Pseudomonas aeruginosa* SCREEN

I. Introduction

Specimens are submitted for the screening of multi-drug resistant *Pseudomonas aeruginosa*.

II. Specimen Collection and Transport

See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

III. Reagents/Materials/Media

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)

IV. Procedure

1. Processing of Specimen:



Specimen	Processing	Media	Incubation
Water	Centrifuge the entire sample at 3500 rpm for 20 minutes. Pour off all supernatant. Transfer the contents of a 2 mL tube of BHI broth into in the falcon tube containing the sediment Subculture BHI broth after overnight incubation to MCPOD by the IC Bench technologist	BHI Broth MCPOD	O ₂ at 35°C overnight O ₂ at 35°C for 24 hours
Environmental swabs	Incubate the BHI Broth Subculture BHI broth after overnight incubation to MCPOD by the IC Bench tech using a new sterile swab	MCPOD	O ₂ at 35°C overnight O ₂ at 35°C for 24 hours
Patient pharmaceutical	≤1 mL	TH14 SD14	O ₂ at 35°C for 14 days O ₂ at RT ^o for 4 days

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Specimen	Processing	Media	Incubation
infusates/injectables (QC bench)	>1 mL	ETH14 ESD14	O ₂ at 35°C for 14 days O ₂ at RT ^o for 4 days
Swabs from patients	Directly inoculate MCPOD plate with specimen	MCPOD	O ₂ at 35°C for 24 hours

2. Interpretation of Cultures:

For water, environmental swabs, patient swabs:

Work up these cultures on the IC Bench.

Work up oxidase-positive gram negative bacilli ONLY.

Set up Vitek MS

When identified as *P. aeruginosa* set up Vitek susceptibility card.

For patient samples, if resistant to all antimicrobials from the vitek card, set up colistin etest.

Freeze resistant strains of *Pseudomonas aeruginosa* into IGR boxes.

For Patient pharmaceutical infusates/injectables:

Work up these cultures on the QC/Sterility Bench.

Work up any growth as per Sterility Manual.

V. Reporting

For water, environmental swabs, patient swabs:

Negative Report: No resistant *Pseudomonas aeruginosa* isolated.

Positive (Resistant strains only) Report: *Pseudomonas aeruginosa* with susceptibility result.

Add Isolate comment: “Susceptibility testing results are provided for infection control purposes only.” \ICSN

Email / Call ICP.

For Patient pharmaceutical infusates/injectables:



Negative Report: No growth.

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Positive: Report *Pseudomonas aeruginosa* with susceptibility result. Call ICP
Add Isolate comment: “Susceptibility testing results are provided for infection control purposes only.” \ICSN

VI. References

Clinical and Laboratory Standards Institute 2016 Performance Standards for Antimicrobial Susceptibility Testing; Documents M100-S26, M2-A12, M7-A10 CLSI, Wayne, PA.

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GROUP A STREPTOCOCCUS SCREEN

I. Introduction

Throat, rectal or wound swabs are the most common that are submitted for the diagnosis of Group A streptococcal infection, to determine cross-transmission between patients or to identify an environmental source of patient infection.

II. Specimen Collection and Transport

See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

III. Reagents / Materials/ Media

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)

IV. Procedure

A. Processing of Specimens

See [Specimen Processing Procedure](#)

- a) Direct Examination: Not routinely performed.
- b) Culture:

Media	Incubation
CNA (rectal/wound)	AnO ₂ , 35°C x 18-24 hours
Carrot Broth	O ₂ , 35°C x 18-24 hours
BA (for throat)	AnO ₂ , 35°C x 18-24 hours

If original plates are negative;

Subculture the Carrot Broth to a second CNA plate and incubate overnight in AnO₂, 35°C x 18-24 hours

B. Interpretation of Cultures:



- a) Examine the CNA/ BA plate after 18-24 hours incubation and identify all morphologically distinct beta haemolytic colonies by performing:
 - i) Catalase test
 - ii) Strep grouping

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- b) For all specimens processed after 1600 hours, re-incubate CNA/BA anaerobically for a further 24 hours.
- c) Subculture the Carrot broth to a second CNA/BA plate and incubate overnight in anaerobic conditions
- d) Examine the subculture CNA/BA plate after overnight incubation for distinct beta haemolytic colonies.
- e) Perform catalase and strep grouping if any beta haemolytic colonies appear.
- f) Freeze all isolates and prepare for PFGE (whether in house or to be sent to PHL)
- g) No Susceptibility Testing Required
- h) E-mail or call Infection Control Practitioner and ward as per [Isolate Notification Table](#).

V. Reporting

A. Culture:



Negative report: “No Group A streptococcus isolated”.

Positive report: Report as isolate - “Group A streptococcus” with LIS ISOLATE COMMENT: “isolated”

E-mail or call all positive Group A streptococci isolates to ward / Infection Control Practitioners as per Isolate Notification Table.

VI. References

Clinical and Laboratory Standards Institute 2016 Performance Standards for Antimicrobial Susceptibility Testing; Documents M100-S26, M2-A12, M7-A10 CLSI, Wayne, PA

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KLEBSIELLA OXYTOCA OR KLEBSIELLA PNEUMONIAE SCREEN

I. Introduction

These specimens may be submitted to identify carriage of drug resistant ESBL producing *Klebsiella oxytoca* or *Klebsiella pneumoniae*, to determine cross-transmission between patients or to identify an environmental source of patient infection.

II. Specimen Collection and Transport

See [Pre-analytical Procedure - Specimen Collection QPCMI02001](#)

III. Reagents/Materials/Media

See [Analytical Process - Bacteriology Reagents_Materials_Media List QPCMI10001](#)

IV. Procedure

A. Processing of Specimen:



- a) Direct Examination: Not indicated
- b) Culture:

For Cefpodoxime resistant *Klebsiella oxytoca* or *Klebsiella pneumonia*

Media	Incubation
ESBL Isolation agar with 2ug/mL cefpodoxime	O2, 350C x 18 h

B. Interpretation of cultures:

1. Read cultures plates after 18 to 24 hours of incubation.
2. Perform id on lactose fermenting gram negative bacilli using Vitek MS.
3. Set up Double Disk Kirby Bauer (KBESBLR) on those isolates identified as *Klebsiella oxytoca* or *Klebsiella pneumonia* to rule out ESBL producers.
4. Record susceptibility results and freeze organism

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- Communicate with requesting Infection Control Practitioner or Microbiologist as appropriate and freeze all positive isolates. PFGE will only be performed on request from Infection Control.

N.B. Susceptibilities can be referred for 3 months

V. Reporting

Negative report: “No <requested organism> isolated”

Positive report: “<requested organism> isolated”

Report their susceptibility results as per Susceptibility Manual

with one of the following ISOLATE COMMENT if applicable:

“The susceptibility pattern suggests that this organism contains a class A extended spectrum beta-lactamase (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains class A and C extended spectrum beta-lactamases (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains a class C extended spectrum beta-lactamase (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains an inducible class C extended spectrum beta-lactamase (ESBL).”

OR

“The susceptibility pattern suggests that this organism contains an extended spectrum beta-lactamase (ESBL) other than class A or C.”

AND

“Susceptibility testing results are provided for infection control purposes only.”

VI. References



Clinical and Laboratory Standards Institute 2016 Performance Standards for Antimicrobial Susceptibility Testing; Documents M100-S26, M2-A12, M7-A10 CLSI, Wayne, PA

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APPENDIX II HOW TO SET UP AND INTERPRET A MIC PANEL

I. Materials

MIC panel
 Panel inoculator set
 Sterile distilled water
 Sterile transfer pipettes
 Blood agar plate
 Sealable bag

II. Procedure



1. Remove the desired MIC panel from the -70°C freezer. Place a cover over the panel and place into the O_2 incubator to thaw.
2. When thawed, label the panel and a blood agar plate with the LIS order number.
3. Make a suspension of the organism in saline to match a 0.5 McFarland standard.
4. Place 1.5 mL of organism into a 50mL tube. Add sterile distilled water to reach 40mL in same falcon tube (~38.5mL). Pour into the inoculator base. Gently mix by agitating slowly
5. Place the inoculator into the base making sure there are no bubbles and that all prongs are in contact with the bacterial suspension.
6. Align the left side (lettered) of the panel with the left side (lettered) of the inoculator.
7. Lift the inoculator straight up and place it, prong side down, into the wells of the MIC panel.
8. Using a transfer pipette, transfer 1 drop of suspension from within the inoculator base to a blood agar plate and streak for isolated colonies.
9. Pour the suspension into a sharps container containing hypochloride and discard the inoculator into a sharps disposal box.

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10. Place a lid onto the panel and place into a sealable bag. Seal the bag and incubate the panel in the appropriate atmosphere and temperature (See below).

<u>Panel</u>	<u>Temp.</u>	<u>Atmosphere</u>	<u>Incubation</u>
VRE (vancomycin)	35 ⁰ C	O ₂	24 h
MRSA (oxacillin)	35 ⁰ C	O ₂	24 h
GNB	35 ⁰ C	O ₂	24 h

III. Interpretation

Use a coordinating MIC panel sheet to record wells with any growth. Each panel contains a positive growth control well (no antibiotic) and a negative growth control well (no inoculum). The MIC for each drug is the lowest dilution showing no growth. Record results in the LIS.

Interpretation of MIC results is performed in accordance with NCCLS breakpoint criteria found in the Performance Standards for Antimicrobial Susceptibility Testing Informational supplement M100-S**. This informational supplement is updated annually and breakpoint criteria for all antibiotics used should be checked yearly for changes.

MIC breakpoints for antimicrobial agents tested in MIC panels that do not have NCCLS criteria available should be obtained from the literature (see references for agents such as mupirocin and fusidic acid). When breakpoints are not available in the literature, no interpretation of MIC should be reported.

IV. References



1. **Clinical Laboratory Standards Institute** 2016. Performance Standards for Antimicrobial Susceptibility Testing; 26th ed. CLSI Approved Standard M100S, Clinical and Laboratory Standards Institute, Wayne, PA
2. **Clinical Laboratory Standards Institute** 2015. Methods for Dilution antimicrobial susceptibility tests for bacteria that grow aerobically 10th ed. CLSI Approved Standard M7A, Clinical and Laboratory Standards Institute Wayne, PA

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3. **Fuchs P. C., R. N. Jones, A. L. Barry** Interpretive Criteria for Disk Diffusion Susceptibility Testing of Mupirocin, a Topical Antibiotic *J Clin Microbiol* 1990; 28: 608-609
4. **Skov R., N., Frimodt-Moller, F. Espersen** Correlation of MIC methods and tentative interpretive criteria for disk diffusion susceptibility testing using NCCLS methodology for fusidic acid *Diag Microbiol Infect Dis* 2001; 40: 111-116

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Record of Edited Revisions

Manual Section Name: Infection Control Manual



Page Number / Item	Date of Revision	Signature of Approval
Annual Review	March 13, 2002	Dr. T. Mazzulli
Annual Review	October 25, 2003	Dr. T. Mazzulli
Annual Review	May 26, 2004	Dr. T. Mazzulli
Annual Review	May 12, 2005	Dr. T. Mazzulli
Annual Review	July 23, 2006	Dr. T. Mazzulli
Link to IC\Infection Control Pulsed-field Gel Electrophoresis.doc added	January 30, 2007	Dr. T. Mazzulli
Link to IC\VRE PCR Procedure.doc added	January 30, 2007	Dr. T. Mazzulli
Enter the no. of pink colonies grown on MRSA-Select if <5	January 30, 2007	Dr. T. Mazzulli
Added quantitation for MRSA	February 28, 2007	Dr. T. Mazzulli
Change to Denim Blue plates for MRSA Screen	March 13, 2007	Dr. T. Mazzulli
Change negative resulting phrases for MRSA, VRE and ESBL screen	March 13, 2007	Dr. T. Mazzulli
Included P. mirabilis for ESBL screen	March 13, 2007	Dr. T. Mazzulli
Annual Review	March 13, 2007	Dr. T. Mazzulli
Revised VRE Identification Procedure	March 22, 2008	Dr. T. Mazzulli
VRE – VANCS resistant <i>E. faecium</i> or <i>E. faecalis</i> report to MSH ICP if it is MSH patient; change to report as Presumptive VRE to all ICP	September 20, 2008	Dr. T. Mazzulli
Pseudo screen, patient swabs – change incubation period from 48 hours to 24 hours	September 20, 2008	Dr. T. Mazzulli
Annual Review	September 20, 2008	Dr. T. Mazzulli
Annual Review	September 20, 2009	Dr. T. Mazzulli
Annual Review	September 20, 2010	Dr. T. Mazzulli
ESBL screen updated to include KPC and NDM screen	November 10, 2010	Dr. T. Mazzulli
Removed send by taxi for carbapenemase PCR send out for Monday, Wednesday and Thursday	January 20, 2011	Dr. T. Mazzulli
Modified carbapenemase screening procedure to match Susceptibility manual	April 04, 2011	Dr. T. Mazzulli
Change VRE screening to Brilliance VRE Agar	April 04, 2011	Dr. T. Mazzulli

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Removed VRE Table 3; added link to Susceptibility manual	May 11, 2011	Dr. T. Mazzulli
VRE Screen, added VANCS back to heavy growth from BVRE or SBVRE	May 31, 2011	Dr. T. Mazzulli
VRE Screen – modified, finalized all day 2 reading in the morning of day 2	October 17, 2011	Dr. T. Mazzulli
Annual Review	October 17, 2011	Dr. T. Mazzulli
Modified Serratia screen	November 25, 2011	Dr. T. Mazzulli
Modified VRE resulting phrases	December 13, 2011	Dr. T. Mazzulli
Added CRE only screen	December 13, 2011	Dr. T. Mazzulli
Modified VRE reporting for vanA gene positive but phenotype vancomycin=S strains	February 1, 2012	Dr. T. Mazzulli
Added link to VRE PCR by Cepheid	July 16, 2012	Dr. T. Mazzulli
Modified planting volume into BHI broth for VRE/MRSA	August 28, 2012	Dr. T. Mazzulli
Annual Review	August 28, 2012	Dr. T. Mazzulli
Annual Review	May 31, 2013	Dr. T. Mazzulli
Manual updates in each section (Maldi procedure review)	October 10, 2013	Dr. T. Mazzulli
Changed CRE screen from ERTA to MERO discs	October 10, 2013	Dr. T. Mazzulli
Update VRE Identification	April 19, 2014	Dr. T. Mazzulli
Annual Review	April 19, 2014	Dr. T. Mazzulli
CRE reporting changes (Mero screen I/R)	June 27, 2014	Dr. T. Mazzulli
VITEK SXT=R SUPPRESS SXT confirm result by KB BEFORE reporting.	June 27, 2014	Dr. T. Mazzulli
vanB gene detected by Cepheid Xpert vanA/vanB Assay reporting	August 6, 2014	Dr. T. Mazzulli
Insert proper headers/footers, UHN/MSH Logo Fix broken link to appendix 2	August 12, 2014	Dr. T. Mazzulli
Added Group A Strep Screen and Klebsiella Screen	September 30, 2014	Dr. T. Mazzulli
Reviewed and updated procedure steps in all sections	September 30, 2014	Dr. T. Mazzulli
Added MRSA scant growth repeat broth culture comments	December 10, 2014	Dr. T. Mazzulli
Annual Review MRSA Removed MACRO use p.6 add “Scant growth (1-5 colonies) Upon Infection Control request to replant into BHIB (2mL): ”	May 19, 2015	Dr. T. Mazzulli

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p.19 changed serratia media to new ctcz media' p.22 ESBL testing is only performed on specimens from pregnant patients, specimens originating from mothers and baby units or upon special request. p.35 BA (for throat) added to GAS screen procedure culture media Changed Mac with cefpodoxime to ESBL isolation agar with cefpodoxime p.39 added ESBL comments to reporting comments p.24: added suscep. comment: \MRSS		
Specimen collected and transport for each section transferred to Specimen collection manual QPCMI02001 And replaced with link to specimen collection manual	May 26, 2015	Dr. T. Mazzulli
VRE outbreak: Temporary Procedure change in effect :VRE PCR on any amount of purple colonies	June 11, 2015	Dr. T. Mazzulli
VRE outbreak: Temporary Procedure change ended. Section removed.	July 15, 2015	Dr. T. Mazzulli
Prev + ESBL and Prev + CRE new statements	August 20, 2015	Dr. T. Mazzulli
p.23 Previously ESBL reporting phrased changed from "Susceptibility not done, please refer to sample collected on __Date_" to "Phenotypic screening suggests this organism is ESBL POSITIVE as previously confirmed on "yyyy.mm.dd"." LIS isolate comment code: \ESBP	December 2, 2015	Dr. T. Mazzulli
Updated ESBL+CRE and CRE sections with new reporting phrases Updated CRE section with new BCARB/CARB-R/ROSCO procedure	December 21, 2015	Dr. T. Mazzulli
MRSA reporting section: added link for susceptibility comments to MRSA reporting phrases in susceptibility manual. Resistant GNB reporting section & Pseudomonas screen section added for Positive reports: Add comment: "Susceptibility testing results are provided for infection control purposes only." \ICSN	April 4, 2016	Dr. T. Mazzulli
Added links in TOC to CNISP Surveillance Study for MRSA, VRE, and CRE and PHOL CRE Surveillance Study as well as in the CRE procedure section.	April 12, 2016	Dr. T. Mazzulli

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Annual Review	May 19, 2016	Dr. T. Mazzulli
VRE outbreak: Temporary Procedure change in effect :VRE PCR on any amount of purple colonies	June 13, 2016	Dr. T. Mazzulli
MRSA for new MRSA added step “Send to NML in batches when requested by IC for CNISP surveillance” VRE - added HEAVY growth workup	July 29, 2016	Dr. T. Mazzulli
VRE: Updated comment to include : “The effectiveness of vancomycin in this setting is uncertain and is not recommended. Please contact Infectious Diseases or Medical Microbiology for treatment advice.”	December 1, 2016	Dr. T. Mazzulli
Annual Review	May 20, 2017	Dr. T. Mazzulli
Updated Direct VRE PCR results with instructions to phone/email as per Isolate Notification and freezing table.	February 2, 2018	Dr. T. Mazzulli
Annual Review Temporary Procedure change in effect:VRE PCR on any amount of purple colonies. PFGE for all new VRE from MSH NOT UHN. Instructions to release ID once PCR is done of <i>E. faecium</i> or <i>faecalis</i> for suspect colonies if Vitek MS fails, confirmation of ID to follow. WASPLAB screening/incubation time changes: <ul style="list-style-type: none"> • MRSA 12hr & 24hrs modified to one 24hr read on Wednesday May 18th, 2018 evening. • MRSA 24 hr read changed to 18 & 24 hr read to Monday May 21, 2018 evening. • VRE changed from 12 & 36 to 18, 30, 36 on Monday May 21, 2018 evening. 	May 22 nd , 2018	Dr. T. Mazzulli
Minor format change	September 14, 2018	Dr. T. Mazzulli

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